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Import risk analysis: *Persea americana* plants for planting

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Approved for general release

Version information

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New Zealand is a member of the World Trade Organisation and a signatory to the Agreement on the Application of Sanitary and Phytosanitary Measures (“The Agreement”). Under The Agreement, countries must base their measures on an International Standard or an assessment of the biological risks to plant, animal or human health.

This document provides a scientific analysis of the risks associated with the importation of *Persea americana* plants for planting. It assesses the introduction of pests and pathogens associated with *Persea americana* budwood imported from around the world, and assesses the potential impacts of those organisms should they enter and establish in New Zealand. The document has been internally and externally reviewed and is now released publicly for feedback. Any significant new science information received that may alter the level of assessed risk will be included in a review, and an updated version released.

Import Risk Analysis

Table of Contents

1. Background

This import risk analysis (IRA) has been developed in response to a request to develop a new Import Health Standard (IHS) for the importation of avocado (*Persea americana*) plants for planting.

Pathways and commodities covered in this IRA are provided in Table 1-1.

Table 1-1: List of pathways and commodities assessed in this IRA

Commodity type	Pathway description
Persea americana plants for planting	Sea or air cargo from countries that have commercial avocado orchards and publish their research in English or Spanish languages.

2. Objectives

Biosecurity New Zealand's objectives in developing an IRA are to:

1. Identify which pests and diseases present a level of risk to New Zealand on the commodity(ies) and pathway(s) included in the scope, and
2. Evaluate the effect of risk management measures on the level of that identified risk,

in order to ensure that the risk managers have the information they need to recommend appropriately effective risk management measures.

3. IRA Scope

The scope of the IRA considers the following key questions:

- What are the detailed commodity and pathway descriptions?
- Are there any additional risk management questions?

Detailed commodity description	
Purpose	The commodity description defines the form of the commodity that is covered by this IRA, e.g. the commodity species and form under consideration. The commodity description may also include commercial production and/or grading requirements or other commodity quality specifications, and any actions taken that may remove or treat pests or diseases.
Commodity Description	• Budwood/ leafless cuttings[1] of *Persea americana* and all its varieties and cultivars, brought into level 3A post-entry quarantine (3A PEQ) for a period of active growth with a minimum of three distinct phenological growing events (referred to as 'shoot flush'). • Budwood/ leafless cuttings are cuttings taken from mother plants. The cuttings do not have leaves, roots or soil attached to them, only buds. • Budwood/ leafless cuttings are dipped in 1% w/v sodium hypochlorite for 2 minutes upon entry into the quarantine facility. • The PEQ period involves visual inspection of plants for the presence of pests and symptoms. Visual inspections by an MPI inspector are usually carried out during active growing seasons - for other plants for planting there are two inspections in the spring, two in the summer and one during the slower growth periods. A similar regime will be designed for avocados. Inspections by a facility operator are carried out at least twice a week.

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[1] As *Persea americana* is an evergreen species, the frequently used commodity type in the IHS of 'budwood/dormant cuttings' will be changed to 'budwood/leafless cuttings' to acknowledge the non-dormancy of the species.

Detailed commodity description	
	All imported plants for planting must comply with basic conditions as set out in Ministry for Primary Industries (MPI) import health standard 155.02.06 “Importation of Nursery Stock” (IHS 155.02.06). In addition to a period of PEQ, these include: • Clean (including free from soil), inert/ synthetic materials for protection, packaging, and shipping. • Phytosanitary certification stating that the exporting country has inspected the commodity and found it free of visually detectable regulated pests. • Pesticide treatments for insects and mites. • Please note: measures for specific pests listed under the “Basic Conditions” section of IHS 155.02.06 are not counted as basic measures but are specified measures.

Detailed pathway description	
Purpose	The pathway description defines the countries and methods the commodity will be sourced from and travel to New Zealand. The pathway description may also include commercial-specific pathway requirements.
Pathway Description	Sea or air cargo from countries that have commercial avocado orchards and publish their research in English or Spanish languages.

Risk management questions	
Purpose	Specific questions that the risk manager needs answered in order to make a decision. They are based on the scope of the overall project.
Description	PRAs will include information on the following risk management questions. What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ? • What symptoms will the pest or disease exhibit? • What are the known environmental conditions conducive to symptom expression? What are the limitations to taking samples for potential testing? • What plant part should be tested? • What is the optimum season for conducting detection testing?

4. IRA methodology

The IRA process involves three principal stages:

1. Hazard identification
2. Pest risk assessment
3. Evaluating risk management options

The IRA methodology used by Biosecurity New Zealand (BNZ) is described in more detail in the *BNZ Guidelines for Undertaking an Import Risk Analysis Version 1.0*.

4.1 Hazard identification

A hazard is a pest or disease that:

- is not present in New Zealand (or is present but still represents a biosecurity risk, e.g. is capable of vectoring pathogens not present in New Zealand);
- is associated with imported risk goods and import pathways; and
- has the ability to establish and cause harm in New Zealand.

Hazard identification (hazard ID) is the process where a list of pests and diseases potentially associated with the commodity is compiled and then assessed against specified risk evaluation criteria, in order to determine which potential hazards require further assessment.

The results of the hazard ID provide the following:

- Pests and diseases identified as potentially meeting the criteria to be quarantine pests[2] and diseases, associated with the commodity, and potentially not being managed by compliance with the commodity description and therefore requiring further assessment are listed in the Annex (Chapter 9).
- Groups (e.g. genera or families) and high-profile pests and diseases that did not meet the criteria to require pest risk assessment are listed in Appendix 1, along with the rationale for excluding them.
- Given the scope of the IRA (Chapter 3), only pathogens (i.e. bacteria, fungi, nematodes, viroids and viruses), or diseases were considered.

4.2 Pest risk assessment

The purpose of risk assessment is to determine the level of risk that hazard pests and diseases (identified at the hazard ID stage as requiring further assessment) pose to New Zealand. Specifically, the pest risk assessment (PRA) concludes whether the pest or disease meets the risk evaluation criteria for requiring additional measures over and above the commodity description.

For this import risk analysis, pests and diseases that could potentially be associated with the commodity on entry into New Zealand and meet the criteria to be considered for additional measures are those that:

- cannot be contained in a Level 3A PEQ facility and/or
- cannot be detected during a period of active growth with a minimum of three distinct phenological growing events (‘spring-flush’) in Level 3A PEQ.

The PRA addresses the following:

- Likelihood of entry:
    - The likelihood of entry into New Zealand given the commodity and pathway descriptions
- Likelihood of exposure and establishment:
    - The ability of the pest or disease to move from the commdity into a suitable environment to allow establishment (exposure)
    - The suitability of the New Zealand environment for the pest or disease (including climate, host plants and vectors)
- The potential impacts in New Zealand:
    - Economic impacts: on economically important plants (symptoms on individual plants, crop yield, costs of management practices, trade restrictions, rate of spread, etc.)
    - Environmental impacts: on native plants/taonga and ecologies/hauropi
    - Human health, sociocultural impacts: on people/tangata whenua

4.3 Management considerations

For those pests or diseases for which a PRA concluded that the criteria to be considered for additional measures have been met on at least one pathway, information relevant to management options, where available in the literature, have been evaluated. This information pertains to such options as:

- Post entry quarantine Level 3B
- Specified environmental conditions
- Diagnostic tests

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[2] In the context of this risk assessment a quarantine pest is an organism that is not present in New Zealand (or is present but still represents a biosecurity risk, e.g. is capable of vectoring pathogens not present in New Zealand) and is capable of establishing and causing harm in New Zealand.

5. Summary of IRA conclusions

A summary of the pest and diseases that meet the criteria to be considered for additional measures are listed in Table 5-1.

Table 5-1: Pests and diseases that meet the criteria to be considered for additional measures.

Pest/disease group	Species meeting the criteria to be considered for additional measures	PRA conclusions	
		Level of Risk	Level of uncertainty
Bacteria	Phytoplasmas (i.e. *Candidatus* Phytoplasma solani)that have a vector in New Zealand	Moderate	High
	Xanthomonas campestris and *X. axonopodis*	Moderate	High
	Xylella fastidiosa	High	Moderate
Fungi	*Colletotrichum* spp.	Moderate	Moderate
	Diaporthe perseae	Low	Moderate
	Diaporthe sterilis	Very low	Moderate
	Dothiorella aromatica	Low	Low
	Gliocladiopsis spp.	Very low	Moderate
	Lasiodiplodia pseudotheobromae	Moderate – High	Moderate
	Neocosmospora perseae	Low	Moderate
	Neofusicoccum nonquaesitum	Moderate	Moderate
	Pestalotiopsis longiseta	Low	Moderate
	Pleiocarpon algeriense and *Cylindrocladiella peruviana*	Moderate	Moderate
	Podosphaera perseae-americanae	Very low	Low
	Pseudoidium perseae-americanae	Very low	Low
Oomycetes	*Phytophthora mengei*	Moderate	Moderate
	Phytophthora palmivora	Moderate	Moderate
Viroids	*Avocado sunblotch viroid*	Moderate	Low
	Potato spindle tuber viroid	Moderate	Low

6. Summary of Pest Risk Assessments

The following sections provide a summary of the pest risk assessments described in detail within the Annex to this IRA.

6.1 Bacteria

6.1.1 Phytoplasmas on avocado

Phytoplasmas are bacteria that lack cell walls. They colonise the phloem of their plant hosts as well as various parts of their insect vectors. Phytoplasma infection is associated with a wide range of symptoms affecting all plant parts and can be fatal to the plant. Phytoplasmas belonging to the stolbur group (16SrXII), likely '*Candidatus* Phytoplasma solani', (16SrXII-A) have been identified in symptomatic avocado trees in Spain, and an unidentified phytoplasma has been identified in symptomatic avocado trees in Chile.

Summary of PRA conclusions

Given the arguments and evidence presented:

- The avocado-infecting phytoplasmas meet the criteria to be hazards on imported *Persea americana* budwood imported to New Zealand because they:
  - Are not known to be present in New Zealand, and
  - have the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description), and
  - have the potential to establish in New Zealand and cause harm to New Zealand.
- The avocado-infecting phytoplasmas pose an overall moderate risk on *Persea americana* budwood imported to New Zealand if there is a competent[3] vector in New Zealand, and negligible risk if there is no competent vector because they:
  - have a very low likelihood of entering New Zealand on *Persea americana* budwood (with moderate uncertainty) because:
    - *Persea americana* is a known host of "stolbur" (likely '*Ca.* P. solani'), and the phytoplasma found in Chile;
    - It is unlikely that avocado budwood will be taken from phytoplasma infected trees, since infections appear to be uncommon;
    - There is only a low volume of budwood entering Post Entry Quarantine (PEQ), which lowers the likelihood that infected budwood would enter PEQ even further;
    - Phytoplasmas would occur in association with budwood, as they are systemic through the plant phloem sieve tube elements and surface sterilisation of budwood would not affect them;
    - Plants will be held in PEQ for long enough to go through 3 flush growth periods (at least 12 months), so any avocado plants with phytoplasma infection are likely to show symptoms, and if so, diagnostic tests will detect phytoplasma.
    - However, some phytoplasma infections are asymptomatic and these would not be detected in PEQ which would result in infected plants being released into New Zealand.
    - 3A and 3B PEQ do not automatically include pre-determined phytoplasma PCR testing.
  - have a high likelihood of transferring from *Persea americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
    - Avocado plants derived from budwood will be planted and nurtured to survive in New Zealand and used to propagate new plants for planting.
    - If the plants are infected with avocado-infecting phytoplasmas at the time of their release from PEQ they will have the infection when they are multiplied and planted.
  - have a moderate likelihood (with high uncertainty) of establishing in New Zealand if there is a competent vector here, and a low likelihood of establishing in New Zealand (with high uncertainty) with no competent vector because:
    - Asymptomatic infected avocado plants released from PEQ will be used for grafting. Avocado trees originating from these are likely to be infected;
    - However if phytoplasma symptoms appear in avocado trees in New Zealand the avocado industry and nurseries will likely start testing for phytoplasma and only use clean stock;
    - It is uncertain whether there will be a competent vector in New Zealand. Known 'Ca. P. solani' vectors are not known to be present, but insects in the same family are present.
    - Two insects with the potential to be vectors have been reported on avocado in New Zealand;
    - Climate is not likely to be a barrier to the establishment of these avocado-infecting phytoplasmas in New Zealand;
  - may cause moderate overall impact on New Zealand if there is a competent vector present here (with high uncertainty), and very low overall impact (with low uncertainty) on New Zealand if there is no competent vector here because:

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[3] Vector competence is defined as "the ability of a vector to transmit a disease". It normally comprises the capacity of a vector to be infected, maintain and transmit an infectious agent.

  - If there is no competent vector in New Zealand, the plants infected would likely be limited to avocado trees grafted from infected plants from PEQ and their offspring, and possibly industry would stop grafting from infected plants. Phytoplasmas are not transmitted mechanically.
  - If there is a competent vector in New Zealand, other crop plants may be affected but it is difficult to know which without knowing the phytoplasma or vector. Some phytoplasmas such as '*Ca.* P. solani' can have major impacts on many crop plants including reduced crop yield and plant decline and death.
  - Other phytoplasma species have caused harm to native plants in New Zealand but it is not known if avocado-infecting phytoplasmas will. Environmental impacts are likely to be low (with high uncertainty) with a competent vector and negligible (with low uncertainty) without a competent vector.
  - Human health impacts are likely to be negligible (with low uncertainty) as this is a plant pathogen limited to plants and insects.
  - There is a very low likelihood that avocado-infecting phytoplasmas will affect amenity trees, home gardens, taonga species to tangata whenua, so sociocultural impacts are likely to be low (with high uncertainty) with a competent vector present, and negligible (with low uncertainty) without a competent vector.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Very low	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	Moderate (vector)	High
	Low (no vector)	High
Impact on the New Zealand economy, environment, health and society	Low (vector)	High
	Very low (no vector)	Low
Overall level of assessed risk to New Zealand	Moderate (vector)	High
	Negligible (no vector)	Low
Likelihood of there being a competent vector in New Zealand	Moderate	High

6.1.2 *Xanthomonas campestris* and *X. axonopodis* avocado pathovars

Xanthomonas species are gram-negative bacteria that infect a wide range of crops and wild plants. Two pathovars (strains that cause the same disease on the same host range) of *Xanthomonas* associated with disease in avocado (*Persea americana*) have been reported in the literature: *X. campestris* causing bacterial canker in California; and *X. axonopodis* causing bacterial leaf spot in the Seychelles.

Because there is very little information associated with *Xanthomonas* on avocado, information for the genus will be used where information for the pathovars is lacking.

Summary of PRA conclusions

Given the arguments and evidence presented:

- The *Xanthomonas campestris* and *X. axonopodis* avocado pathovars meet the criteria to be hazards on *Persea americana* budwood imported to New Zealand because they:
  - are not known to be present in New Zealand,
  - have the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description) and,
  - have the potential to establish in New Zealand, and cause harm to New Zealand.
- The *X. campestris* and *X. axonopodis* avocado pathovars pose an overall moderate risk on *P. americana* budwood imported to New Zealand because they:
  - have a very low likelihood of entering New Zealand on *P. americana* budwood (with high uncertainty) because:
    - they could survive as epiphytes on avocado budwood in PEQ without causing symptoms for the entire PEQ period, but

        - in the case of internal infection, symptoms are more likely to appear within a matter of days or weeks.
    - have a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
        - imported avocado budwood is intended to generate plants for planting in New Zealand and avocado plants (which are a suitable host for these pathogens) derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments, and
        - if the commodity is infected with either of these pathogens at the time of their release from PEQ and they are used as propagative material then the pathogens will be transferred to a suitable host multiple times.
    - have a high likelihood of establishing in New Zealand (with moderate uncertainty) because:
        - suitable hosts (in the form of cultivated avocados or native and introduced members of the Lauraceae family) are available for the establishment of both pathovars,
        - suitable climate is available for the establishment of the of the *X. campestris* avocado pathovar,
        - the wetter conditions of New Zealand compared to southern California are likely to favour establishment of the *X. campestris* avocado pathovar, and
        - the suitability of the New Zealand environment for establishment of the *X. axonopodis* pathovar cannot be definitively ruled out.
    - may cause moderate overall impact on New Zealand (with high uncertainty) because:
        - avocados are the only hosts of the *X. campestris* and *X. axonopodis* avocado pathovars commercially produced in New Zealand, and
        - infection of avocado trees with the *X. campestris* or *X. axonopodis* avocado pathovar could cause yield losses, and control of an outbreak could involve wide scale destruction of avocado trees (e.g., removal of all trees in one major avocado production area).

Infection of avocado trees with the *X. campestris* or *X. axonopodis* avocado pathovar could cause yield losses, and control of an outbreak could involve wide scale destruction of avocado trees (e.g., removal of all trees in one major avocado production area). The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Very low	High
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Moderate
Impact on the New Zealand economy, environment, health and society	Moderate	High
Overall level of assessed risk to New Zealand	Moderate	High

6.1.3 *Xylella fastidiosa* (bacterial leaf scorch)

Xylella fastidiosa is a fastidious xylem-limited bacterium (i.e. it has specialised nutrient needs and can be difficult to culture). It can colonise a very broad range of plants, including economically important crops and environmentally significant plants. It is a very serious pathogen of some of these plants, blocking water transport and causing wilting and death. Natural transmission is via specialist xylem-feeding insects.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Xylella fastidiosa* meets the criteria to be a hazard on avocado budwood imported to New Zealand because it:
    - is not known to be present in New Zealand. New Zealand has country freedom status for the species.
    - has the potential to enter New Zealand on avocado budwood (as described in the commodity description), establish in New Zealand, and cause harm to New Zealand.

- *Xylella fastidiosa* poses an overall high risk (with moderate uncertainty) on avocado budwood imported into New Zealand because it:
    - has a very low likelihood of entering New Zealand on avocado budwood (with high uncertainty) because;
        - infection of avocado by *X. fastidiosa* appears to be very rare (it has only ever been documented in the published literature once, with another unpublished report); however
        - infections of avocado and many other plant species are reported to be asymptomatic, and where infections are symptomatic the latent period is not known but may be long. Therefore established *X. fastidiosa* infections may not be detected before release from PEQ; and
        - for the above reasons, *X. fastidiosa* infections of avocado may also be more common than is reported;
    - has a high likelihood of transferring from avocado budwood to a suitable host in New Zealand (with low uncertainty) because:
        - avocado is a suitable host for *X. fastidiosa*.
        - avocado plants derived from grafting imported budwood onto suitable rootstock will be planted in New Zealand.
    - has a moderate likelihood of establishing in New Zealand (with moderate uncertainty) because:
        - plants suitable for colonisation by *X. fastidiosa* are widely grown throughout New Zealand;
        - New Zealand's temperate climate may be limiting, but at least some parts of the country are likely to be suitable for the establishment and expression of diseases caused by *X. fastidiosa*;
        - at least one competent vector species is present in New Zealand, it is widely distributed and has a broad host range which includes avocado as well as other economically important and environmentally significant plants.
    - may cause very high overall impacts on New Zealand (with high uncertainty) because:
        - *Xylella fastidiosa* can cause serious damage to crops and plant-based industries that are of high economic importance to New Zealand, for example, grapes, summerfruit and citrus;
        - the introduction of *X. fastidiosa* may result in market access restrictions for nursery stock, ornamentals, and cut flowers, particularly to Australia (New Zealand's main trading partner);
        - the cost of a response to an incursion of *X. fastidiosa* and/or long-term management costs resulting from establishment of the bacterium are likely to be very high;
        - the bacterium is known to be able to colonise environmentally and culturally significant native plants, although it is not known how likely this is to have an impact at the population or ecosystem level;
        - very significant socio-cultural impacts (possibly similar to those that resulted from New Zealand's *Pseudomonas syringae* pv. *actinidiae* (Psa) outbreak) could be expected if some subspecies establish, e.g., *X. fastidiosa* subsp. *fastidiosa*, the cause of Pierce's disease of grapevines;
        - however impacts may be limited by New Zealand's temperate climate (depending on the bacterial genotype introduced), and the impact on human health is considered to be negligible.

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	Very low	High
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	Moderate	Moderate
Impacts on the New Zealand economy, environment, health and society	Very high	High
Overall level of assessed risk to New Zealand	High	Moderate

6.2 Fungi

6.2.1 *Akaropeltopsis* sp. (sooty blotch of avocado)

Akaropeltopsis sp. is a fungus that causes sooty blotch on the branches, stems, leaf veins and fruit of *Persea americana* (avocado).

Summary of PRA conclusions

Akaropeltopsis sp. meets the criteria to be a hazard on imported *P. americana* budwood imported to New Zealand because:

  - it is not known to be present in New Zealand,
  - has the potential to establish in New Zealand, and cause harm to New Zealand, and
  - has the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description).
- *Akaropeltopsis* sp. poses an overall negligible risk on *P. americana* budwood imported to New Zealand because it:
  - has a negligible likelihood of entering New Zealand on *P. americana* budwood (with low uncertainty) because:
    - it can be present on the surface of budwood, but such infections are likely to be managed by dipping in 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility,
    - if *Akaropeltopsis* sp. is imported into PEQ then it is likely to produce visible symptoms in PEQ.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall level of assessed risk to New Zealand	Negligible	Low

6.2.2 Ambrosia fungi of the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp. vectored by beetles of the *Euwallacea fornicatus* species complex

Ambrosia fungi of the Ambrosia *Fusarium* Clade (AFC), *Graphium* and *Paracremonium* are symbionts associated with small (1–2 mm) ambrosia beetles in the genus *Euwallacea*. These fungi supply essential diet components for the ambrosia beetles and in return rely on the beetles to carry them to a new host. By cultivating their fungal symbionts in the xylem of plants, some *Euwallacea* species have managed to expand their host range and become invasive in new areas of the world. In recent years, for example, the *Euwallacea fornicatus* complex, together with their symbiotic fungi, have become a concern to the avocado industry in California, Florida, Australia, Israel and South Africa.

Summary of PRA conclusions

Given the arguments and evidence presented:

- The ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) meet the criteria to be a hazard on *Persea americana* budwood imported to New Zealand because they:
  - are not known to be present in New Zealand,
  - have the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description), and
  - have the potential to establish in New Zealand, and cause harm to New Zealand.
- The ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) pose an overall negligible risk on *Persea americana* budwood imported to New Zealand because they:
  - have a very low likelihood of entering New Zealand on *Persea americana* budwood (with low uncertainty) because:

        - they are associated with and spread by ambrosia beetles or their beetle galleries. Level 3A PEQ has measures in place to manage the risks associated with insects, wastewater, waste and accidental transfer, therefore, ambrosia beetles carrying ambrosia fungi are highly unlikely to escape level 3A PEQ to infect a suitable host;
        - their infection of avocado budwood is likely visible as necrotic lesions and sugar volcanos. The ambrosia fungi of the *Euwallacea fornicatus* species complex are reported to have localised pathogenicity (<10cm from beetle gallery). Ambrosia beetle entry points and galleries will be visible during inspection and, thus, infected budwood would be detected and destroyed before being released from PEQ.
    - have a very low likelihood of transferring from the imported commodity onto a suitable host in New Zealand (with moderate uncertainty) because:
        - there are no *Euwallacea* species reported to be present in New Zealand that could serve as vectors of ambrosia fungi (AFC); and there are no alternative vectors reported to be present in New Zealand, such as *Quadrastichus erythrinae* or *Xyleborus ferrugineus*;
        - some uncertainty arises from the fact that most fungi from the AFC have only recently been described, and species within it and their symbiotic associations with their ambrosia beetles are still being described.

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	Very low	Low
The likelihood of transferring from the imported commodity onto a suitable host	Very low	Moderate
Overall level of assessed risk to New Zealand	Negligible	Low

6.2.3 *Colletotrichum* spp.

This PRA assesses species associated with avocado found in two species complexes: the *Colletotrichum gloeosporioides* species complex and the *Colletotrichum acutatum* species complex. The following species will be assessed: *Colletotrichum aenigma*, *C. chrysophilum*, *C. endophyticum*, *C. jiangxiense*, *C. queenslandicum* and *C. tropicale* are in the *C. gloeosporioides* species complex and *C. nymphaceae* is in the *C. acutatum* species complex.

Colletotrichum species in both the *gloeosporioides* and *acutatum* species complexes are fungi with a wide host and environment range. They are important plant pathogens but can have endophytic and saprotrophic lifestyles. In avocado, *Colletotrichum* species cause anthracnose in fruit and sometimes stem dieback.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Colletotrichum* spp. (*C. aenigma, C. chrysophilum, C. endophyticum, C. jiangxiense, C. nymphaeae, C. queenslandicum, C. tropicale*) meet the criteria to be hazards on imported *Persea americana* budwood imported to New Zealand because they:
    - are not known to be present in New Zealand,
    - have the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description), and
    - have the potential to establish in New Zealand, and cause harm to New Zealand.
- *Colletotrichum* poses an overall moderate risk on *P. americana* budwood imported to New Zealand because it:
    - has a low likelihood of entering New Zealand on *P. americana* budwood (with moderate uncertainty) because:
        - *Colletotrichum* infection under high temperatures (25 – 30℃) and elevated relative humidity can produce symptoms in a few days, but
        - there is uncertainty around the endophytic capabilities of some *Colletotrichum* species in *P. americana* budwood which may allow asymptomatic *Colletotrichum* infection during the PEQ period.

    - has a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
        - imported avocado budwood is intended to generate plants for planting in New Zealand and avocado plants (which are a suitable host for these pathogens) derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments, and
        - if the commodity is infected with these pathogens at the time of their release from PEQ and they are used as propagative material then the pathogens could be transferred to a suitable host on multiple occasions.
    - has a high likelihood of establishing in New Zealand (with low uncertainty) because:
        - hosts (in the form of cultivated avocados and other cultivated crops) are available for the establishment of *Colletotrichum* species
        - the climate in parts of New Zealand is suitable for the establishment of *Colletotrichum*
    - may cause moderate overall impact on New Zealand (with moderate uncertainty) because:
        - the economic impact of *Colletotrichum* to New Zealand is considered to be moderate, with low uncertainty
        - the impact on the environment from the establishment of *Colletotrichum* in New Zealand is considered to be very low, with moderate uncertainty
        - the human health impact of *Colletotrichum* to New Zealand is considered to be negligible, with low uncertainty
        - the sociocultural impact of *Colletotrichum* to New Zealand is considered to be low, with low uncertainty

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Low	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low
Impact on the New Zealand economy, environment, human health and society	Moderate	Moderate
Overall level of assessed risk to New Zealand	Moderate	Moderate

6.2.4 *Clonostachys pseudochroleuca* (avocado branch dieback and wilting)

Clonostachys pseudochroleuca is a fungus that is associated with a wide range of plants and environments. It is commonly found as a saprotroph or endophyte but has been recorded as a pathogen in avocado, vectored by ambrosia beetles, causing avocado branch dieback and wilting.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Clonostachys pseudochroleuca* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because it:
    - is not known to be present in New Zealand,
    - has the potential to establish in New Zealand, and cause harm to New Zealand, and
    - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description).
- *Clonostachys pseudochroleuca* poses an overall negligible risk on *Persea americana* budwood imported to New Zealand because it:
    - has a negligible likelihood of entering New Zealand on *Persea americana* budwood (with low uncertainty) because:
        - *Clonostachys pseudochroleuca* is associated with and spread by ambrosia beetles. Budwood will be treated with miticides and insecticides. Furthermore, Level 3A PEQ has measures in place to manage the risks associated with insects, wastewater, waste and accidental transfer, therefore ambrosia beetles carrying *C. pseudochroleuca* are highly unlikely to escape level 3A PEQ to infect a suitable host.

        - *Clonostachys pseudochroleuca* infection in avocado budwood is only found in association with ambrosia beetle galleries. Ambrosia beetle entry points and galleries will be visible during inspection and thus infected budwood would be detected and destroyed before they are released from PEQ.

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall level of assessed risk to New Zealand	Negligible	Low

6.2.5 *Cophinforma tumefaciens*

Cophinforma tumefaciens is a pathogenic ascomycete fungus that causes stem galls/tumours in young and old avocado plants. It is known to cause galls/tumours in *Citrus* spp. as well as in other host plants.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Cophinforma tumefaciens* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because it:
    - is not known to be present in New Zealand;
    - has the potential to establish (and spread) in New Zealand, and cause harm to New Zealand, and
    - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description).
- *Cophinforma tumefaciens* poses an overall negligible risk on *Persea americana* budwood imported to New Zealand because it:
    - has a negligible likelihood of entering New Zealand on *Persea americana* budwood (with low uncertainty) because:
        - established *C. tumefaciens* infections are highly likely to be detected during pre-export phytosanitary inspection, on arrival in New Zealand, or while the budwood is prepared for grafting,
        - recently infected budwood from contaminated pruning tools would likely develop symptoms 2- 24 weeks after infection within the PEQ period. Searches found no evidence of asymptomatic infection or longer latent period in avocado and other host plants.
        - Level 3A PEQ has measures in place to manage the risks associated with wastewater, waste, and accidental transfer, therefore viable *C. tumefaciens* propagules are highly unlikely to escape level 3A PEQ to infect a suitable host.

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall level of assessed risk to New Zealand	Negligible	Low

6.2.6 *Diaporthe perseae*

Diaporthe perseae is a fungal plant pathogen that causes stem-end rot (postharvest) disease in avocado.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Diaporthe perseae* meets the criteria to be a hazard on imported *Persea americana* (avocado) budwood imported to New Zealand because it:
    - is not known to be present in New Zealand
    - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description).
    - has the potential to establish in New Zealand, and cause harm to New Zealand.
- *Diaporthe perseae* poses an overall **low** risk on *Persea americana* budwood imported to New Zealand because it:
    - has a high likelihood of entering New Zealand on *Persea americana* budwood (with low uncertainty) because:
        - It is likely that *D. perseae* will not be detected from visual inspection during export preparation and pre-export phytosanitary inspection to New Zealand and on arrival to New Zealand. In its endophytic phase, it can be present on leafless avocado without showing visible symptoms
        - The growing period in Post Entry Quarantine (PEQ) level 3A is not likely to impact the likelihood of entry of *D. perseae* because infected budwood that are grafted in PEQ level 3A might not show visible symptoms within the PEQ growing period
        - The main disease symptom caused by *D. perseae* (stem-end rot) will not be expressed during the growing period in PEQ because it is usually expressed on the fruits after harvest. Fruits are not likely to form and mature to harvest state during the time of PEQ.
    - has a high likelihood of transferring from *Persea americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
        - Avocado (*P. americana*) is a suitable host of *D. perseae*
        - Avocado budwood derived from grafted imported budwood onto rootstock will be planted in New Zealand.
    - has a high likelihood of establishing in New Zealand (with low-moderate uncertainty) because:
        - *Diaporthe perseae* is found in places such as South Africa, Netherland and Australia, part of which have similar climate to parts of New Zealand
        - Avocado is a suitable host and is commercially produced and commonly grown in northern parts of New Zealand, and less frequently in other parts of the country
        - *Diaporthe perseae* can be spread by human movement of infected nursery stock for planting and movement of infected fruits (fruits producing spores).
    - may cause low overall impact on New Zealand (with moderate uncertainty) because:
        - *Diaporthe perseae* can result in some postharvest losses that can reduce crop value and marketability, but overall losses may not differ from current level of stem-end rot diseases in New Zealand
        - Current control methods of control of other stem-end rot causing pathogens in New Zealand may be effective in controlling diseases caused by *D. perseae*.
        - It can potentially result in trade barriers with countries where it has not been reported to be present. However, it is recorded as present in Australia which is New Zealand's major export market. As such, it might not have a significant impact on export.

Although there are high likelihoods of entry, exposure and establishment, *D. perseae* is a less important pathogen of stem-end rot disease than other stem-end rot causing fungi such as *Dothiorella aromatica* in Australia and *Colletotrichum gloeosporioides* and *Diaporthe* spp. already in New Zealand.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	High	Low
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low-moderate
Impact on the New Zealand economy, environment, health and society	Low	Low
Overall level of assessed risk to New Zealand	Low	Moderate

6.2.7 *Diaporthe sterilis*

Diaporthe sterilis is a fungus that has relatively recently been isolated from branch cankers on avocado orchard plants in Italy.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Diaporthe sterilis* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because it:
  - is not known to be present in New Zealand
  - has the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description), and
  - has the potential to establish in New Zealand, and cause harm to New Zealand.
- *Diaporthe sterilis* poses an overall very low risk on *P. americana* budwood imported to New Zealand because it:
  - has a high likelihood of entering New Zealand on *P. americana* budwood (with moderate uncertainty) because:
    - *Diaporthe sterilis* has been isolated from aboveground plant parts of avocado plants (stems, branches and fruits) and is likely to be present in infected leafless avocado budwood.
    - if *D. sterilis* is imported on leafless avocado budwood, it is likely to express symptoms in the duration of the time in PEQ and the biosecurity risk are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events in 3A PEQ,
    - however, it can potentially be present in asymptomatic leafless, rootless avocado budwood that might not be the detected during preparation for export, or in phytosanitary inspection pre-export or on arrival to New Zealand (although with high uncertainty).
  - has a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with moderate uncertainty) because:
    - avocado (*P. americana*) is a suitable host of *D. sterilis*.
    - avocado budwood derived from grafted imported budwood onto rootstock will be planted in New Zealand.
  - has a moderate likelihood of establishing in New Zealand (with moderate uncertainty) because:
    - *Diaporthe sterilis* is found in Italy which has a similar climate to most parts of New Zealand.
    - avocado is a suitable host and is commercially produced and commonly grown in northern parts of New Zealand, and less frequently in other parts of the country.
    - It is known to also infect *Vaccinium corymbosum* (blueberry), which is widely grown commercially in parts of the North and South Island of New Zealand.
    - *Diaporthe sterilis* can possibly be spread by human movement of infected nursery stock for planting and movement of infected fruits (fruits producing spores).
  - may cause very low overall impact on New Zealand (with moderate uncertainty) because:
    - *Diaporthe sterilis* can cause trunk cankers and potentially result in some postharvest losses of avocado. However, overall losses caused may not differ from the current losses caused by other canker and stem-end rot pathogenic fungi already present in New Zealand.
    - it has been reported to cause disease symptoms on *V. corymbosum* (blueberry) (cankers at their bases and brown lesions on stems resulting in twig blight). While such diseases can affect blueberry production, the impact of *D. sterilis* on blueberry production has not yet been quantified.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	High	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	High	Moderate
The likelihood of establishing in the New Zealand environment	Moderate	Moderate
Impact on the New Zealand economy, environment, human health and society	Very low	Moderate
Overall level of assessed risk to New Zealand	Very low	Moderate

6.2.8 *Dothiorella aromatica*

Dothiorella aromatica is a fungal plant pathogen that causes stem-end rot (postharvest) disease in avocado. It has also been associated with stem-end rot disease in mangos.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Dothiorella aromatica* meets the criteria to be a hazard on imported *Persea americana* (avocado) budwood imported to New Zealand because it:
  - is not known to be present in New Zealand
  - has the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description), and
  - has the potential to establish in New Zealand, and cause harm to New Zealand.
- *Dothiorella aromatica* poses an overall low risk on *P. americana* budwood imported to New Zealand because it:
  - has a high likelihood of entering New Zealand on *P. americana* budwood (with low uncertainty) because:
    - it is likely that *D. aromatica* will not be detected by visual inspection during export preparation and pre-export phytosanitary inspection to New Zealand and on arrival to New Zealand. In its endophytic phase, it can be present on leafless avocado budwood without showing visible symptoms.
    - the main disease symptom caused by *D. aromatica* (stem-end rot) will not be expressed during the growing period in PEQ because it is usually expressed on the fruits after harvest. Fruits are not likely to form and mature to harvest state during the time of PEQ.
  - has a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
    - avocado (*P. americana*) is a suitable host of *D. aromatica.*
    - avocado budwood derived from grafted imported budwood onto rootstock will be planted in New Zealand.
  - has a high likelihood of establishing in New Zealand (with moderate uncertainty) because:
    - *Dothiorella aromatica* is found in South Africa and Australia, parts of which have similar climate to parts of New Zealand,
    - avocado is a suitable host and is commercially produced and commonly grown in northern parts of New Zealand, and less frequently in other parts of the country,
    - *Dothiorella aromatica* can be spread by human movement of infected nursery stock for planting and movement of infected fruits (fruits producing spores).
  - may cause low overall impact on New Zealand (with moderate uncertainty) because:
    - *Dothiorella aromatica* can result in some postharvest losses that can reduce crop value and marketability, but overall losses may not differ from current level of stem-end rot diseases in New Zealand
    - current control methods of control of other stem-end rot causing pathogens in New Zealand may be effective in controlling diseases caused by *D. aromatica*.
    - it can potentially result in trade barriers with countries where it has not been reported to be present. However, it is recorded as present in Australia which is New Zealand's major export market. As such, it might not have a significant impact on export.

    - Although there are high likelihoods of entry, exposure and establishment, *D. aromatica* is a less important pathogen of stem-end rot disease than other stem-end rot causing fungi such as *Colletotrichum gloeosporioides* and *Diaporthe* spp. already in New Zealand.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	High	Low
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Moderate
Impact on the New Zealand economy, environment, health and society	Low	Low
Overall level of assessed risk to New Zealand	Low	Low

6.2.9 *Elsinoe perseae* – avocado scab

Elsinoe perseae (avocado scab) is a fungal pathogen that causes brown scabby lesions on fruit, leaves and stems of avocado plants. It can reduce avocado fruit yield, quality and marketability.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Elsinoe perseae* meets the criteria to be a hazard on *Persea americana* budwood imported to New Zealand because it:
  - is not known to be present in New Zealand. *Elsinoe perseae* was recorded in error in New Zealand in 1991 and older literature and some databases reflect this.
  - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description), establish in New Zealand, and cause harm to New Zealand.
- *Elsinoe perseae* poses an overall negligible risk on *Persea americana* budwood imported to New Zealand because it:
  - has a negligible likelihood of entering New Zealand on *Persea americana* budwood (with low uncertainty) because:
    - Avocado scab lesions are likely to be detected during pre-export phytosanitary inspection, on arrival in New Zealand, or while the budwood is being prepared for grafting.
    - Ungerminated spores remaining on the budwood after surface disinfection upon arrival in PEQ will either die or germinate once the buds break dormancy and produce visible symptoms very rapidly, and most probably within the PEQ period.
    - Symptoms develop in 7–10 days once spores germinate on susceptible leaves, stems or fruit. Searches found no evidence of asymptomatic *E. perseae* infections or longer latent periods in avocado plants.
    - *Elsinoe perseae* spores are spread by watersplash, windborne rain and insects (thrips). Given the measures in level 3A PEQ to manage risks associated with insects and wastewater, waste and accidental transfer, *E. perseae* propagules are highly unlikely to escape to infect a suitable host.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall level of assessed risk to New Zealand	Negligible	Low

6.2.10 *Gliocladiopsis forsbergii*, *Gliocladiopsis peggii* and *Gliocladiopsis whileyi*

Gliocladiopsis is a genus of soilborne fungi found mostly in tropical and sub-tropical regions of the world. Species of this genus are regarded as secondary plant pathogens or saprobes. *Gliocladiopsis forsbergii, G. peggii* and *G. whileyi* have been isolated frequently from necrotic roots of avocado trees, but pathogenicity studies have so far only been done for one strain of *G. peggii*. Genus level information is being used where necessary as limited information about the ecology of these fungi is available.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Gliocladiopsis forsbergii, G. peggii* and *G. whileyi* meet the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because they:
    - are not known to be present in New Zealand, and
    - have the potential to establish in New Zealand, and cause harm to New Zealand, and
    - have the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description).
- *Gliocladiopsis forsbergii, G. peggii* and *G. whileyi* pose an overall very low risk on *P. americana* budwood imported to New Zealand because they:
    - have a low likelihood of entering New Zealand on *P. americana* budwood (with high uncertainty) because:
        - they are likely soilborne, although other *Gliocladiopsis* species have been recovered as endophytes from stems and leaves, and
        - it is unknown if symptoms would develop in PEQ as these species are likely secondary pathogens that can also be saprobic or endophytic
    - have a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
        - imported avocado budwood is intended to generate plants for planting in New Zealand and avocado plants (which are a suitable host for these pathogens) derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments, and
        - if the commodity is infected with either of these pathogens at the time of their release from PEQ and they are used as propagative material, then the pathogens will be transferred to a suitable host
    - has a high likelihood of establishing in New Zealand (with low uncertainty) because:
        - a suitable host in the form of cultivated avocados is available for their establishment, and
        - a suitable climate is available for their establishment, and
        - other *Gliocladiopsis* species have established in New Zealand
    - may cause very low overall impact on New Zealand (with moderate uncertainty) because:
        - their economic impact likely depends on a variety of abiotic and biotic factors (such as co-infection with other pathogens), and
        - good orchard management practises are likely to manage these species

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Low	High
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low
Impact on the New Zealand economy, environment, human health and society	Very low	Moderate
Overall level of assessed risk to New Zealand	Very low	Moderate

6.2.11 *Grovesinia moricola* (zonate leaf spot)

Grovesinia moricola (zonate leaf spot) is a fungal pathogen that produces characteristic target-shaped necrotic rings leading to leaf blight and defoliation in a broad range of host species. The disease has been reported infecting *Persea americana* (avocado) in the USA and Brazil, causing round leaf lesions with dark margins. It has the potential to infect a number of economically significant hosts in New Zealand.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Grovesinia moricola* meets the criteria to be a hazard on *P. americana* budwood imported to New Zealand because it:
  - has the potential to enter New Zealand on *P. americana* budwood, establish and cause harm to New Zealand, and
  - is not known to be present in New Zealand.
- *Grovesinia moricola* poses an overall negligible risk on *P. americana* budwood imported to New Zealand because it:
  - has a negligible likelihood of entering New Zealand on *P. americana* budwood as described in the commodity description (with low uncertainty) as:
    - the fungus is primarily a foliar pathogen and is not strongly associated with plant stems,
    - disinfection dip in 2% sodium hypochlorite (as per the commodity description) is likely to be effective in eliminating any infection on the surface of avocado budwood, and
    - if *G. moricola* is imported into PEQ then it is likely to produce visible symptoms within the period of growth in PEQ.

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall Level of assessed risk to New Zealand	Negligible	Low

6.2.12 *Lasiodiplodia pseudotheobromae*

Lasiodiplodia pseudotheobromae is a fungal plant pathogen that has been reported to cause dieback, cankers and postharvest diseases (stem-end rot and fruit rot disease) in many woody plant species which includes avocado.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Lasiodiplodia pseudotheobromae* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because it:
  - is not known to be present in New Zealand,
  - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description), and
  - has the potential to establish in and cause harm to New Zealand.
- *Lasiodiplodia pseudotheobromae* poses an overall moderate risk on *P. americana* budwood imported to New Zealand because it:
  - has a low likelihood of entering New Zealand on *P. americana* budwood (with low-moderate uncertainty) because:
    - *Lasiodiplodia pseudotheobromae* has been isolated from the stems, branches and fruits of avocado plants and therefore can be associated with leafless avocado budwood.
    - Infected budwood is likely to exhibit symptoms such as necrotic lesions and discolouration of internal tissues that can be visually detectable during preparation for export or on phytosanitary inspection pre-export or on arrival in New Zealand.
    - However, *L. pseudotheobromae* can potentially be present on asymptomatic leafless, rootless avocado budwood that might not be the detected during preparation for export, or in phytosanitary inspection pre-export or on arrival to New Zealand although with high uncertainty.
    - If *L. pseudotheobromae* is imported on leafless avocado budwood, it is likely to express symptoms in the duration of the time in PEQ. The biosecurity risk is likely to be managed by the period of active growth with a minimum of three distinct phenological growing events in 3A PEQ.

  - has a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
    - avocado (*P. americana*) is a suitable host of *L. pseudotheobromae*
    - avocado budwood derived from grafted imported budwood onto rootstock will be planted in New Zealand.
  - has a high likelihood of establishing in New Zealand (with low uncertainty) because:
    - *L. pseudotheobromae* is found in about 23 countries, some of these countries have a similar climate to New Zealand
    - avocado is a suitable host and is commercially produced and commonly grown in northern parts of New Zealand, but less frequently in other parts of the country.
    - it is known to affect a wide range of other host plants that are widely grown commercially in orchards and in home gardens in parts of the North and South Island of New Zealand.
    - *L. pseudotheobromae* can spread by human movement of infected nursery stock for planting, movement of infected fruits (spores produced from the fungus present on infected fruit), contaminated soil, and pruning material.
  - may cause moderate overall impact on New Zealand (with moderate uncertainty) because:
    - *L. pseudotheobromae* can potentially cause unwanted impacts in New Zealand by reducing the crop value and marketability of avocados.
    - it can also cause diseases which result in reduced yields and affect the marketability of other horticultural crops such as *Citrus* spp., *Vitis vinifera* (grapes) and *Diospyros kaki* (persimmons). These are crops of economic value to New Zealand both domestically and internationally which together are worth over a billion dollars.
    - if it establishes in New Zealand, it can potentially result in trade barriers with countries where it has not been reported to be present.

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	Low	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	High	Moderate
The likelihood of establishing in the New Zealand environment	High	Moderate
Impact on the New Zealand economy, environment, health and society	Moderate-High	Moderate
Overall level of assessed risk to New Zealand	Moderate-High	Moderate

6.2.13 *Mycosphaerella perseae*

Mycosphaerella perseae is a fungal plant pathogen that causes silver spot disease in *Persea americana* (avocado). Silver spot disease causes lesions on the leaves of the plant.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Mycosphaerella perseae* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because it:
  - is not known to be present in New Zealand;
  - has the potential to establish in New Zealand, and cause harm to New Zealand, and
  - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description).
- *Mycosphaerella perseae* poses an overall negligible risk on *Persea americana* budwood imported to New Zealand because it:
  - has a negligible likelihood of entering New Zealand on *Persea americana* budwood (with low uncertainty) because:
    - established *M. perseae* infections are highly likely to be detected, and infected material removed or destroyed, during preparation for export, in

        - phytosanitary inspections pre-export or with surface disinfection on arrival at PEQ in New Zealand;
        - residual biosecurity risks from recent infections or ungerminated conidia on the surface of avocado budwood are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ;

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall level of assessed risk to New Zealand	Negligible	Low

6.2.14 *Neocosmospora perseae*

Neocosmospora perseae is a fungal pathogen reported to cause avocado trunk cankers and is probably an opportunistic pathogen infecting the plant via wounds.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Neocosmospora perseae* meets the criteria to be a hazard on *Persea americana* budwood imported to New Zealand because it:
    - is not known to be present in New Zealand,
    - has the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description), and
    - has the potential to establish in New Zealand, and cause harm to New Zealand.
- *Neocosmospora perseae* poses an overall low risk on *P. americana* budwood imported to New Zealand because it:
    - has a very low likelihood of entering New Zealand on *P. americana* budwood (with moderate uncertainty) because:
        - symptoms will usually develop and will be detected prior to export or within the PEQ period; but
        - estimates of time to symptom development are based on pathogenicity testing and natural infections may develop more slowly; and
        - internal necrosis caused by *N. perseae* may not be detected in routine inspections of plants in PEQ.
    - has a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
        - avocado (*P. americana*) is a suitable host for *N. perseae*;
        - avocado plants derived from grafting imported budwood onto suitable rootstock will be planted in New Zealand.
    - has a high likelihood of establishing in New Zealand (with low uncertainty) because:
        - avocado is a host and is commercially produced and commonly grown domestically in northern parts of New Zealand, though less frequently elsewhere in the country;
        - *Neocosmospora perseae* is reported from an area that has a similar climate to parts of New Zealand;
        - *Neocosmospora perseae* can almost certainly spread between host plants in New Zealand because conidia (asexual spores) of *Fusarium solani* species complex such as *N. perseae* are typically produced on plant debris on the soil surface and dispersed by soil water or water splash to infect roots or above ground tissues of suitable hosts.
    - may cause low overall impact to New Zealand (with moderate uncertainty) because:
        - *Neocosmospora perseae* outbreaks are likely to be rare and sporadic but can cause unwanted impacts on avocado production in New Zealand by damaging some trees in some orchards or growing regions;
        - however, there is moderate uncertainty that impacts of *N. perseae* will be limited to avocado, since *N. perseae* is recently described and some closely related species in the *Fusarium solani* species complex are reported as opportunistic pathogens, causing disease symptoms in a diverse range of

plant hosts as well as insects, marine animals, and humans. Therefore *N. perseae* has the potential for broader unwanted economic, environmental and human health impacts.

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	Very low	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low
Impact on the New Zealand economy, environment, health and society	Low	Moderate
Overall level of assessed risk to New Zealand	Low	Moderate

6.2.15 *Neofusicoccum nonquaesitum*

Neofusicoccum nonquaesitum is a pathogenic fungi from the family Botryoshaeriaceae. They can cause diseases such as stem/branch cankers, necrosis and dieback in several woody crop plant species (grapevine, avocado, and apples) and non-crop plant species.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Neofusicoccum nonquaesitum* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because they:
    - are not known to be present in New Zealand:
    - have the potential to establish in New Zealand, and cause harm to New Zealand, and
    - have the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description).
- *Neofusicoccum nonquaesitum* poses an overall moderate risk on *Persea americana* budwood imported to New Zealand because they:
    - have a low likelihood of entering New Zealand on *Persea americana* budwood (with moderate uncertainty) because:
        - both species have been isolated from the stems and branches of avocado plants and can be present on leadless avocado budwood.
        - avocado budwood with established *N. nonquaesitum* infections can exhibit symptoms such as cankers on plant tissues that could be detected during preparation for export or on arrival in New Zealand
        - *Neofusicoccum nonquaesitum* can potentially be present on asymptomatic leafless avocado budwood that might not be detected during preparation for export and upon arrival in New Zealand, with high uncertainty
        - If *N. nonquaesitum* is imported on leafless avocado budwood, the active growing period at level 3A PEQ (a minimum of three distinct phenological growing events) will likely result in expression of symptoms.
    - have a high likelihood of transferring from *Persea americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
        - avocado (*P. americana*) is a suitable host of *N. nonquaesitum.*
        - avocado plants derived from grafting imported budwood onto rootstock will be planted in New Zealand.
    - have a high likelihood of establishing in New Zealand (with low uncertainty) because:
        - *Neofusicoccum nonquaesitum* is reported to be present in five countries, parts of which have similar climate to New Zealand. The climatic conditions in some parts of New Zealand are likely to be suitable for them to establish and spread especially the North Island where climatic conditions are warmer
        - both species are associated with a range of crop and non-crop host plants such as avocado, blueberry, grapevine, apples, plum and oak tree species, which are commercially grown in both the North and South Island of New Zealand

        - *Neofusicoccum nonquaesitum* can be spread through the human movement of infected nursey stock and contaminated pruning tools.
    - may cause moderate overall impact on New Zealand (with moderate uncertainty) because:
        - *Neofusicoccum nonqaesitum* can potentially result in reduced productivity and yield loss of avocado. This can have an unwanted impact on both domestic and international marketability of avocado
        - infections by *N. nonquaesitum* can affect market access of export of avocado to other parts of the world where they are not reported to be present
        - *Neofusicoccum nonquaesitum* can cause disease and result in reduced yields of other economic important host plants such as grapevine, apples and blueberries resulting in additional economic loss.

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	Low	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low
Impact on the New Zealand economy, environment, human health and society	Moderate	Moderate
Overall level of assessed risk to New Zealand	Moderate	Moderate

6.2.16 *Pestalotiopsis longiseta*

Pestalotiopsis longiseta has been reported to cause dieback disease in *Persea americana* (avocado) in China.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Pestalotiopsis longiseta* meets the criteria to be a hazard on imported *P. americana* budwood imported to New Zealand because it:
    - is not known to be present in New Zealand
    - has the potential to establish in New Zealand, and cause harm to New Zealand, and
    - has the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description).
- *Pestalotiopsis longiseta* poses an overall low risk on *P. americana* budwood imported to New Zealand because it:
    - has a low likelihood of entering New Zealand on *P. americana* budwood (with moderate uncertainty) because:
        - *Pestalotiopsis longiseta* is reported to be associated with avocado as well as other crop plants.
        - In its pathogenic phase of infection, it is likely to exhibit symptoms if infected budwood is grafted in PEQ level 3A.
        - It can be present on avocado budwood (in a latent or endophytic phase) without exhibiting symptoms
    - has a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with moderate uncertainty) because:
        - It can spread by the human movement of infected nursery stock that is grafted on to rootstock that will be planted in orchards in New Zealand.
    - has a moderate likelihood of establishing in New Zealand (with moderate uncertainty) because:
        - Avocado which is a known host is grown commercially and in home gardens in most parts of New Zealand.
        - It is not likely to be restricted by climate because it occurs in both tropical and temperate countries some of which have similar climate to New Zealand.
        - It can spread by the human movement of infected nursery stock for planting.
        - It is not host specific but opportunistic and can infect a wide range of host plants. Other crop plants are available in New Zealand which it can infect.

    - may cause low overall impact on New Zealand (with moderate uncertainty) because:
        - it was only recently associated with avocado in China, with up to 30% of young plants dying. The impact it might have on mature avocado trees and crop yield has not been assessed and quantified.
        - about 16 species from the genus *Pestalotiopsis* are already recorded to be present in New Zealand and they have not been reported to affect crop plants significantly.
        - It caused between 10-20% yield loss of persimmons in Japan. Persimmons are grown in New Zealand with a value of about NZ$ 12 million in export and domestic market in 2020.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Low	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	High	Moderate
The likelihood of establishing in the New Zealand environment	Moderate	Moderate
Impact on the New Zealand economy, environment, human health and society	Low	Moderate
Overall level of assessed risk to New Zealand	Low	Moderate

6.2.17 *Phyllachora gratissima* (tar spot)

Phyllachora gratissima is a fungal leaf pathogen that causes tar spot on the leaves of *Persea americana* (avocado). It is considered a minor pathogen that is more frequent in ‘Creole’ avocados.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Phyllachora gratissima* meets the criteria to be a hazard for New Zealand because it:
    - is not known to be present in New Zealand, and
    - has the potential to enter New Zealand on *P. americana* budwood, establish in and cause harm to New Zealand.
- *Phyllachora gratissima* poses an overall negligible risk on *P. americana* budwood imported to New Zealand because it:
    - has a negligible likelihood of entering New Zealand on *P. americana* budwood (with low uncertainty) as:
        - the fungus is primarily a foliar pathogen and is not strongly associated with plant stems,
        - disinfection dip in 2% sodium hypochlorite (as per the commodity description) is likely to be effective in eliminating any infection on the surface of avocado budwood, and
        - if *P. gratissima* is imported into PEQ then it is likely to produce visible symptoms within the period of growth in PEQ.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall Level of assessed risk to New Zealand	Negligible	Low

6.2.18 *Phyllosticta perseae* (leaf spot disease)

Phyllosticta perseae is a fungal pathogen known to cause leaf spot disease in multiple *Persea* species. Affected plants typically have dark spots in lesions at leaf edges and may lead to leaf death.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Phyllosticta perseae* meets the criteria to be a hazard for New Zealand because it:

    - is not known to be present in New Zealand, and
    - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description), and it can potentially establish and cause harm to New Zealand.
- *Phyllosticta perseae* poses an overall negligible risk on *P. americana* budwood imported to New Zealand because it:
    - has a negligible likelihood of entering New Zealand on *P. americana* budwood (with low uncertainty) as:
        - *Phyllosticta perseae* is primarily transmitted on the leaves where it is highly likely to be detected, and infected material removed or destroyed, during preparation for export, in phytosanitary inspections pre-export or with surface disinfection on arrival at PEQ in New Zealand;
        - residual biosecurity risks from new infections or ungerminated conidia on the surface of avocado budwood are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ;
        - no evidence was found to show that the pathogen has latent infections or is endophytic in avocado

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall Level of assessed risk to New Zealand	Negligible	Low

6.2.19 *Pleiocarpon algeriense* and *Cylindrocladiella peruviana*

Pleiocarpon algeriense and *Cylindrocladiella peruviana* are causative agents of black-foot disease of grapevines which causes necrosis at the base of root stocks, reduced vigor of plants and occasional mortality. In avocado trees, both of these organisms cause stem and crown rot.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* meet the criteria to be hazards for New Zealand because they:
    - are not known to be present in New Zealand,
    - have the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description), and
    - have the potential to establish [and spread] in New Zealand, and cause harm to New Zealand.
- *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* poses an overall moderate risk on *P. americana* budwood imported to New Zealand because it:
    - has a low likelihood of entering New Zealand on *P. americana* budwood (with high uncertainty) as:
        - latent infections of *C. peruviana* and *P. algeriense* have been observed in avocado orchards as disease has only been observed in ≥3 year old plants,
        - *Cylindrocladiella peruviana* and *P. algeriense* infect cuttings of other host species, but there is uncertainty regarding the association with *P. americana* budwood
    - has a high likelihood of transferring from *P. americana* budwood to an appropriate host in New Zealand (with low uncertainty) as:
        - imported avocado budwood is intended to generate plants for planting in New Zealand and avocado plants (which are a suitable host for these pathogens) derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments, and
        - if the commodity is infected with these pathogens at the time of their release from PEQ and they are used as propagative material then the pathogens could be transferred to a suitable host on multiple occasions.
    - has a high likelihood of establishing in New Zealand (with low uncertainty) as:
        - hosts (in the form of cultivated avocados and other cultivated crops) are available for the establishment of *C. peruviana* and *P. algeriense*

        - the climate in parts of New Zealand is suitable for the establishment of *C. peruviana* and *P. algeriense*
    - may cause moderate overall impacts on New Zealand (with moderate uncertainty) as:
        - the economic impact of *C. peruviana* and *P. algeriense* to New Zealand is considered to be moderate, with moderate uncertainty
        - the impact on the environment from the establishment of *C. peruviana* and *P. algeriense* in New Zealand is considered to be very low, with moderate uncertainty
        - the human health impact of *C. peruviana* and *P. algeriense* to New Zealand is considered to be negligible, with low uncertainty
        - the sociocultural impact of *C. peruviana* and *P. algeriense* to New Zealand is considered to be low, with low uncertainty

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Low	High
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low
Impacts on the New Zealand economy, environment, health and society	Moderate	Moderate
Overall Level of assessed risk to New Zealand	Moderate	Moderate

6.2.20 *Podosphaera perseae-americanae*

Podosphaera perseae-americanae is a fungus causing powdery mildew on leaves of *Persea americana* (avocado) plants.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Podosphaera perseae-americanae* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because it:
    - is not known to be present in New Zealand,
    - has the potential to establish in New Zealand, and cause harm to New Zealand, and
    - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description).
- *Podosphaera perseae-americanae* poses an overall very low risk on *Persea americana* budwood imported to New Zealand because it:
    - has a low likelihood of entering New Zealand on *Persea americana* budwood (with moderate uncertainty):
        - *Podosphaera perseae-americanae* is likely to enter PEQ in dormant buds, and
        - *Podosphaera perseae-americanae* is not likely to survive on avocado budwood without causing symptoms throughout the PEQ period.
        - If symptoms develop, rapid production of airborne spores may not be detected early enough and measures in PEQ L3A may not prevent spores escaping containment facilities.
    - has a low likelihood of transferring from *Persea americana* budwood to a suitable host in New Zealand (with moderate uncertainty) because:
        - if airborne conidia escape from Level 3A PEQ, exposure of the pathogen to the New Zealand environment may then occur if avocado plants or plants from Lauraceae family are present near the PEQ facility. However, the amount of inoculum likely to escape is small, and the conidia would have to land on a suitable host (avocado is the only known host of *Podosphaera perseae-americanae*).
    - has a low likelihood of establishing in New Zealand (with moderate uncertainty) because:
        - potential hosts (in the form of cultivated avocados or native and introduced members of the Lauraceae family) are available for the establishment of *Podosphaera perseae-americanae*

        - climate consistently suitable for the establishment of the *Podosphaera perseae-americanae* is likely not readily available
    - may cause low overall impact on New Zealand (with moderate uncertainty) because:
        - avocados are the only known host of *Podosphaera perseae-americanae* commercially produced in New Zealand,
        - infection of avocado trees with *Podosphaera perseae-americanae* could cause yield losses,
        - an outbreak would likely spread quickly within and between regions,
        - an outbreak could affect avocados grown in gardens having some sociocultural impact, and
        - *Podosphaera perseae-americanae* could cause low environmental impact if native Lauraceae plants were susceptible to the disease

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Low	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	Low	Moderate
The likelihood of establishing in the New Zealand environment	Low	Moderate
Impact on the New Zealand economy, environment, human health and society	Low	Moderate
Overall level of assessed risk to New Zealand	Very low	Moderate

6.2.21 *Pseudocercospora purpurea*

Pseudocercospora purpurea is a fungal plant pathogen causing Pseudocercospora spot disease resulting in leaf, stem, and fruit spot symptoms in *Persea* spp. including avocado (*P. americana*). The disease is a serious pre-harvest disease affecting all cultivars of avocado in warm, humid, and rainy climates.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Pseudocercospora purpurea* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because it:
    - is not present in New Zealand and is listed as a regulated organism,
    - has the potential to establish in New Zealand, and cause harm to New Zealand, and
    - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description.
- *Pseudocercospora purpurea* poses an overall negligible risk on *Persea americana* budwood imported to New Zealand because it:
    - has a negligible likelihood of entering New Zealand on *Persea americana* budwood (with moderate uncertainty) because:
        - Symptomatic lesions are likely to be detected during pre-export phytosanitary inspection, on arrival inspection or while in the PEQ facility for 12 months.
        - The pathogen is known to be latent for approximately 3 months after penetration of susceptible plant material, after which spots are observed. However, given the process undertaken to import avocado budwood includes pre-export inspection, importation, on arrival inspection and further inspections over the period in PEQ for 12 months (assuming optimised conditions such as a warm, moist, and humid environment) the fungus is highly likely to produce symptoms before release.
        - Ungerminated spores on the surface of the imported material are likely to be managed by the 1% sodium hypochlorite treatment upon arrival in PEQ. If any ungerminated spores remain viable after the treatment on the surface of the budwood, they will likely either die or germinate once the buds break dormancy and produce visible symptoms within the PEQ period.
        - Spores are spread by watersplash (rain and irrigation), wind, insects, plant debris and contaminated tools and equipment. Given the measures undertaken in level 3A PEQ to manage risks associated with wastewater,

insects, waste, and accidental transfer, *P. purpurea* spores are unlikely to escape and infect a suitable host.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Negligible	Moderate
Overall level of assessed risk to New Zealand	Negligible	Moderate

6.2.22 *Pseudoidium perseae-americanae* (powdery mildew of avocado)

Pseudoidium perseae-americanae is a fungus that causes powdery mildew on the leaves, buds and herbaceous stems of *Persea americana* (avocado) plants.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Pseudoidium perseae-americanae* meets the criteria to be a hazard on imported avocado budwood imported to New Zealand because it:
    - is not known to be present in New Zealand,
    - has the potential to enter New Zealand on avocado budwood (as described in the commodity description).
    - has the potential to establish in New Zealand, and cause harm to New Zealand, and
- *Pseudoidium perseae-americanae* poses an overall very low risk on avocado budwood imported to New Zealand because it:
    - has a very low likelihood of entering New Zealand on avocado budwood (with low uncertainty) because:
        - *Pseudoidium perseae-americanae* has a reasonably wide geographical distribution but is mostly limited to tropical/subtropical climates,
        - It could be unknowingly imported into PEQ on leafless avocado budwood because mycelium can overwinter on dormant buds,
        - It is likely that *P. perseae-americanae* would produce symptoms on infected plants under suitable conditions and be detected in PEQ. Therefore, the likelihood of infected plants being released from PEQ is considered to be negligible, with low uncertainty, but
        - There is a very low likelihood that airborne conidia could escape level 3A PEQ because they could be produced between inspections and are small enough to fit though the vents. However, the amount of conidia likely to escape is small.
    - has a very low likelihood of transferring from avocado budwood to a suitable host in New Zealand (with low uncertainty) because:
        - if airborne conidia escape from Level 3A PEQ, exposure of the pathogen to the New Zealand environment may then occur if avocado plants are present near the PEQ facility. However, the amount of inoculum likely to escape is small, and the conidia would have to land on a suitable host (avocado is the only known host of *P. perseae-americanae*) under suitable conditions to germinate and cause disease,
    - has a moderate likelihood of establishing in New Zealand (with moderate uncertainty) because:
        - *Pseudoidium perseae-americanae* is mostly distributed in tropical/subtropical climates, indicating that the northern avocado growing regions of New Zealand may have suitable conditions for establishment (warm and humid), especially during summer,
        - the pathogen could overwinter as dormant asexual mycelia in buds,
        - *Pseudoidium* spp. can spread rapidly because they produce large amounts of airborne conidia within a short period of time (3–7 days) and do not require free water on plant surfaces to initiate infection,
        - conidia can be transmitted long distances by wind, but also by human activity on people, equipment or vehicles.
    - may cause a negligible overall impact on New Zealand (with low uncertainty) because:

        - *Pseudoidium perseae-americanae* is host specific to avocado and is only reported to cause disease in countries with tropical/subtropical climates. It is usually a minor disease problem but can become severe in prolonged warm, humid and shaded conditions, especially in nurseries,
        - it is not reported to be associated with fruit and can be effectively managed by fungicide treatments, suggesting that it is unlikely to have major disease or trade impacts,
        - *Pseudoidium perseae-americanae* is unlikely to cause any environmental, human health or sociocultural impacts because no records of such impacts were found in the literature.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Very low	Low
The likelihood of transferring from the imported commodity onto a suitable host	Very low	Low
The likelihood of establishing in the New Zealand environment	Moderate	Moderate
Impact on the New Zealand economy, environment, human health and society	Low	Low
Overall level of assessed risk to New Zealand	Very low	Low

6.2.23 *Pyrrhoderma noxium* (brown root rot)

Pyrrhoderma noxium is a tropical and sub-tropical fungus that causes root rot in a wide range of host species, frequently resulting in plant death. Avocado is reported to be a highly susceptible host of this fungus.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Pyrrhoderma noxium* meets the criteria to be a hazard on *P. americana* budwood imported to New Zealand because it:
    - is not known to be present in New Zealand, and
    - has the potential to enter New Zealand on *P. americana* budwood, establish in New Zealand, and cause harm to New Zealand.
- *Pyrrhoderma noxium* poses an overall negligible risk on *P. americana* budwood imported to New Zealand because it:
    - has a negligible likelihood of entering New Zealand on *P. americana* budwood as described in the commodity description (with low uncertainty) as:
        - *P. noxium* is not reported to be associated with shoots, stems or leaves of its hosts so it is unlikely that avocado budwood cuttings would contain mycelia,
        - *P. noxium* may produce airborne basidiospores which may then be present on the surface of avocado budwood. However, surface sterilisation of avocado budwood (as per the commodity description) is likely to make these spores inviable, and
        - symptoms on avocado are severe and develop rapidly, so are likely to be observed within the period of PEQ.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall Level of assessed risk to New Zealand	Negligible	Low

6.2.24 *Raffaelea* spp. (laurel wilt disease)

The following *Raffaelea* species were assessed: *R. aguacate*, *R. campbellii*, *R. canadensis* and *R. lauricola*.

Raffaelea spp. are typically saprotrophic fungi symbiotically associated with ambrosia beetles. However, *Raffaelea lauricola* is unique in that it causes the vascular wilt disease in trees and shrubs in the Lauraceae family including *Persea americana*. Similarly, *R. canadensis* has also been associated with wilt disease.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Raffaelea* species (*R. aguacate*, *R. campbellii*, *R. canadensis* and *R. lauricola*) meet the criteria to be a hazard on imported *P. americana* budwood imported to New Zealand because they:
    - are not known to be present in New Zealand:
    - have the potential to establish in New Zealand, and cause harm to New Zealand, and
    - have the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description).
- *Raffaelea* species pose an overall negligible risk on *P. americana* budwood imported to New Zealand because it:
    - has a negligible likelihood of entering New Zealand on *P. americana* budwood (with low uncertainty) because:
        - established *Raffaelea* infections are highly likely to be detected, and infected material removed or destroyed, during preparation for export, in phytosanitary inspections pre-export or on arrival in New Zealand;
        - residual biosecurity risks from ambrosia entry points or recent infections on the surface of avocado budwood are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ;

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall level of assessed risk to New Zealand	Negligible	Low

6.2.25 *Rhizoctonia noxia* (thread blight)

Rhizoctonia noxia is a fungal pathogen causing thread blight of coffee and many other plants including avocado. Damage from the disease typically occurrs on all parts of the plant except the roots.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Rhizoctonia noxia* meets the criteria to be a hazard for New Zealand because it:
    - is not present in New Zealand, and
    - has the potential to enter New Zealand on the commodity/pathway (as described), establish [and spread] in New Zealand, and cause harm to New Zealand.
- *Rhizoctonia noxia* poses an overall negligible risk on *Persea americana* budwood imported to New Zealand because it:
    - has a negligible likelihood of entering New Zealand on *Persea americana* budwood (with moderate uncertainty) as:
        - *Rhizoctonia noxia* is not likely to be imported into New Zealand on avocado budwood due to conspicuous symptoms,
        - if an intrepid infection on imported avocado budwood was not detected visually, then the pathogen would likely produce visible symptoms in PEQ.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Negligible	Moderate
Overall Level of assessed risk to New Zealand	Negligible	Moderate

6.2.26 *Rhizoctonia theobromae* (vascular-streak dieback)

Rhizoctonia theobromae is a leaf-penetrating, xylem-infecting, fungal pathogen that is the causal agent of vascular-streak dieback of cacao. One report was found of it causing vascular-streak dieback in avocado plants.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Rhizoctonia theobromae* meets the criteria to be a hazard on imported *Persea americana* (avocado) budwood imported to New Zealand because:
    - it is not known to be present in New Zealand,
    - has the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description),
    - has the potential to establish in New Zealand, and cause harm to New Zealand, and
- *Rhizoctonia theobromae* poses an overall negligible risk on *P. americana* budwood imported to New Zealand because it:
    - has a negligible likelihood of entering New Zealand on *P. americana* budwood (with moderate uncertainty) because:
        - *Rhizoctonia theobromae* has only been reported to infect *Theobroma cacao* (cacao) plants with the exception of one report of it infecting avocado plants,
        - new infections may not be detected prior to export as symptoms take 3-5 months to occur,
        - *Rhizoctonia theobromae* could be present inside leafless avocado budwood and such infections are unlikely to be managed by surface disinfection,
        - cacao budwood harvested from infected branches cannot be used for grafting because grafts do not take, meaning that infections are highly unlikely to be spread through grafting in cacao, however, this has not been tested in avocado,
        - if *R. theobromae* is imported into PEQ then it is likely to produce visible symptoms during the period of containment.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall level of assessed risk to New Zealand	Negligible	Low

6.2.27 *Scolecobasidium musae* (sooty blotch and flyspeck (SBFS))

Scolecobasidium musae is an epiphytic fungus that has occasionally been reported to colonise the surface of banana and avocado plants. It is considered to be a part of the sooty blotch and flyspeck complex (SBFS) which contains more than 100 species in 30 genera of Ascomycota and Basidiomycota.

Summary of PRA conclusions

Scolecobasidium musae meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because:

- it is not known to be present in New Zealand,
- has the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description), and
- has the potential to establish in New Zealand, and cause harm to New Zealand.

- *Scolecobasidium musae* poses an overall negligible risk on *P. americana* budwood imported to New Zealand because it:
    - has a negligible likelihood of entering New Zealand on *P. americana* budwood (with low uncertainty) because:

        - it can be present on the surface of budwood but such infections are likely to be manged by dipping in 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility,
        - if *S. musae* is imported into PEQ then it is likely to produce visible symptoms in PEQ.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall level of assessed risk to New Zealand	Negligible	Low

6.2.28 Sooty moulds

Sooty mould is a generic name used to describe a wide range of fungus species that cover the surfaces of above ground plant parts with a black, sooty coating and are generally associated with the sugary exudates of sap-sucking insects. Heavy infestation may inhibit photosynthesis and the unsightly mould may degrade the market value of produce.

Summary of PRA conclusions

Given the arguments and evidence presented:

- The sooty moulds (*Antennulariella batistae*, *Calothyrium apiahynum*, *Capnodium* spp., *Lembosia perseae*, *Meliola antioquiensis*, *Asteridiella perseae*, *Periconiella perseae*) meet the criteria to be a hazard on *Persea americana* budwood imported to New Zealand because they:
    - are not known to be present in New Zealand, and
    - have the potential to enter New Zealand on *P. americana* budwood, establish in New Zealand, and cause harm to New Zealand.
- These sooty mould species pose an overall negligible risk on *P. americana* budwood imported to New Zealand because they:
    - have a negligible likelihood of entering New Zealand on *P. americana* budwood as described in the commodity description (with low uncertainty) as:
        - sooty moulds may be present on the surface of budwood imported into New Zealand, but such infections are highly likely to be manged by 1% sodium hypochlorite dip for 2 minutes upon entry into the quarantine facility,
        - if sooty moulds are imported into PEQ, they may not produce visible symptoms during the period of containment but would become inviable,
        - if growth of sooty moulds occurs the symptoms are highly likely to be detected and it is unlikely that spores will be produced prior to detection, and
        - conditions aiding spore dispersal are unlikely to occur in PEQ.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Negligible	Low
Overall Level of assessed risk to New Zealand	Negligible	Low

6.3 Oomycetes

6.3.1 *Phytophthora mengei* (Phytophthora trunk canker)

Phytophthora mengei is a soil borne water mould (oomycete) that causes trunk cankers in avocado (*Persea americana*) plants. It can also infect the structural roots, lower limbs and fruit of the plant/tree.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Phytophthora mengei* meets the criteria to be a hazard on *P. americana* budwood imported to New Zealand because it:
    - is not known to be present in New Zealand,
    - has the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description), and
    - has the potential to establish in New Zealand, and cause harm to New Zealand.
- *Phytophthora mengei* poses an overall low risk on *P. americana* budwood imported to New Zealand because it:
    - has a low likelihood of entering New Zealand on *P. americana* budwood (with moderate uncertainty) because:
        - cankers/lesions on woody tissues have a brown necrotic appearance and are likely to be detected on arrival in New Zealand, or in PEQ.
        - in two studies, *P. mengei* symptoms developed on avocado plants within 40 days once spores germinated on susceptible tissue. Although no evidence was found of asymptomatic *P. mengei* infections or longer latent periods in avocado plants, other *Phytophthora* pathogens can be asymptomatic, or infect the xylem of hosts, and such infections may not be visually detectable. This highlights uncertainty regarding whether *P. mengei* infections can be asymptomatic or latent for longer periods.
        - *Phytophthora* spores are spread by watersplash, windborne rain and insects (ants, snails etc). Given the measures in level 3A PEQ to manage risks associated with insects and wastewater, waste and accidental transfer, *P. mengei* propagules are highly unlikely to escape to infect a suitable host.
    - has a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
        - avocado (*P. americana*) is a suitable host for *P. mengei*
        - avocado plants derived from grafting imported budwood onto suitable rootstock will be planted in New Zealand. These plants are likely be susceptible to *P. mengei* infection.
    - has a high likelihood of establishing in New Zealand (with low uncertainty) because:
        - avocado is the only reliably recorded host of *P. mengei*. There is a degree of uncertainty regarding the host associations of *P. mengei* because it is a recently described species (2009), formerly recognised as *P. citricola*. Host associations attributed to *P. citricola* may in fact correspond to *P. mengei*.
        - avocado is in commercial production and plants are commonly grown in northern parts of New Zealand, though less frequently elsewhere in the country.
        - *Phytophthora mengei* is found in California, USA, which includes some areas that have a similar climate to the parts of New Zealand.
        - *Phytophthora mengei* lesions produce abundant vegetative chlamydospores, sexual oospores and flagellated zoospores. *Phytophthora* spores can survive in adverse environmental conditions for several years and are spread by watersplash, windblown rain, animals, insects, humans, equipment and vehicles.
    - may cause moderate overall impact on New Zealand (with moderate uncertainty) because:
        - *Phytophthora mengei* has the potential to cause harm to New Zealand by reducing orchard health and productivity. The management costs associated with the pathogen may be partially mitigated because management of *P. cinnamomi* (already present in NZ) is likely to manage *P. mengei*.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Low	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low
Impact on the New Zealand economy, environment, health and society	Moderate	Moderate
Overall level of assessed risk to New Zealand	Moderate	Moderate

6.3.2 *Phytophthora palmivora* (bud rot of palms)

Phytophthora palmivora is an oomycete (or water mould) that causes a range of rot, canker and dieback symptoms on various host tissues.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Phytophthora palmivora* meets the criteria to be a hazard for New Zealand because it:
    - is not known to be present in New Zealand,
    - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description), establish in and cause harm to New Zealand, and
- *Phytophthora palmivora* poses an overall low risk on *P. americana* budwood imported to New Zealand because it:
    - has a low likelihood of entering New Zealand on *P. americana* budwood (with moderate uncertainty) because:
        - *Phytophthora palmivora* has an extensive global distribution,
        - *Phytophthora palmivora* can infect stems, indicating that it could be associated with budwood,
        - there is no evidence of asymptomatic *P. palmivora* infections or longer latent periods in *P. americana* plants, but asymptomatic infections are considered possible because other *Phytophthora* pathogens can be asymptomatic,
        - infections may be supressed and not visually detectable if suitable environmental conditions (wet and warm (above 22℃)) are not met in PEQ,
    - has a high likelihood of transferring from *P. americana* budwood to an appropriate host in New Zealand (with low uncertainty) because:
        - *Phytophthora palmivora* has a very wide host range including avocado and suitable hosts are widespread in New Zealand,
        - avocado plants derived from grafting imported budwood onto suitable rootstock will be planted in New Zealand. These plants are likely be susceptible to *P. palmivora* infection.
    - has a high likelihood of establishing in New Zealand (with low uncertainty) because:
        - hosts are widespread in New Zealand,
        - climate matching and modelling suggest that the pathogen is likely to infect plants and spread in northern New Zealand, especially in the summer. *Phytophthora palmivora* may be able to persist in the environment over winter as resistant spores.
    - may cause moderate overall impacts on New Zealand (with moderate uncertainty) because:
        - disease expression and spread are likely to be limited by climate, particularly in the south of New Zealand, though it may be more damaging in sheltered or protected environments such as nurseries and greenhouses.
        - *Phytophthora palmivora* causes a range of symptoms/diseases on numerous hosts in wet tropical and subtropical areas of the world. Some host plants are important economical crops in New Zealand, such as avocado, tomato, potatoes, kiwifruit, citrus and orchids.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	Low	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low
Impacts on the New Zealand economy, environment, health and society	Moderate	Moderate
Overall level of assessed risk to New Zealand	Moderate	Moderate

6.4 Viroids

6.4.1 Avocado sunblotch viroid

Avocado sunblotch viroid (ASBVd) is distributed in most avocado-producing areas globally and causes significant reductions in yield and fruit quality. All avocado cultivars have been reported as susceptible and infected asymptomatic trees play an important role in the epidemiology and spread of this disease

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Avocado sunblotch viroid* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because it:
  - is not known to be present in New Zealand
  - has the potential to establish in New Zealand, and cause harm to New Zealand, and
  - has the potential to enter New Zealand on *P. americana* budwood (as described in the commodity description).
- *Avocado sunblotch viroid* poses an overall moderate risk on *P. americana* budwood imported to New Zealand because it:
  - has a high likelihood of entering New Zealand on *P. americana* budwood (with low uncertainty) because:
    - *Avocado sunblotch viroid* can remain asymptomatic for the entire PEQ period.
  - has a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
    - imported avocado budwood is intended to generate plants for planting in New Zealand and avocado plants (which are a suitable host for these pathogens) derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments, and
    - if the commodity is infected with either of these pathogens at the time of their release from PEQ and they are used as propagative material then the pathogens will be transferred to a suitable host multiple times.
  - has a high likelihood of establishing in New Zealand (with low uncertainty) because:
    - a suitable host (in the form of cultivated avocados) is available for the establishment of *Avocado sunblotch viroid*, and
    - a suitable climate is available for the establishment of *Avocado sunblotch viroid*, and
    - *Avocado sunblotch viroid* had been reported from New Zealand prior to its eradication.
  - may cause moderate overall impact on New Zealand (with low uncertainty) because:
    - avocados are the only natural hosts of *Avocado sunblotch viroid* commercially produced in New Zealand, and
    - infection of avocado trees with *Avocado sunblotch viroid* could cause large yield losses and significant downgrading in quality of the fruit, and
    - control of an outbreak could involve wide scale destruction of avocado trees (e.g., removal of all infected trees), and
    - there are likely to be negligible trade restrictions, and
    - a slow spread of disease incidence.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	High	Low
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low
Impact on the New Zealand economy, environment, human health and society	Moderate	Low
Overall level of assessed risk to New Zealand	Moderate	Low

6.4.2 Potato spindle tuber viroid

Potato spindle tuber viroid (PSTVd) is a plant pathogen which consists of a small, unencapsidated (no coat protein), circular, single-stranded RNA molecule 356-361 nucleotides in length. It causes disease primarily in solanaceous plants, including potato, tomato and capsicum. However, the reported natural host range is large and includes avocado.

Summary of PRA conclusions

Given the arguments and evidence presented:

- *Potato spindle tuber viroid* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because it:
    - is not known to be present in New Zealand, and
    - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description), and
    - has the potential to establish in New Zealand, and cause harm to New Zealand.
- *Potato spindle tuber viroid* poses an overall moderate risk on *Persea americana* budwood imported to New Zealand because it:
    - has a high likelihood of entering New Zealand on *P. americana* budwood (with low uncertainty) because:
        - *Potato spindle tuber viroid* can infect its hosts systemically, and
        - can remain asymptomatic for the entire post entry quarantine (PEQ) period.
    - has a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
        - imported avocado budwood is intended to generate plants for planting in New Zealand and avocado plants (which are a suitable host for this pathogen) derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments, and
        - if the commodity is infected with this pathogen at the time of release from PEQ and used as propagative material then the pathogen will be transferred to a suitable host multiple times.
    - has a high likelihood of establishing in New Zealand (with low uncertainty) because:
        - suitable hosts in the form of cultivated avocados, tomatoes, capsicum, egg plants, potatoes, and reservoir plants in the form of weeds and ornamentals are available for the establishment of *Potato spindle tuber viroid*, and
        - the climate is suitable for the establishment of *Potato spindle tuber viroid*, and
        - *Potato spindle tuber viroid* had occurred transiently in New Zealand prior to its eradication, and
        - *Potato spindle tuber viroid* is readily transmitted by graft and mechanical means, seeds, pollen and irrigation systems.
    - may cause moderate overall impact on New Zealand (with low uncertainty) because:
        - direct impacts would likely occur on the tomato, potato and capsicum industry in New Zealand through yield losses, and
        - indirect impacts would occur from export or trade restrictions imposed by other countries, and
        - control of an outbreak would involve wide scale testing of growing areas and destruction of infected plants and trees.

The following table contains a summary of risk assessment against the criteria:

Criteria	Rating	Uncertainty
The likelihood of entry on the commodity, given the commodity description	High	Low
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low
Impact on the New Zealand economy, environment, human health and society	Moderate	Low
Overall level of assessed risk to New Zealand	Moderate	Low

Annex: Assessment Details

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1. Risk analysis process

The World Trade Agreement on the Application of Sanitary and Phytosanitary Measures (SPS agreement) states that phytosanitary measures must be supported by risk assessment and not maintained without sufficient evidence (SPS agreement; WTO 1995). That is, to require additional measures, MPI must have evidence that a pest or disease would not be sufficiently managed by the application of the minimum requirements.

The Biosecurity New Zealand process for undertaking an IRA builds on the existing international frameworks for risk analysis under the World Organisation for Animal Health (OIE) and the International Plant Protection Convention (IPPC), and extends the scope, under the SPS Agreement, to include all of the values required by the Biosecurity Act (1993)[4] (the Act).

The main output is an IRA which is used in the development or review of an IHS under the Act. An IHS specifies the requirements to be met for the effective management of risks associated with importing risk goods[5].

The Act requires a chief technical officer to begin the process of developing an Import Health Standard by: "*analysing or assessing the risks associated with importing a class or description of goods"[6].* While the Act does not state how the risks are to be assessed or analysed, it does state that the chief technical officer must have regard to certain matters when developing an IHS for recommendation to the Director-General. A number of these are part of an IRA as described by the OIE and IPPC:

- The likelihood that the goods will import organisms[7]
- The nature of the organisms that the goods may import
- The possible effect on human health, the New Zealand environment[8], and the New Zealand economy of the organisms that the goods may import
- In relation to requirements proposed for inclusion in an IHS, the extent to which the requirements reduce or manage the likelihood or impacts of adverse effects from organisms that may be imported on or in association with goods[9]

An IRA is also a relevant factor in how a country meets its obligations under the SPS agreement and other agreements such as the Convention on Biological Diversity (CBD). Under the SPS agreement, risk management measures either must be based on existing international standards, guidelines or recommendations, or must be supported by a scientific justification[10]. Measures must not be maintained without sufficient scientific evidence[11]. Measures must also not be unnecessarily trade restrictive[12]. In order to meet these obligations, the SPS agreement requires that measures are based on a risk analysis. Under the CBD, countries must consider environmental impacts in decision-making and prevent the introduction of, and control or eradicate alien species that threaten ecosystems, habitats and species.

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[4] Biosecurity Act section 24(4)(b)(iii) "…human health, the New Zealand environment, and the New Zealand economy…" As defined in section 2(1), environment includes (a) ecosystems and their constituent parts, including people and their communities; and (b) all natural and physical resources; and (c) amenity values; and (d) the aesthetic, cultural, economic, and social conditions that affect or are affected by any matter referring to paragraphs (a) to (c).

[5] Section 22, Biosecurity Act 1993

[6] Section 23(1) Biosecurity Act 1993

[7] From Section 2(1) Biosecurity Act 1993: "**organism**—(a) does not include a human being or a genetic structure derived from a human being: (b)includes a micro-organism: (c) subject to paragraph (a), includes a genetic structure that is capable of replicating itself (whether that structure comprises all or only part of an entity, and whether it comprises all or only part of the total genetic structure of an entity): (d) includes an entity (other than a human being) declared by the Governor-General by Order in Council to be an organism for the purposes of this Act: (e) includes a reproductive cell or developmental stage of an organism: (f) includes any particle that is a prion"

[8] From section 2(1) Biosecurity Act 1993, "**environment** includes— (a) ecosystems and their constituent parts, including people and their communities; and (b) all natural and physical resources; and (c) amenity values; and (d) the aesthetic, cultural, economic, and social conditions that affect or are affected by any matter referred to in paragraphs (a) to (c)"

[9] Section 23(4)(b) and (d) Biosecurity Act 1993

[10] SPS Agreement 1995 Article3(1) and (3)

[11] SPS Agreement 1995 Article 5(7)

[12] SPS Agreement Article 5(6)

The Biosecurity New Zealand (BNZ) process is equivalent to the process for plant health risk analysis under the International Standards for Phytosanitary Measures (ISPM) under the IPPC[13] (the main differences being in terminology).

1.1 Overview of the risk analysis process

The individual steps in the process for developing an IRA are the same for both the OIE and IPPC. However the standards, guidelines and recommendations for animal health (sanitary) and plant health (phytosanitary) measures are developed by different international bodies. This means that there are differences in the detail of how IRA is done for animal health (OIE) and plant health (IPPC). The BNZ process and methodology for undertaking an IRA is summarised in Figure 1-1.

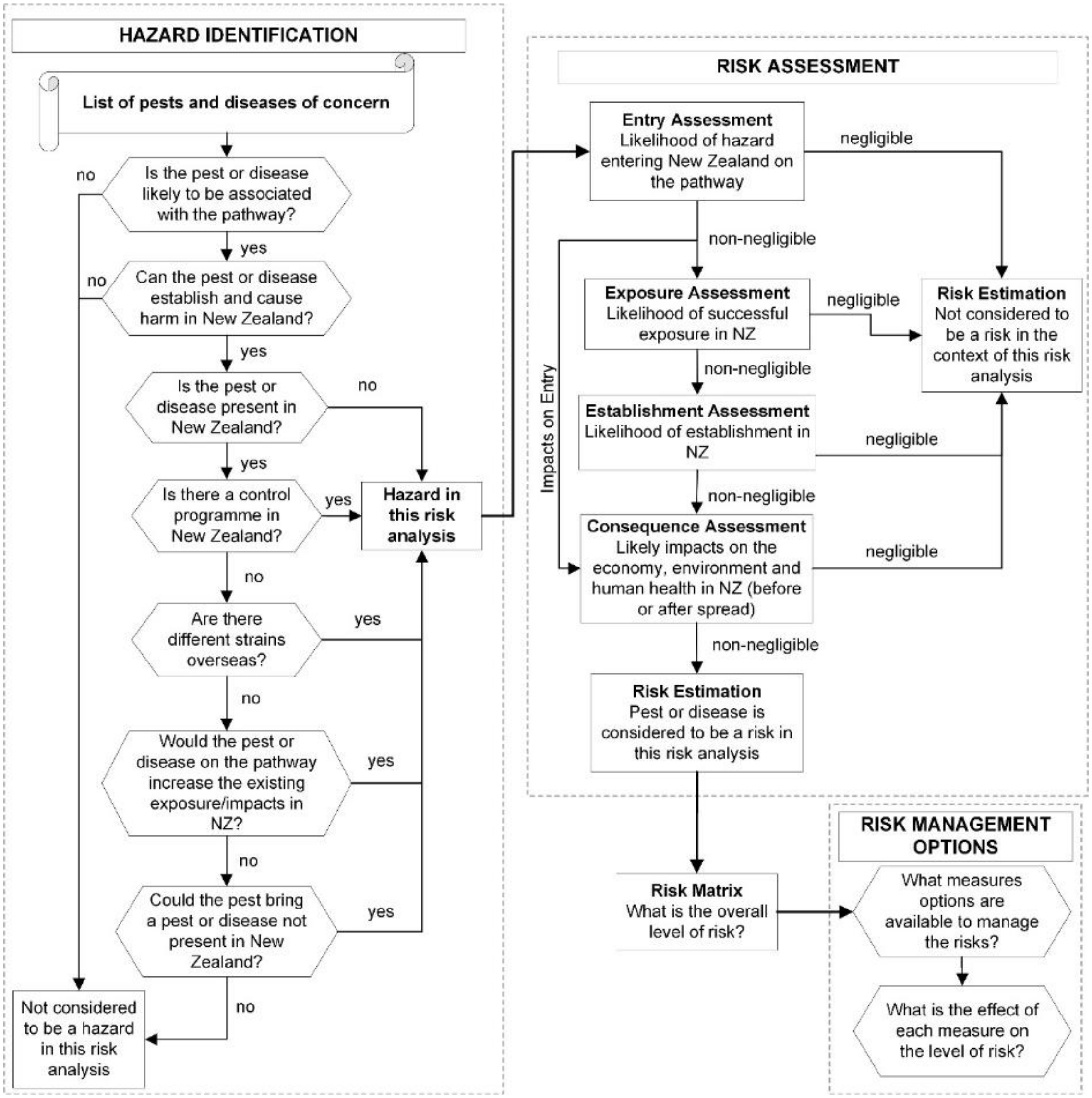

Figure 1-1: BNZ process and methodology for undertaking an import risk analysis

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[13] ISPM2: Framework for Pest Risk Analysis (FAO 1995).

1.2 Risk assessment criteria

For each of the likelihoods of entry, exposure, establishment, the following ranking scales will be used. The column containing the "quantitative guidance" provides a quantitative scale to help guide the analyst on the scale of each ranking.

Scale	Criteria	Quantitative guidance
High	Extending above the normal or average level	More than 2 events in 3 years
Moderate	Around the normal or average level	1 to 2 events in 3 years
Low	Less than average, coming below the normal level	1 event in 3 to 20 years
Very low	Close to insignificant	1 event in 20 to 100 years
Negligible	Not worth considering; insignificant	Less than 1 event in 100 years

For consequences, the following ranking scales will be used against the listed risk evaluation criteria:

Scale	Criteria	Quantitative guidance
Very High	Well above the normal or average level	More than 10 billion $NZ
High	Extending above the normal or average level	Between 1 and 10 billion $NZ
Moderate	Around the normal or average level	Between 100 million and 1 billion $NZ
Low	Less than average, coming below the normal level	Between 10 and 100 million $NZ
Very low	Close to insignificant	Between 1 and 10 million $NZ
Negligible	Not worth considering; insignificant	Less than 1 million $NZ

While the values provided in the quantitative guidance are New Zealand dollars over a 20 to 30 year period, the analyst considers how non-economic impacts can be monetarised (e.g., the dollar value of damage to ecosystem services) or considered equivalent (e.g., how much social impact is equivalent to these economic values).

In risk assessment, the uncertainty associated with each conclusion/contention for each likelihood and consequence is stated in each conclusion. The rationale for the uncertainty rating will be stated in the text as follows:

Scale	Criteria
High	• Scarce or no data available; evidence provided in unpublished reports of unknown authenticity; or • Few observations and personal communications; and/or • Authors' or experts' conclusions vary considerably
Moderate	• Some or only incomplete data available; evidence provided in small number of references; authors' or experts' conclusion vary; or • Limited evidence from field/lab observations; or • Solid and complete data available from other species that can be extrapolated to the species being considered
Low	• Solid and complete data available; strong evidence in multiple references with most authors coming to the same conclusions; or • Considerable and consistent experience from field observations

1.3 Hazard identification

Hazard identification[14] is the process for identifying pests and diseases associated with imported risk goods and which have the ability to cause harm to New Zealand. The process consists of compiling a list of potential hazards and then assessing them against criteria to see whether they warrant further consideration. The hazard identification answers two different questions:

- Does the species meet the criteria to be considered a biosecurity risk to New Zealand?
- Is the species potentially associated with the commodity that is being assessed?

The criteria for a quarantine pest for New Zealand are derived from the Biosecurity Act and the IPPC and OIE. These criteria are:

- Is the pest or disease absent from New Zealand?
- OR is the pest or disease present in New Zealand, but it meets one of the following criteria?
    - The pest or disease is under official control.
    - The species is a vector of a quarantine pest or disease.
    - There are subspecific taxa (subspecies, varieties, strains, etc.) within the pest species that are an increased risk to New Zealand compared with those already present.
    - There are other factors that would mean that the pest or disease may still be of concern in associated with imported goods (e.g. increased exposure to people through imported goods[15]).
- AND does the pest or disease have the potential to establish in New Zealand and harm "human health, the New Zealand environment, and the New Zealand economy[16]"?

Association with the commodity is based on:

- association with the commodity species or genus;
- association with the specific parts of the commodity; and
- whether a particular pest will stay associated with a commodity during preparation and/or transit to New Zealand – for example, a large flying insect is unlikely to stay on a piece of fruit when it is picked.

Different approaches may be taken to compiling and presenting the list of potential hazards depending on the information needed for pests or diseases in each pest/disease group. The approach for each group may be determined once the risk management question and risk evaluation criteria have been established. The specific approaches to hazard identification used in this import risk analysis are discussed further in Chapter 3.

1.4 Risk assessment

At the end of hazard identification, the list of hazards that require risk assessment is compiled. A risk assessment evaluates the likelihood of introduction (entry, exposure and establishment) and magnitude of consequence for a particular hazard, as well as the uncertainty in the conclusions. The SPS agreement describes the factors to take into account when assessing risk. These factors include:

- available scientific evidence;
- relevant processes and production methods;
- relevant inspection, sampling and testing methods;
- prevalence of specific pests or pathogens;
- relevant ecological and environmental conditions; and

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[14] Under the IPPC, the hazard identification process is known as pest categorisation in ISPM 2 (FAO 1995) and ISPM 11 (FAO 2013).

[15] One example is venomous spiders on fresh fruit. Even if present in a country, there may be a higher likelihood of people getting bitten if the spiders are associated with fruit sold at a supermarket.

[16] Biosecurity Act 1993

- potential damage in the event of the entry, establishment or spread of a pest or disease.

A qualitative approach is suitable for the majority of import risk analyses and is currently the most common type of assessment undertaken to support the development and review of import requirements.

1.5 Assessment of uncertainties

The SPS agreement states that measures must be applied *only to the extent necessary* and must be supported by *sufficient scientific evidence*[17]. Therefore, if there is insufficient evidence indicating that an organism meets the criteria for requiring additional measures (the risk evaluation criteria), then the lack of evidence suggests that it does not meet the critera to be a hazard (see section 1.3). However, in some cases there may be good reason to consider a pest or disease even when evidence is insufficient (e.g. similarity to known pests or diseases), and that pest or disease may be assessed further. If there is insufficient evidence but significant uncertainty, then the decision maker may apply measures, but under those circumstances the measures are provisional and further conditions may be required[18].

Therefore, documenting significant uncertainty is an essential part of a risk assessment. Uncertainties such as contradictions in the evidence or a lack of evidence are documented in the IRA. Where the risk assessment identifies significant uncertainty affecting the conclusion, this is indicated in the wording of the conclusion.

1.6 Review and consultation

Expert review is a fundamental component of a risk analysis to ensure the analysis is based on the most up-to-date and credible information available.

The IRA may be reviewed to check that the IRA is based on the best available and most credible information, that the document is clear and logical, that assumptions are valid and that conclusions are consistent with the evidence, with other conclusions in the IRA and with relevant conclusions in other assessments.

1.7 References for Chapter 1

FAO (1995) *International Standard for Phytosanitary Measures (ISPM) No. 2. Guidelines for pest risk analysis.* Secretariat of the International Plant Protection Convention, Food and Agricultural Organization of the United Nations, Rome, Italy.

FAO (2013) International Standards for Phytosanitary Measures (ISPM) No. 11. Pest rick analysis for quarantine pests. In *International Standard for Phytosanitary Measures No. 11*. Rome, Italy: Secretariat of the International Plant Protection Convention, Food and Agricultural Organization of the United Nations.

WTO (1995) World Trade Organization (WTO) Sanitary and Phytosanitary Measures Agreement (SPS). www.wto.org/english/tratop_e/sps_e/sps_e.htm Accessed February

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[17] SPS Agreement 1995 Article 2.2

[18] SPS Agreement 1995 Article 5.7

2. IRA scope and information

This import risk analysis (IRA) has been developed in response to a request to develop a new import health standard (IHS) for the importation of avocado (*Persea americana*) plants for planting. The purpose of the IRA is to identify and assess biosecurity risks associated with *Persea americana* budwood/leafless cuttings, and to identify risk management options to inform decisions on risk management. The decisions that this IRA will inform are:

- **a.** whether any pests or diseases on *Persea americana* budwood require additional measures.
- **b.** what measures are available that could manage the pest or disease risks to an appropriate level.

The objective of the IRA is to:

- **a.** identify pests (mainly pathogens such as viruses, viroids, bacteria, fungi, oomycetes and nematodes) and diseases of unknown aetiology that are associated with the commodity and may require additional measures (i.e. cannot be contained in a level 3A post-entry quarantine facility or be detected in a level 3A post-entry quarantine during a period of active growth with a minimum of three distinct phenological growing events ('shoot flush', see section 2.2)).
- **b.** assess these pests or diseases using a method that provides sufficient evidence about the biosecurity risks for a robust and transparent decision on whether additional measures are required to manage these pests or diseases.
- **c.** ascertain options for managing the identified risks to an appropriate level where this information is available in the literature.

For any particular risk good[19], there is a commodity definition that describes the minimum condition of the commodity that is covered by this IRA. The commodity definition is important as many pests and diseases may not be associated with a commodity that is defined in a certain way (e.g. more pests or diseases may potentially be associated with a commodity definition of fresh beans for consumption than a commodity definition of frozen beans for consumption).

2.1 Descriptions of the commodity and pathway

This risk analysis considers the effects on the New Zealand economy, environment, society and human health[20] from pests or diseases potentially associated with the importation of *Persea americana* budwood. A detailed description of the commodity and pathway is provided in Table 2-1.

Table 2-1: Detailed description of the commodity and the pathway(s) of entry to New Zealand

Commodity description
• Budwood/ leafless cuttings[21] of *Persea americana* and all its varieties and cultivars, brought into level 3A post-entry quarantine (3A PEQ) for a period of active growth with a minimum of three distinct phenological growing events (referred to as 'shoot flush'). • Budwood/ leafless cuttings are taken from mother plants. The cuttings do not have leaves, roots or soil attached to them, only buds. • Budwood/ leafless cuttings are dipped in 1% w/v sodium hypochlorite for 2 minutes upon entry into the quarantine facility. • The PEQ period involves visual inspection of plants for the presence of pests and symptoms. Visual inspections by an MPI inspector are usually carried out during active growing seasons - for other

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[19] Regulated article under IPPC

[20] Specifically, this IRA covers sections 23(4)(b)(i), (ii) and (iii) of the Biosecurity Act:
(i) the likelihood that the goods will import organisms:
(ii) the nature of the organisms that the goods may import:
(iii) the possible effect on human health, the New Zealand environment, and the New Zealand economy of the organisms that the goods may import

[21] As *Persea americana* is an evergreen species, the frequently used commodity type in the IHS of 'budwood/ dormant cuttings' will be changed to 'budwood/ leafless cuttings' to acknowledge the non-dormancy of the species.

Commodity description

plants for planting there are two inspections in the spring, two in the summer and one during the slower growth periods. A similar regime will be designed for avocados. Inspections by a facility operator are carried out at least twice a week.

All imported plants for planting must comply with basic conditions as set out in MPI Standard 155.02.06 (2020) “Importation of Nursery Stock” (IHS 155.02.06) . In addition to a period of PEQ, these include:

- Clean (including free from soil), inert/ synthetic materials for protection, packaging, and shipping.
- Phytosanitary certification stating that the exporting country has inspected the commodity and found it free of visually detectable regulated pests.
- Pesticide treatments for insects and mites.

Pathway description

Sea or air cargo from countries that have commercial avocado orchards and publish their research in English or Spanish languages.

2.1.1 Taxonomy and biology of *Persea americana*

Persea americana Mill. (avocado) is an evergreen tree belonging to the Lauraceae family which also includes plants such as *Cinnamomum verum* (cinnamon) and *Cinnamomum camphora* (camphor) (WFO 2021b). Although there are about 120 species in the genus *Persea* (WFO 2021a), avocado is one of the best-known because it is widely cultivated for its edible fleshy fruit.

The probable area of origin of avocado is the eastern and southern highlands of Mexico, through Guatemala to the Pacific coast of Central America (Knight Jr. 2002). *Persea americana* is a diverse species consisting of a number of botanical varieties corresponding to three ecological races (Mexican, Guatemaland and West Indian) and close relatives (Knight Jr. 2002; Scora et al. 2002; CABI 2021) adapted to different climates (Table 2-2):

- *Persea americana* var. *americana* (West Indian)
- *Persea americana* var. *drymifolia* (Mexican)
- *Persea americana* var. *guatemalensis* (Guatemalan)
    - The following contribute to the ancestry of var. *guatemalensis*:
        - *Persea americana* var. *nubigena*
        - *Persea americana* var. *steyermarkii*
        - *Persea americana* var. *tolimanensis*
        - *Persea americana* var. *zentmeyerii*
- *Persea americana* var. *floccosa* (wild botanical variety)

The races can hybridise readily and there are now many cultivars and varieties in commercial production (CABI 2021). Types with predominantly Mexican x Guatemalan germplasm are more suited to the subtropical regions, whereas West Indian or West Indian x Guatemalan hybrids are successful in tropical climates (Knight Jr. 2002; CABI 2021). In general, propagating an avocado variety from seed will yield a different variety to the mother plant, therefore, avocadoes are usually propagated vegetatively (Crane et al. 2019).

Table 2-2: Characteristics of West Indian, Guatemalan and Mexican races of *P. americana, in Florida USA (modified from Crane et al. 2019)**

Characteristics	Race		
	West Indian	Guatemalan	Mexican
Origin	Tropical lowlands	Tropical highlands	Tropical highlands
Blooming season	February to March	March to April	January to February
Maturity season	May to September	September to January	June to October
Development period (fruit set to maturity)	5 to 8 months	10 to 15 months	6 to 8 months
Cold hardiness - General rating	Low	Moderate to High	High
Cold hardiness - Young trees	-2 to -1°C	-3 to -2°C	-4 to -3°C
Cold hardiness - Mature trees	-4 to -1°C	-4 to -2°C	-8 to -3°C

* Tree response to freezing temperatures is influenced by tree health, stage of growth, and cultural practices. Hybrids of these races have intermediate characteristics.

Avocado is now grown in many tropical and subtropical countries in the Americas, Africa, Asia, Europe and Oceania (CABI 2021). Avocado is grown commercially and domestically in New Zealand and is naturalised in climatically suitable areas of the country. Approximately 95% of New Zealand's commercial production is of the 'Hass' variety (New Zealand Avocado 2021b), which is of predominantly Guatemalan origin but with some Mexican genes (Newett et al. 2002).

The avocado tree can grow to 20 metres and are perennial. The leaves are sometimes reddish when young before turning green. Small greenish-yellow flowers bloom in clusters (panicles). The fruit is a berry, consisting of a single large seed surrounded by a buttery pulp, and generally does not ripen until it falls or is picked from the tree (Crane et al. 2019). The average range of productive life in California has been estimated at 20 to 30 years, with an upper estimate of 40 years under favourable conditions (Goodall et al. 1971), while trees growing in the wild in Mexico have been known to live as long as 400 years[22].

Based on the established phenology growth model for avocado (Whiley et al. 1988), avocado has two distinct periods of extensive vegetative shoot growth flushes per year, referred to as 'growth flush' or 'shoot flush'. A growth flush of the roots ('root flush') either follows or coincides with a growth flush of aerial parts of the plant ('shoot flush') (Whiley 2002). Growth flushes are followed by periods of 'quiescence'. Avocados are alternate or irregular bearing trees, meaning that heavy crops in an "on year" alternate with smaller crops in the "off year". Alternate bearing can result from a heavy crop one year inhibiting shoot growth and flowering in the next year or from there being sufficient flowers but poor fruit set. These cycles may be initiated by environmental triggers such as frost or drought and maintained by internal physiological mechanisms. Tree carbohydrate accumulation, boron deficiency and poor pollination may all contribute to this alternate bearing pattern (Thorp 2011). Crop management practices such as mulching, application of plant growth regulators, orchard design and canopy management can all be used to reduce production fluctuations (Whiley 2002).

Avocado trees grown in New Zealand can have two or three growth flushes over a 12-month period. According to a study in the western Bay of Plenty, the first flush generally starts in September and finishes between mid-December and mid-January and the second flush runs from mid-January to the end of February/ mid-March (Dixon et al. 2008). There may also be a short period of shoot growth in autumn (mid-March to May). There is an obvious alternate bearing pattern with heavy fruit set in one year followed by poor or absent fruit set the next year. Different numbers of root flushes occurred at different times of the year in each study year (Dixon et al. 2008). In general, phenological events differed by several weeks from year to year. This variation in timing makes timing of tree management decisions challenging (Dixon et al. 2008).

Fruit matures on the tree but generally does not ripen until it falls or is picked from the tree. As a result, individual fruits may be harvested any time during their period of maturity which can vary according to variety (Crane et al. 2019). The main harvest season for Hass avocados in New Zealand is September to April (New Zealand Avocado 2021a).

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[22] http://ucavo.ucr.edu/general/answers.html

Rationale for minimum of 12 months in L3A PEQ:

To account for this unpredictable/fluctuation, holding plants in PEQ for at least one shoot flush period of each year (spring or summer) in two subsequent years would provide sufficient opportunity for plant and pathogen interactions and life-cycles to develop if the pathogen is present and for plants to be inspected for signs of disease. It also provides sufficient growth cycles and time for pre-determined testing for presence/absence of specified pathogens to be conducted. A suitable sequence of growth flushes may be one spring, one summer and one subsequent spring growth flush; or alternatively consist of two summer and one spring flush periods.

While plants held in PEQ will not mature and develop to the stage of flowering or fruit bearing (A grafted avocado tree can start producing fruit after three years (New Zealand Avocado 2022)), the principle of alternate (biennial) or irregular bearing applies as that characteristic is encoded into the avocado genome and not predictable (CABI 2021; New Zealand Avocado 2022).

Generally, only two growth flushes may be expected annually ('spring' and 'summer' flush); however, environmental conditions in greenhouse facilities may be artificially optimised to facilitate additional growth flushes and/or suppresses periods quiescence ('dormancy') during cold weather periods ('winter' period) (CABI 2021). Therefore, the minimum period in PEQ is set to no less than 12 months.

2.2 General information related to likelihood of entry

The following aspects of the commodity and pathway description will affect the likelihood that pests or diseases will enter New Zealand:

- The scope of the import risk analysis includes *Persea americana* budwood collected from all growing conditions and production methods from around the world and thus, various climates and growing regions need to be considered.
- While it is reasonable to consider that obviously diseased plants would not be used as a source of germplasm, many pathogens are not visible on the plant surface. Some pathogens have obvious symptoms on leaves, flowers, or fruit, but not stems. Plants infected with such pathogens are unlikely to be used as a source of budwood by commercial growers using known source plants. However, infected plants may be used for other purposes in the source location, such as for breeding programmes, or budwood may be harvested from plants that have not been observed over a long period of time.
- The import risk analysis therefore covers some pathogens or diseases with limited distributions or infrequent associations with *P. americana*, or pathogens that have obvious symptoms during at least part of the year. These pathogens have a much lower likelihood of being present in *P. americana* nursery stock than more widespread pathogens of *P. americana*; however, they cannot be ruled out as a risk, given the import risk analysis scope.

As described in the commodity description:

- Budwood/ leafless cuttings[23] of *Persea americana* and all its varieties and cultivars, brought into level 3A post-entry quarantine (3A PEQ) for a period of active growth with a minimum of three distinct phenological growing events (referred to as 'shoot flush').
- Budwood/ leafless cuttings are taken from mother plants. The cuttings do not have leaves, roots or soil attached to them, only buds.
- Budwood/ leafless cuttings are dipped in 1% w/v sodium hypochlorite for 2 minutes upon entry into the quarantine facility.
- The PEQ period involves visual inspection of plants for the presence of pests and symptoms. Visual inspections by an MPI inspector are usually carried out during active growing seasons - for other plants for planting there are two inspections in the spring, two in the summer and one during the slower growth periods. A similar regime will be designed for avocados. Inspections by a facility operator are carried out at least twice a week.

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[23] As *Persea americana* is an evergreen species, the frequently used commodity type in the IHS of 'budwood/ dormant cuttings' will be changed to 'budwood/ leafless cuttings' to acknowledge the non-dormancy of the species.

All imported plants for planting must comply with basic conditions as set out in MPI Standard 155.02.06 (2020) “Importation of Nursery Stock” (IHS 155.02.06). In addition to a period of PEQ, these include:

- Clean (including free from soil), inert/ synthetic materials for protection, packaging, and shipping.
- Phytosanitary certification stating that the exporting country has inspected the commodity and found it free of visually detectable regulated pests.
- Pesticide treatments for insects and mites.

As above, the standard also states that following arrival in New Zealand all nursery stock (unless specified in the schedules of special entry conditions) must undergo a period of post entry quarantine (PEQ) in order to check for the presence of regulated pests and/or diseases.

The minimum intervention to manage pest risk on *Persea americana* budwood/leafless cuttings that are not coming from approved offshore facilities will be:

- Budwood/ leafless cuttings are dipped in 1% w/v sodium hypochlorite for 2 minutes upon entry into the quarantine facility, and
- Level 3A post-entry quarantine (L3A PEQ) for a period of active growth with a minimum of three distinct phenological growing events (referred to as ‘shoot flush’).

Specification for the minimum period of post-entry quarantine for the IHS may be defined as follows. The post-entry quarantine period for *P. americana* plants for planting:

a. begins after imported plants held in a quarantine greenhouse have started active growth.
    i. for plants derived from imported budwood/ leafless cuttings, active growth begins when all buds grafted from the imported cuttings have developed fully expanded leaves;
b. must include a period of active growth with a minimum of three distinct phenological growing events (referred to as ‘shoot flush’), each of at least three months in length.
c. must not be less than a minimum of 12 months of active growth.
d. a period of quiescence (‘dormancy’) between summer and subsequent spring flush period must not be artificially suppressed or shortened.

As outlined in the Facility standard: Post Entry Quarantine for Plants, the structural and operational requirements of L3A PEQ include (MPI 2021):

- Vents must be screened with stainless steel insect-proof mesh with a maximum opening size of 0.04 mm (4.3.1.1 (7))
- A temperature controlled growing environment based on a mechanically ventilated heating and cooling system (4.3.1.1 (8)),
- All tools must be decontaminated (or discarded) between use on each plant (4.3.2.7 (2))
- All plants must be grown on raised benches with adequate drainage (4.3.2.7 (6))
    - All plants must be grown in individual containers (4.3.2.10 (3)).
- In addition, plants are inspected at least twice per-week during periods of active growth for signs of fungal infection (4.3.2.9 (1)), and symptoms are diagnosed (3.7.2) and controlled when found, so spore production is minimised. Plants showing symptoms of fungal infection will be destroyed, also to prevent spore production.
- Requirements for wastewater treatment
    - 4.3.2.5 Water (1): Only potable water may be used
    - 4.3.2.5 Water (2): Any water that is collected for re-use must be disinfected before reuse to ensure that it is free from pathogens
    - 4.3.1.1. (2): All water must be decontaminated before leaving the facility using a method proven to kill plant disease organisms
- Prevention of vector-driven pathogen dissemination, contamination of the surrounding environment and cross-contamination,
    - 4.3.1.1 Construction (7): Vents must be screened with stainless steel insect-proof mesh with a maximum opening size of 0.04 mm.
    - 4.3.2.4 Growing medium (1): Plants must be grown in pasteurised or inert medium

    - 4.3.2.10 Plant growing conditions (2): All plants must be grown in individual containers.
    - 3.4.4 Facility hygiene (2): Plant material form different consignments or different lots within the same consignment, must be effectively segregated to prevent cross-contamination
    - 3.4.4 (3): Procedures must be put in place to ensure that there is no mixing of plants from different lots or consignments
    - 4.3.2.7 Facility hygiene (6): All plants must be grown on raised benches with adequate drainage
    - 4.3.2.3 (2) yellow sticky traps must be used to monitor insects in the facility.
    - 4.3.1.3: the facility must be surrounded by a buffer strip minimum 1m wide which must be covered and free of plants
- Irrigation: Drip irrigation rather than over-head irrigation to be used
    - 4.3.2.10 Plant growing conditions (1): Plant growing conditions in L3A – (2) Specific plant requirements for irrigation, nutrition, temperature and winter chilling must be met.

In summary, in a Level 3A post-entry quarantine facility, all water leaving the facility must be decontaminated using a method proven to kill plant pathogens. The facility must have vents screened with insect-proof mesh with a maximum aperture of 0.2 mm, must be monitored for insects and must be built in such a way that no plants are within 1 m of the facility (MPI 2021). Additionally, plants are inspected for signs of infestation or disease at least twice a week during active growth and once a week during dormancy (MPI 2021). If found, any signs or symptoms are diagnosed and controlled. Therefore, if symptoms are due to fungal infection, spore production will be minimised.

All requirements must be met before plants are eligible for biosecurity clearance and released from PEQ. In addition, the Emerging Risks register is checked for new information on potential hazards on the pathway that may need assessment.

A summary of the import pathway of *P. americana* budwood into New Zealand from non-MPI accredited facilities is given in Figure 2-1

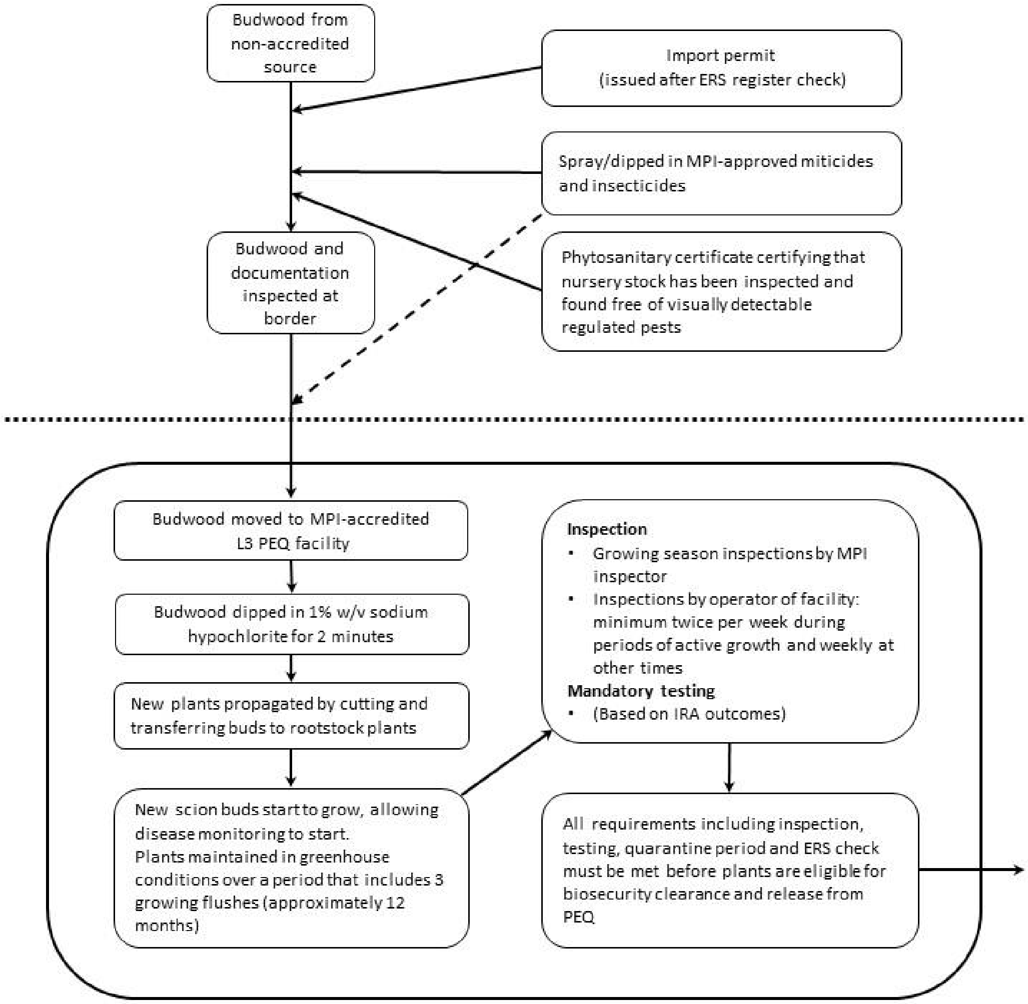

Figure 2-1 Summary of the pathway for the import of leafless *Persea americana* budwood from non-MPI accredited sources

2.3 General information related to likelihood of exposure and establishment

2.3.1 Exposure

When a pest or disease arrives in a new area, it usually needs to find or encounter a growing host plant or substrate in order to establish. The process of a pest or disease encountering a growing host plant or substrate is termed “exposure” in Biosecurity New Zealand risk assessments.

In the case of nursery stock, imported plant material is maintained in a habitat favourable for plant growth and is likely to be multiplied up to substantial numbers. Promoting the survival and persistence of the plant material also promotes the survival and persistence of the associated pests and diseases, resulting in a high likelihood of exposure, with low uncertainty for pests and diseases associated with germplasm/propagation pathways.

Imported avocado budwood (as described, see section 2.1) is intended to generate plants for planting in New Zealand. Therefore, avocado plants derived from the avocado budwood and released from Post Entry Quarantine (PEQ) will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand and used to propagate new plants for planting. Consequently, any undetected pests or diseases associated with avocado plant material at the time it is released from PEQ are highly likely to be associated with growing host plants in New Zealand, with low uncertainty.

2.3.2 Establishment and Climate

Climate factors may impact on the likelihood of pest or disease establishment in New Zealand.

New Zealand climates

New Zealand, in general, exhibits a mid-latitude oceanic temperate climate (Cfb temperate with maritime climate in the Köppen classification (Köppen 1936; Rubel and Kottek 2010)). Annual precipitation varies substantially due to topography, from dry eastern and inland areas to very high precipitation (on a global scale) in west coast areas[24]. The driest New Zealand climates barely reach a conventional water deficit on average (although they can do so in extreme years and months). The New Zealand climate 'space' can be visualised as a three-dimensional simplified climate niche (Figure 5 in Halloy 2021). This represents the average monthly temperature and precipitation for each month of the year for 42 New Zealand climate stations.

As a consequence of being a small landmass in a large ocean, New Zealand has relatively low diurnal and seasonal temperature fluctuations (equable climate). Rainfall is close to evenly distributed between summer and winter. These features are similar in equatorial climates. However, there can be relatively rapid variations between days as large weather systems alternate over the oceans. Although such variations may also occur in continental climates, a key difference for organisms is the proximity of these variations to biological thresholds (such as frost).

Unlike many Northern Hemisphere continental climates with reliable lengthy warm and cold periods, a small temperature variation in a mild oceanic climate means a more significant change in the number of days crossing threshold degrees, which may in some cases restrict the ability of some organisms to establish. This feature is also common to smaller land masses and mountains of the Southern Hemisphere, which is why, globally, some of the most similar climate conditions to New Zealand are in the montane regions of the Andean Yungas cloud forests and grasslands (Halloy et al. 2008). This is reflected in high morphological similarities of adaptations (Halloy and Mark 1996).

Together with tussock grasslands, temperate rainforests are the global biome climate types most represented in New Zealand. The northernmost areas of New Zealand approximate subtropical temperature conditions, with an ability to grow a range of subtropical plants.

The northern part of New Zealand is the most climatically suitable for the establishment of new pests and diseases coming from subtropical or tropical climates. The area includes Kaitaia, Kerikeri, Whangarei, Auckland (New Zealand's largest city) and Tauranga. The latter two cities both contain large active seaports. These are also the main avocado producing areas of New Zealand (see section 2.4.3).

Avocado is grown commercially mainly in the Bay of Plenty and Northland (87% of production areas). Together with Auckland (7% of avocado production) and Waikato (3%), these four regions are grouped in NIWA (2001) 'Northern New Zealand' climate zone because of the broad similarity of their climates. Within the Northern New Zealand climate zone there is some variation, but it can be broadly described as a subtropical maritime climate with low diurnal and moderate seasonal temperature variations. A wide variety of crops grow in this mild climate, including avocados, kūmara, and kiwifruit, as well as citrus, asparagus, berry fruits and a variety of other subtropical fruits (Plant & Food Research 2020).

Mean annual temperatures range from 13.8℃ (Hamilton) to 15.7℃ (Kaitaia, Whangarei)[25]. The mean range of temperature between warmest and coldest months is 8.8℃. The mean of the coldest month

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[24] This summary is written with crop pests and diseases in mind, focusing on the climate in inhabited and cultivated areas of New Zealand. Rainy outliers (e.g. Milford and Mt Cook) and high mountain climates are not considered, as they represent extremes with relatively low human and agricultural activity

[25] Data for this section is calculated from NIWA`s climate database (https://cliflo.niwa.co.nz/) representing the 'normal' from 1981-2010. For futher detail please refer to (Halloy 2021).

increases from 8.9℃ (Hamiltion) to 12.1˚C (Kaitaia). Means for the warmest months vary from 18.8℃ to 20.2℃. In Northland typical summer daytime maximum air temperatures range from 22℃ to 26℃, but seldom exceed 30℃. Winter daytime maximum air temperatures range from 12℃ to 17℃ (NIWA 2021).

Both hot and cold temperatures are mitigated by the maritime equable climate (Halloy 2021). Temperatures above 30℃ are rare, as are frosts. There are typically over 300 frost free days (for 0℃) and frosts below -2℃ occur rarely north of Auckland (Ward and Clothier 2020).

Growing degree days above 10℃ (GDD_{10}) for 2019 ranged from above 1700 (Hamilton) to close to 2400 (Whangarei) (Stats NZ Tatauranga Aotearoa 2020).

Annual precipitation ranges from just over 1100 mm (Hamiltion) to over 1300 mm (Pukekohe and most or Northland). Precipitation is distributed fairly evenly over the year, with slightly lower amounts in summer. Combined with warmer summers and cooler winters, there may be water stress during the summer, requiring supplementary irrigation (Ward and Clothier 2020) and there may be excess water in winter.

Avocados have a requirement for well drained, well aerated soils (Dirou 2003; Ward et al. 2019). This is an important consideration, in addition to climate, in understanding where avocado (and its pests and diseases) can thrive. Some of avocado's serious pathogens become a problem when roots are subjected to insufficient aeration and excess waterlogging.

The climate overlap between New Zealand and where a pest or disease has been reported can be assessed using the tool described in Phillips et al. (2018). This tool is based on the composite (or climate) match index (CMI) CLIMEX-MCR of CLIMEX version 3.3. The tool allows the comparison of New Zealand's climate in general or more specifically, with areas where a pest or disease occurs overseas. The CMI range extends from 0 to 1; the higher the CMI, the more similar the climate of the region to which the CMI pertains and the New Zealand climate. A CMI of 0.7 or more between the New Zealand climate and the existing range of the pathogen or disease may indicate a high likelihood of climatic suitability for that pest or diseases to establish in New Zealand (Phillips et al. 2018).

For many of the pests or diseases assessed, this tool may be a sufficient indication of climatic suitability in New Zealand for the pest or disease to establish. A whole-world map with CMI data from Phillips et al. (2018) is provided in Figure 2-2. When using the tool, the map can be enlarged to show more detail at a regional level.

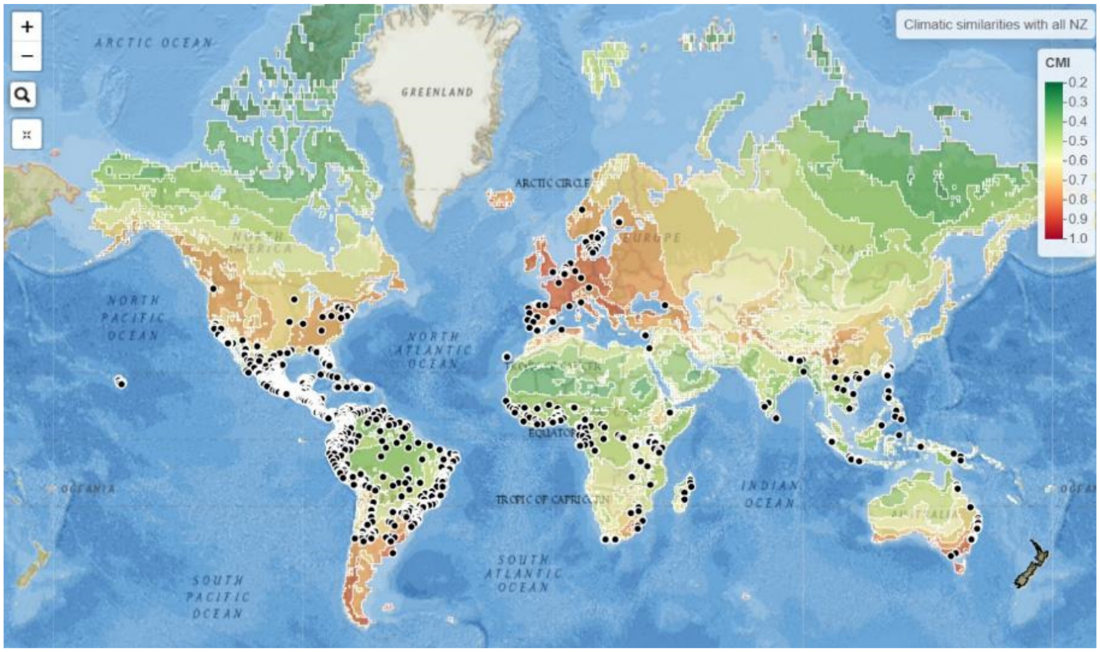


Figure 2-2 Climate match index (CMI). World climate similarities with All New Zealand (Phillips et al. 2018).

Shifting New Zealand climates with climate change

The global climate is warming rapidly and will continue to warm even faster as a result of greenhouse gas emissions (IPCC 2013, 2018; WMO 2019). Several studies have examined the effects of these changes on the New Zealand climate (NIWA 2017; Ministry for the Environment 2018). Warming temperatures and changing precipitation patterns will affect existing biodiversity, pests and diseases, and will change the likelihood of establishment of new arrivals (Gerard et al. 2013; Kean et al. 2015).

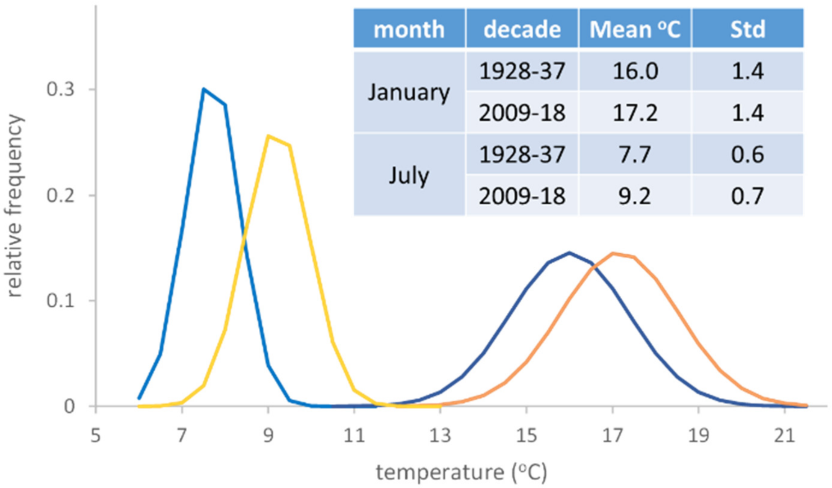
However, climate change is not just a scenario for the future. Average New Zealand temperatures have risen substantially over the last century, with concomitant but more variable changes in threshold and extreme conditions (including frosts, heatwaves, droughts, tropical storms, etc). Sea surface temperatures (SSTs) around New Zealand are closely correlated with atmospheric warming, and in recent decades (since 1981), have been rising in the order of 0.1–0.3℃ per decade (Sutton and Bowen 2019). This would equate to 1–3℃ per century, close to the modelled predictions for 2100. Such trends are, to some degree, hidden by inter-annual variability, but are already significant for the biota, agriculture and human occupation.

These ‘shifting goal posts’ may lead to some confusion when reading climate change scenarios. Climate scenarios are expressed as degrees of change up to 2100, for example, a 2℃ increase. However, the baseline for that increase may not be explicit to the reader. The baseline is sometimes referred to as ‘conditions pre-industrial age’, or ‘beginning of 20th century’. Other times, it may be the ‘1961–1990 average’, or more recently, the ‘1981–2010’ average. Because of the continuous change since the pre-industrial period, the first baseline would lead to a 2℃ increase by 2100. However, adding the same 2℃ to a 1981–2010 baseline would mean ~3℃ increase by 2100 in respect to pre-industrial conditions.

In New Zealand, some of the last years have already approximated a 2℃ increase on pre-industrial levels. Although cool years will still occur, for an organism, some conditions in some years already approximate some models for 2100. Model scenarios for temperature increases for 2100 depend on many factors, but most scientists now expect there is little chance of keeping the increases below 2℃ (Wallace-Wells 2017). For illustration’s sake, exploring a conservative 2℃ average increase (as in

NIWA scenarios[26]) and a 10-mm monthly increase in precipitation allows us to investigate the consequences without making any judgement as to which is more likely.

We can then consider the likelihood of such scenarios. To give an idea of how conservative a +2°C is, consider that the average temperature in the last century has already climbed >1°C over pre-industrial values. As averages shift, so do the extremes of the distribution and the thresholds for frosts, degree days, etc. Note, for example, that the midpoint of July 2009–2018 is where extreme warm years were in 1928–1937; the extreme has already become the norm (Figure 2-3)[27]. Moreover, the pace of increase is accelerating, greenhouse gas emissions are still rising, and a series of potential tipping points may soon be breached. Due to prolonged lag times, the abrupt decline in emissions triggered from March 2020 due to the Covid-19 pandemic are not likely to affects trends in the next decades. If such declines were to continue, they could reduce the centennial temperature increase.



month	decade	Mean °C	Std
January	1928-37	16.0	1.4
	2009-18	17.2	1.4
July	1928-37	7.7	0.6
	2009-18	9.2	0.7

Figure 2-3: **Schematic normalised frequency distribution of mean monthly temperatures for July (left) and January (right) for Wellington, New Zealand.**

The blue line is 1928–1937, the orange line 2009–2018, calculated from NIWA National Climate Database (https://cliflo.niwa.co.nz/). Note that actual frequency distributions tend to skew further right. As a result of such shifts, in future scenarios, frosts decrease substantially, by 30% (RCP 2.6) to 50% (RCP 8.5[28]) for the year 2040 (Ministry for the Environment 2018).

2.4 General information related to impacts of pests and diseases

2.4.1 Potential for spread

Availability of host species is unlikely to be a limiting factor for the spread in New Zealand of pests associated with avocado. Avocado is grown commercially and domestically in New Zealand and is naturalised in areas climatically suitable. For oligophagous pests (a narrow host range, but wider than avocado itself), plants in the same family as avocado (Lauraceae) are the most likely potential hosts. There are a number of native and exotic Lauraceae species present in New Zealand (section 2.4.2).

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[26] 2°C is an 'in between' of the 0.7°C to 3°C range of scenarios in NIWA: https://niwa.co.nz/our-science/climate/information-and-resources/clivar/scenarios, 20171221. However, in more detail, https://ofcnz.niwa.co.nz/#/localCharts shows an increase to 2100 of 2°C (RCP 6) to 3°C (RCP 8.5) annual mean (for six-model average) for Christchurch. Annual precipitation remains almost the same. Spring °C and mm follow the same pattern. Summer (DJF) increases less (1.5°C to 2.3°C). For rainfall, possible increases are around 10 mm; however, there will be considerable geographic variation and this is only illustrative.

[27] Note that climatologists typically prefer to use 30 years to represent a climatic period. Decadal periods are, however, useful for biological systems, as a decade is a long enough time to determine whether an organism can perish, establish or invade.

[28] In the latest IPCC report, RCPs (representative concentration pathways) are used to indicate emission scenarios, where 2.6 and 8.5 are the lowest and highest (e.g. IPCC 2013; Ministry for the Environment 2018).

For polyphagous pests (a wide host range) many suitable host species are likely to be present in New Zealand.

The majority of commercial avocado production (see section 2.4.3) takes place in the warmer frost-free areas of the upper North Island from Northland to the Bay of Plenty; however, some commercial production occurs in other regions of the North Island and in the upper South Island (Table 2-4). Domestic avocado trees are likely to be found in frost-free areas especially in the upper North Island.

Other *Persea* relatives in New Zealand are assumed to be grown in captivity or cultivation only and not naturalised or widely grown (see section 2.4.2).

The New Zealand native species of Lauraceae (see section 2.4.2) are most commonly found in the upper North Island, although *Beilschmiedia tawa* occurs as far south as the northern South Island (Allan 1982; NZPCN 2021). Non-native species of Lauraceae in New Zealand are largely grown in captivity/cultivation and are not naturalised, however, camphor laurel *(Cinnamomum camphora*) and bay tree (*Laurus nobilis*) are fully naturalised (Table 2-3).

The probable area of origin of avocado is Mexico/Central America. It is now grown in many tropical and subtropical countries in the Americas, Africa, Asia, Europe and Oceania (CABI 2021). The Climate Match Index (CMI) for New Zealand with many of these countries is below 0.7, indicating a poor climate match (see section *New Zealand climates*). However, for some countries/regions, e.g. France, Greece, Portugal, Spain, California, Chile, Argentina, the CMI is 0.7–0.9, indicating a good or very good climate match. This means that at least some pathogens that are associated with imported avocado plants are likely to encounter favourable conditions for establishment and spread and for disease expression, at least in the warmer parts of New Zealand.

2.4.2 Potential impacts on plant species other than *Persea americana*

Members of the same plant family share many pests and diseases in common. There is a strong relationship between how closely related different plant species are and how likely they are to share pests and diseases (for example, see Gilbert and Webb 2007)[29]. In addition to *P. americana* the following *Persea* species are known to be present in New Zealand: *P. borbonia*, *P. lingue*, and *P. thunbergia*. However, these species are probably growing only infrequently under cultivation and not naturalised as only *P. americana* is listed in the Manaaki Whenua – Landcare Research New Zealand plants database and the NZPCN website.

Persea is a member of the Lauraceae family. There are a number of native and exotic Lauraceae species present in New Zealand. Other genera in the family in New Zealand include *Beilschmiedia*, *Cassytha* and *Litsea* (native and some endemic; none are considered threatened (NZPCN 2021)) and *Laurus*, *Cinnamomum* and *Cryptocarya* (exotic, introduced as ornamentals and, in the case of the bay tree *Laurus nobilis*, for culinary purposes; some species are naturalised) (Table 2-3). Therefore, pests and diseases which are associated with avocado, but have a wider host range, have the potential for environmental and socio-cultural impacts.

Table 2-3: Species in the family Lauraceae reported as present in New Zealand in the Biota NZ (2022) and NZPCN (2021) databases.

Species	Common name	New Zealand Biostatus
Native species		
Beilschmiedia tarairi (A.Cunn.) Benth. & Hook.f. ex Kirk	taraire	Endemic. Not threatened
Beilschmiedia tawa (A.Cunn.) Benth. & Hook.f. ex Kirk	tawa	Endemic. Not threatened
Cassytha paniculata R.Br.	taihoa, mawhai	Native – not endemic. Not threatened
Cassytha pubescens R.Br.	taihoa, mawhai	Non-endemic

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[29] This phenomenon is widely used in risk assessment and policy setting for plant health (Gilbert and Webb 2007). See www.pnas.org_cgi_doi_10.1073_pnas.0607968104

Species	**Common name**	**New Zealand Biostatus**
Litsea calicaris (Sol. ex A.Cunn.) Benth. & Hook.f. ex Kirk	mangeao, tangeao	Endemic. Not threatened
Exotic species		
Cinnamomum camphora (L.) J.Presl	camphor laurel	Fully naturalised
Cinnamomum loureiroi Nees		Present in captivity/cultivation/culture
Cinnamomum cassia		Present in captivity/cultivation/culture
Cryptocarya obovata R.Br.		Present in captivity/cultivation/culture
Laurus nobilis	bay tree, laurel	Fully naturalised
Laurus azorica (Seub.) Franco		Present in captivity/cultivation/culture
Persea americana Mill	avocado	Fully naturalised
Neolitsea sericea (Blume) Koidz		Present in captivity/cultivation/culture
Lindera benzoin (L.) Blume		Present in captivity/cultivation/culture
Sassafras tzumu (Hemsl.) Hemsl.		Present in captivity/cultivation/culture
Sassafras albidum (Nutt.) Nees		Present in captivity/cultivation/culture
Machilus thunbergii Siebold & Zucc.		Present in captivity/cultivation/culture

2.4.3 Potential for economic impacts to the avocado industry

The New Zealand avocado industry comprises around 3,937 hectares divided between approximately 1,034 growers with most commercial production taking place in the Bay of Plenty, Auckland and Northland (Table 2-4, Figure 2-4). The most commonly grown variety is ‘Hass’ (NZIER 2016). Most avocados are sold as fresh produce although some are processed as avocado oil. Most produce is grown for the export market.

Table 2-4: Distribution of avocado (area planted, ha) by Regional Councils, year ended 30 June 2017 (Stats NZ Tatauranga Aoteaora 2021)

Region	Hectares	Region	Hectares
North Island of New Zealand		**South Island of New Zealand**	
Northland	2,000	Tasman	-
Auckland	430	Nelson	-
Waikato	100	Marlborough	-
Bay of Plenty	2,200	West Coast	-
Gisborne	50	Canterbury	30
Hawke's Bay	20	Otago	10
Taranaki	20	Southland	S*
Manawatū-Whanganui	-	Chatham Islands	-
Wellington	10		
Total North Island	**4,820**	**Total South Island**	**40**
		Total New Zealand	**4,870**

*S – data suppressed for reasons of confidentiality

A number of packhouses, fruit marketing companies and exporters are involved in the industry to market and sell avocados both domestically and overseas (NZIER 2016). In 2020 there were 13 licensed exporters, located in Northland, Auckland and Bay of Plenty, and 12 registered packers dealing with New Zealand avocados (NZ Horticulture Export Authority 2021).

Growers were estimated to have produced 39,078 tonnes of avocados in 2019/2020 (Plant & Food Research 2020). New Zealand domestic sales of fresh avocado fruit were worth NZ$50.6 million (Plant & Food Research 2020). In 2020, export sales (free-on-board value) of fresh fruit were worth NZ$100.1 million (Plant & Food Research 2020). Domestic sales of avocado oil were worth $2.6

million in 2019/20 and export sales (free-on-board value) worth $9.3 million in 2020 (Plant & Food Research 2020).

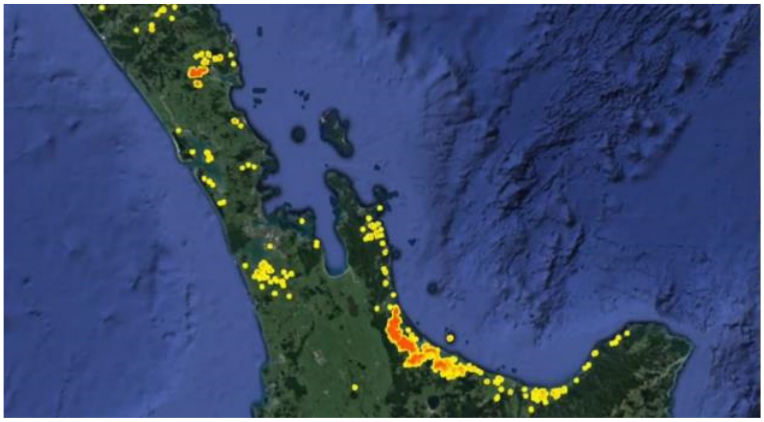

Figure 2-4: Main avocado growing areas in New Zealand

Australia is the main export destination with 75% of total export earnings in the year ended June 2020 (Table 2-5). Asian markets, particularly Thailand, South Korea, Taiwan, Japan and Singapore, account for most of the remainder.

Avocado is of significant economic importance to New Zealand, although not comparable to crop plants such as kiwifruit, grapevine or pine (NZIER 2016). Avocado was one of the four dominant fruit export products in 2020 (the others being kiwifruit, grape wine and apples) (Plant & Food Research 2020).

Table 2-5: Avocado export markets 2020 (year ending June) (NZ Horticulture Export Authority 2021)

Market	Volume (tonnes)	Value ($NZ FOB)
Australia	18,018	83,958,924
Thailand	1,727	9,786,986
South Korea	1,051	5,663,720
Taiwan	603	3,617,522
Japan	673	3,217,124
Singapore	633	2,687,279
China	302	1,258,401
India	194	1,023,353
Malaysia	163	660,228
Hong Kong	66	246,835
New Caledonia	37	180,240
Fiji	16	68,225
United Arab Emirates	17	66,115
Indonesia	9	46,208
United States of America	1	13,937
Pacific Islands	1	8,270
French Polynesia	0	1,254
Total	**23,512**	**$112,504,621**

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3. Hazard identification

A hazard is a pest or disease that is associated with imported risk goods and import pathways and that has the ability to cause harm to New Zealand. Hazard identification (hazard ID) is the process where, depending on the scope of the IRA, a list of pests and diseases potentially associated with the commodity is compiled and then assessed against specified criteria, in order to determine which species require further assessment (section 1.3). The objective for hazard identification in this IRA is to identify all pests and diseases that require further risk assessment to determine which ones may require additional measures to those in the commodity description.

In order to identify pests and diseases that require further risk assessment, potential hazards were considered using the following criteria:

- Potential to meet the criteria to be a quarantine pest
- Association with the commodity
- Potential to meet risk evaluation criteria for additional measures.

Pathogens which meet the risk evaluation criteria are those that cannot be contained in a Level 3A post-entry quarantine facility or be detected in Level 3A post-entry quarantine during a period of active growth with a minimum of three distinct phenological growing events ('spring-flush'). These may include pathogens with the following traits:

- The pathogens can be latent for long periods (more than 12 months, such as fungi which can be endophytic or pathogens which may only show symptoms under certain growing conditions).
- The pathogens can rapidly produce copious quantities of wind-dispersed spores.
- The pathogens are vector-transmitted, and there are vectors present in New Zealand and the vectors are small and mobile enough to enter a 3A post-entry quarantine facility, acquire the pathogen and then escape from the facility.

While the ability of a pest or disease to establish and cause impacts is part of the definition of a hazard, this is generally only considered indicatively at the hazard identification stage. It is assessed in depth at the risk assessment stage. Similarly, the ability of a pathogen to not be contained during a period of active growth in Level 3A PEQ is considered very broadly at the hazard ID stage and is considered at depth during risk assessment.

3.1 Hazard ID scope

- Pests (mainly pathogens such as viruses, viroids, bacteria, fungi, oomycetes and nematodes) and diseases of unknown aetiology associated with *P. americana* plants for planting from any country.
- Information is restricted to that in English and Spanish language sources.

3.2 Summary of hazard ID process

- Automated literature search supplemented by manual searches of additional sources
  - Sources included databases such as CABI, EPPO, USDA Fungal database, Web of Science (process outlined in the BNZ Guidelines for Undertaking an Import Risk Analysis Version 1.0)
- Some organisms are not considered further because risks are considered adequately managed by the commodity and pathway descriptions (e.g, arthropods, molluscs, vertebrates)
- Reduced list screened against hazard id criteria to produce list of hazards for risk assessment

Sixty-one species were identified at the hazard identification stage as requiring further pest risk assessment. These species were grouped into thirty-five PRAs of which 29 pests and diseases were found to present a sufficient level of risk to be considered for measures (Table 5-1). These conclusions were based on a species potentially meeting criteria for being a quarantine pest, being associated with the commodity, and potentially not being managed by basic measures (including the period in L3A PEQ) (Table 5-1).

Groups (e.g. genera or family) or high-profile species that did not meet the criteria for requiring pest risk assessment are listed in Appendix 1, along with the rationale for excluding them at the hazard identification stage.

3.3 List of Identified Hazards

From the list of potential hazards identified from the literature and other sources, the pests and diseases listed in Table 3-1 were identified through hazard ID to be hazards. For each of thse hazards a PRA was completed with the resulting risk level identified.

Table 3-1: Summary of IRA conclusions based on pathogen groups or species for which PRAs were carried out. PRA conclusion includes overall risk [uncertainty] of each pathogen.

Pest/disease group	Species requiring PRAs	PRA conclusions
Bacteria	Phytoplasmas on avocado	Moderate (vector) [high] Negligible (no vector) [low]
	Xanthomonas campestris and *X. axonopodis*	Moderate [high]
	Xylella fastidiosa	High [moderate]
Fungi	*Akaropeltopsis* sp.	Negligible [low]
	Ambrosia fungi of the Ambrosia *Fusarium* Clade	Negligible [low]
	Colletotrichum spp.	Moderate [moderate]
	Clonostachys pseudochroleuca	Negligible, [low]
	Cophinforma tumefaciens	Negligible [low]
	Diaporthe perseae	Low [moderate]
	Diaporthe sterilis	Very low [moderate]
	Dothiorella aromatica	Low [low]
	Elsinoe perseae	Negligible [low]
	Gliocladiopsis spp.	Very low [moderate]
	Grovesinia moricola	Negligible [low]
	Lasiodiplodia pseudotheobromae	Moderate – high [moderate]
	Mycosphaerella perseae	Negligible [low]
	Neocosmospora perseae	Low [moderate]
	Neofusicoccum nonquaesitum	Moderate [moderate]
	Pestalotiopsis longiseta	Low [moderate]
	Phyllachora gratissima	Negligible [low]

Pest/disease group	Species requiring PRAs	PRA conclusions
	Phyllosticta perseae	Negligible [low]
	Pleiocarpon algeriense and *Cylindrocladiella peruviana*	Moderate [moderate]
	Podosphaera perseae-americanae	Very low [low]
	Pseudocercospora purpurea	Negligible [moderate]
	Pseudoidium perseae-americanae	Very low [low]
	Pyrrhoderma noxium	Negligible [low]
	Raffaelea spp.	Negligible [low]
	Rhizoctonia noxia	Negligible [low]
	Rhizoctonia theobromae	Negligible [low]
	Scolecobasidium musae	Negligible [low]
	Sooty moulds	Negligible [low]
Oomycetes	*Phytophthora mengei*	Moderate [moderate]
	Phytophthora palmivora	Moderate [moderate]
Viroids	*Avocado sunblotch viroid*	Moderate [low]
	Potato spindle tuber viroid	Moderate [low]

4. Pest risk assessments: Bacteria

4.1 Phytoplasmas on avocado

Phytoplasmas are bacteria that lack cell walls. They colonise the phloem of their plant hosts as well as various parts of their insect vectors. Phytoplasma infection is associated with a wide range of symptoms affecting all plant parts and can be fatal to the plant. Phytoplasmas belonging to the stolbur group (16SrXII), likely 'Candidatus Phytoplasma solani', (16SrXII-A) have been identified in symptomatic avocado trees in Spain, and an unidentified phytoplasma has been identified in symptomatic avocado trees in Chile.

4.1.1 Taxonomic description

Name: *Persea americana*–infecting phytoplasmas

Kingdom/Class/Family: Bacteria/Mollicutes/Acholeplasmataceae

Taxonomic notes: Phytoplasmas are a well-defined clade in the class Mollicutes but cannot be classified by traditional methods. Phytoplasmas can be classified into "candidate" ('*Candidatus*') species by 16S rRNA gene sequences and they can also be classified into 16S rRNA gene groups and sub-groups based on restriction fragment length polymorphism (RFLP). There are currently over 30 16S rRNA groups and over 160 sub-groups recognised. There are currently around 40 recognised '*Candidatus*' species. Each of these corresponds to a 16S rRNA gene sub-group, but not every sub-group has been classified into a recognised candidate species. In the latter cases the 16Sr sub-group number and name can be used.

The species concept is defined within the phytoplasmas, but the appropriate level at which to conduct a risk assessment is often unclear. In this case, the phytoplasmas involved cannot be assigned definitely to a '*Candidatus*' species; it can only be said that they are *Persea americana*-infecting phytoplasmas (avocado-infecting phytoplasmas), with further information from one record (Laviña et al. 2002) that the phytoplasma detected is in the "stolbur" group, likely '*Ca.* P. solani' (16SrXII-A). The other record (Bäuerle Bascuñán 2002) is of an undetermined phytoplasma.

4.1.2 Hazard identification

New Zealand biostatus

Persea americana–infecting phytoplasmas are not known to be present in New Zealand:

Only two phytoplasmas are known to be present in New Zealand, neither of which have been reported in association with avocado (S Veerakone, pers. comm.; L Liefting pers. comm.; Veerakone et al. 2015; NZOR 2021; PPIN 2021):

- '*Candidatus* Phytoplasma australiense' infecting *Apium*, *Coprosma*, *Cordyline, Gomphocarpus*, *Phormium*, *Potentilla*, *Solanum* and *Rubus*; and
- '*Candidatus* Phytoplasma pruni' causing a (desirable) branching phenotype in *Euphorbia.*

It is very unlikely that the avocado-infecting phytoplasmas are already present in New Zealand because they are not likely to be either of these two phytoplasmas. The phytoplasmas from avocado trees in Spain is not the same strain as the '*Ca.* P. pruni' recorded in New Zealand because this is in group 16SrXIII (not 16SrXII, stolbur group, as recorded by Laviña et al. 2002). The phytoplasmas recorded from avocado trees in Spain by Laviña et al. (2002) or in Chile by Bäuerle Bascuñán (2002) are very unlikely to be '*Candidatus* Phytoplasma australiense', although this is one of the phytoplasmas within the 16SrXII "stolbur" group. '*Ca.* P. australiense' is only known to be present in New Zealand and Australia (L Liefting pers. comm.; Liefting 2020) and has not been reported from Spain or Chile. Furthermore, neither phytoplasma found in New Zealand has been recorded in New Zealand on avocado plants. Therefore, the current assessment has been carried out assuming that the phytoplasmas reported by Laviña et al. (2002) and Bäuerle Bascuñán (2002) are not present in New Zealand.

Avocado-infecting phytoplasmas are associated with *Persea americana* budwood because:

- *Persea americana* is a known host of at least one phytoplasma (Bäuerle Bascuñán 2002; Laviña et al. 2002);
- Phytoplasmas occur in association with budwood, as they are systemic through the plant phloem sieve tubes (Marcone 2009).

Avocado-infecting phytoplasmas have the potential to establish (and spread) in New Zealand because:

- If an infected plant grown from infected budwood is released from PEQ undetected the infected plant will be multiplied and planted in New Zealand avocado orchards. Plants originating from the original budwood may be infected, so avocado trees may be infected by these phytoplasmas.
- If competent vectors are present in New Zealand, they may transmit the phytoplasma/s to other New Zealand originating avocado trees and potentially to other hosts, depending on the vector's host range.

Avocado-infecting phytoplasmas have the potential to cause harm to New Zealand because:

- They may infect avocado trees and potentially cause yield loss, plant decline and death. While this has not been reported in avocado trees infected with phytoplasmas, phytoplasmas have caused this type of damage in other hosts (Agrios 2005).
- Depending on vector availability within New Zealand and the identity of the phytoplasma, the avocado-infecting phytoplasmas may cause decline or death in other plant hosts in New Zealand, as phytoplasmas have previously been reported to do (Agrios 2005). Some phytoplasmas e.g. '*Ca.* P. solani' have very wide host ranges (Quaglino 2020).

Given the arguments and evidence above, avocado-infecting phytoplasmas meet the criteria to be hazards on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

4.1.3 Risk assessment

Detections and identity of avocado-infecting phytoplasmas

Spanish detection (Laviña et al. 2002)

Laviña et al. (2002) reported finding "stolbur phytoplasma" on Hass cultivar avocado trees in Spain. They observed typical phytoplasma symptoms: leaf roll, leaf vein chlorosis with the leaves becoming small and abnormally red, and dwarfing; the symptoms were irregularly distributed on infected trees. Leaves from eight symptomatic plants taken from affected plots and leaves from two symptomless plants taken from healthy plots were analysed for phytoplasma infection by PCR assays. The stolbur phytoplasma was detected in trees by PCR using a stolbur-specific nonribosomal primer pair, or by nested PCR with 16S rDNA primers pairs. Symptomatic and asymptomatic trees were sampled in May, July, and October 2000, but phytoplasmas were detected only in samples collected from symptomatic trees in July. Phytoplasmas were detected by universal primers in all symptomatic samples analysed in July, whereas stolbur-specific primers gave positive results in only 75% of the symptomatic samples. RFLP analysis confirmed the phytoplasma belonged to the stolbur group.

The phytoplasma found in avocado trees in Spain was described as "stolbur" (Laviña et al. 2002). Stolbur can refer to the overall 16SrXII "stolbur" group, or specifically to 16 SrXII-A "Stolbur" (= '*Candidatus* Phytoplasma solani'). The stolbur phytoplasma referred to by Laviña et al. (2002) could be any of the phytoplasmas now recorded in the 16SrXII "stolbur" group (Table 4-1) but is most likely to be '*Ca.* P. solani' (see explanation below). There are currently thirteen groups (XII-A to XII-O) within the 16SrXII "stolbur" group, including five '*Candidatus* Phytoplasma' species. However, ‘*Candidatus* Phytoplasma australiense’ is only known to be present in New Zealand and Australia (although there are some erroneous reports from other countries) (L Liefting pers. comm.).

However, Laviña et al. (2006) studied Bois Noir disease in Spain and said it was caused by “***the stolbur phytoplasma****, which belongs to the stolbur group, also referred to as the* ***16SrXII-A group****. The disease is widespread in different vine growing regions in Spain, such as La Rioja, Navarra, Cataluña and Aragón. In Spain it has also been shown to affect different crops such as carrot, tomato, pepper, strawberry,* ***avocado*** *and asparagus*.” They cited Laviña et al. (2002) as evidence that this phytoplasma was found in avocado. It is therefore likely that when Laviña et al. (2002) refer to “the stolbur phytoplasma” they are referring to the 16SrXII-A group, which they said in 2006 was found in avocado. This is ‘*Candidatus* Phytoplasma solani’.

Table 4-1 Phytoplasma names and groups coming under 16SrXII – “stolbur”. 16SrXII-A to I from Bertaccini and Lee (2018). The “stolbur” found on avocado in Spain by Laviña et al. (2002) could be any of these although it is almost certain not to be ‘Ca. P. australiense’. It is most likely to be ‘Ca. P. solani’.

16Sr grouping	Strain (acronym)	‘*Candidatus’* species
XII-A	“Stolbur”	‘*Ca.* P. solani’
XII-B	Australian grapevine yellows (AUSGY)	‘*Ca.* P. australiense’
XII-C	Strawberry lethal yellows (StrawLY)	
XII-D	Japanese hydrangea phyllody	‘*Ca.* P. japonicum’
XII-E	Yellows diseased strawberry (StrawY)	‘*Ca.* P. fragariae’
XII-F	“Bois noir” (BN-Op30)	
XII-G	“Bois noir” (N-Fc3)	
XII-H	Bindweed yellows (BY-S57/11)	‘*Ca.* P. convolvuli’
XII-I	Potato strain from China (169/Hezuo 88)	
XII-J*		
XII-K*		
XII-N**		
XII-O***		

*New subgroups described in Quaglino et al. (2017)
**New subgroup described by Eroglu et al. (2010)
***New subgroup described in Kazeem et al. (2021)

Chilean detection (Bäuerle Bascuñán 2002)

In a master’s thesis Bäuerle Bascuñán (2002) reported the detection of an unidentified phytoplasma on symptomatic avocado trees in Quillón, Chile, using nested PCR. Phytoplasmas were not detected when conventional PCR was used. Nested PCR is more sensitive than conventional (or direct) PCR (Haqqi et al. 1988). Symptoms observed on infected trees were dwarfism, general chlorosis, short internodes, bud proliferation, and deformation of new leaves.

Biology and epidemiology of avocado-infecting phytoplasmas

Phytoplasmas are bacteria lacking cell walls. They are found in the phloem sieve tubes of host plants, and in insect vectors, and can multiply in both the plant and insect hosts (Bertaccini and Lee 2018). They are obligate biotrophic organisms, which cannot survive outside their plant or insect hosts (Oshima et al. 2004). Phytoplasmas spread systemically through the plant by passing through phloem sieve plate pores (Marcone 2009).

Phytoplasma diseases are known from more than a thousand plant species worldwide (Agrios 2005). They are quite often associated with severe and rapidly spreading plant diseases (Bertaccini and Lee 2018). Phytoplasmas are associated with a wide range of symptoms including virescence and phyllody of flowers, sterility, leaf yellowing, loss of apical dominance generating the proliferation of axillary buds with witches’ broom formation, abnormal internode elongation, abnormal root growth, generalised stunting, and a general decline that is sometimes fatal to the plant (Agrios 2005; Bertaccini and Lee 2018). Infection can also be asymptomatic (Bertaccini and Lee 2018).

There are two main mechanisms of transmission known for phytoplasmas: natural transmission by phloem-feeding insect vectors, but also by propagation or grafting of infected material.

Experimentally, phytoplasmas can be transmitted between plants via the vascular connection formed by dodder (*Cuscuta* spp.). Phytoplasmas are not reported to be spread by mechanical transmission (Lee et al. 2000), and only uncommonly by seed (Satta et al. 2019).

Vectoring

Phytoplasmas are naturally spread between plants by insects in the hemipteran families Cicadellidae (leafhoppers) and Psyllidae (psyllids) and superfamily Fulgoroidea (plant hoppers). These insects feed on the phloem of infected plants; therefore the phytoplasmas' host range is dependent on the host range of their vectors (Bertaccini and Lee 2018). The phytoplasma subgroups vary in the specificity of their relationships with insect vectors (Lee et al. 1998). Insect vectors have been identified for relatively few phytoplasma diseases and typically when new phytoplasma diseases are discovered little is known about the disease epidemiology (Weintraub and Beanland 2006).

Grafting

When phytoplasma-infected plants are grafted, often only a percentage of the resulting plants are infected with phytoplasmas. Some experiments examined rates of transmission of phytoplasma from infected hosts to grafted plants, showing varying rates (Table 4-2).

Table 4-2: ***Percentage of phytoplasma infected plants resulting from grafting. Some care is needed in interpreting these results, as different hosts, phytoplasmas and experimental techniques were used.***

Host	Phytoplasma / phytoplasma caused disease	% of phytoplasma infected plants resulting from the grafting	Reference
Grapevines	Bois noir phytoplasma	3%	Osler et al. (1997)
		1.7%	Credi et al. (2007)
Apple	Apple proliferation	0-30%	Pedrazzoli et al. (2008)
		6.13%	Magnago et al. (2002)
		90%	Jarausch et al. (2000)
		80-97%	Jarausch et al. (1999)
Apple to periwinkle	Apple proliferation	60%	Aldaghi et al. (2007)
Pear	Undetermined phytoplasma	29-85%	Pastore et al. (1998)
	European stone fruit yellows phytoplasma (EFSY)	69-94%	Jarausch et al. (1999)
Japanese plum	'*Ca.* Phytoplasma prunorum'	Initially 53%	Pastore et al. (2008)
Apricot	EFSY	Plants in 5 cultivars out of 10 infected	Pastore et al. (2001)
Japanese plum	EFSY	Plants in all cultivars infected	Pastore et al. (2001)

4.1.4 Likelihood of entry

This assessment is made on the basis that avocado-infecting phytoplasmas are present in the countries from which avocado budwood is being imported.

Phytoplasmas are associated with avocado trees (Bäuerle Bascuñán 2002; Laviña et al. 2002). As phytoplasma infection in avocado plants has only been reported in the literature twice, it is likely that it is not common. Avocado is a high volume, widely grown, high value crop, and it is likely that if phytoplasma infection was a more common problem in avocado it would have been reported more frequently. However, phytoplasma infection can be asymptomatic (Bertaccini and Lee 2018) so it is possible that avocado trees are infected with phytoplasma more frequently than symptoms are observed, although asymptomatic infection was not reported by Laviña et al. (2002) or Bäuerle Bascuñán (2002). So it is unlikely that budwood intended for importation into New Zealand would be taken from phytoplasma infected trees.

The volume of budwood entering New Zealand PEQ will be very low, all budwood is grown into plants which are kept growing in PEQ for a year and PEQ does not have space for many plants. Therefore it is unlikely that phytoplasma infected budwood would enter PEQ – it is unlikely that the few budwood cuttings taken for PEQ would be taken from infected plants.

As phytoplasmas are systemic through the phloem sieve tube elements (Marcone 2009), budwood cuttings taken from infected avocado plants could be infected with phytoplasmas. This infection is internal to the budwood so the likelihood of entry of the phytoplasmas would not be reduced by the surface sterilisation dip of budwood cuttings in dilute sodium hypochlorite (bleach dip).

Avocado budwood will enter PEQ and plants will be grown from the budwood for a period no shorter than 3 growth flush periods (and at least 12 months). During this period the plants are likely to be exposed to New Zealand summer-like conditions for part of the time. It is likely that any phytoplasma infecting the imported avocado budwood would express symptoms in the resulting plants during this PEQ period, although there is no information available on how frequently phytoplasma infections of avocado plants are asymptomatic. Avocado plants are checked at least twice weekly during periods of active growth. If phytoplasma-like symptoms are observed, a universal phytoplasma PCR test would be undertaken, which would be likely to detect any phytoplasma. 3A PEQ does not include pre-determined testing for phytoplasmas using PCR.

Given the arguments and evidence above, that is:

- One or more phytoplasmas have been reported to infect avocado (*P. americana*) trees;
- It is unlikely that avocado budwood is taken from phytoplasma-infected trees, since infections appear to be uncommon;
- There is only a low volume of budwood entering PEQ, which lowers the likelihood that infected budwood would enter PEQ even further;
- Avocado-infecting phytoplasmas occur in association with budwood, as phytoplasmas are systemic through the plant phloem sieve tube elements, and will not be reduced by budwood surface sterilisation;
- Plants will be held in PEQ and any avocado plants with phytoplasma infection are likely to show symptoms, and if so, diagnostic PCR tests will likely detect phytoplasma;
- However, some phytoplasma infections are asymptomatic so it is possible that infection would not be detected in PEQ;
- 3A PEQ does not automatically include pre-determined phytoplasma PCR testing;

Therefore, the likelihood of avocado-infecting phytoplasmas entering New Zealand associated with *Persea americana* budwood is considered to be very low, with moderate uncertainty.

4.1.5 Likelihood of exposure

This assessment is made on the basis that avocado-infecting phytoplasmas have entered New Zealand undetected.

When a pest or disease arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for some phytoplasmas.

If avocado plants are infected with phytoplasmas at the time of their release from Post Entry Quarantine (PEQ) they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3.1 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of avocado-infecting phytoplasmas in New Zealand from *Persea americana* budwood is considered to be high, with low uncertainty.

4.1.6 Likelihood of establishment

This assessment is made on the basis that avocado-infecting phytoplasmas have been successfully exposed to suitable host plants in the New Zealand environment.

As mentioned in the exposure step above, it is likely that avocado-infecting phytoplasmas will be present in avocado trees in New Zealand originating from imported budwood, and plants propagated from these e.g. by grafting, and plants taken from these grafted plants. It is not clear what percentage of grafted avocado plants taken from infected mother plants would be infected, likely less than 100%. Percentages of grafted plants also being infected by phytoplasmas ranging from 97% to as low as 1-2% have been recorded from some hosts (Table 4-2).

Plants that show symptoms of a phytoplasma infection are unlikely to be propagated from. If asymptomatic infected avocado trees are released from PEQ and used to graft new trees, it is likely that phytoplasma symptoms will develop eventually, and the presence of phytoplasma infected avocado in New Zealand will become known. The avocado nurseries and industry are likely to then put in measures such as genetic testing to detect phytoplasma infections in trees which are to be used for grafting, and only use trees with a negative result. In this way, spread via grafting could be reduced and potentially controlled in New Zealand. Then the only main way the phytoplasma could spread is through insect vectors.

There is a high degree of uncertainty in whether there will be a competent vector in New Zealand, for transmission between avocado trees, and from avocado trees to other hosts. As the phytoplasmas infecting avocado in southern Spain and Quillón in Chile are not known (other than in Laviña et al. (2002) it was a stolbur phytoplasma, likely *‘Ca.* P. solani’), the vectors of the avocado-associated phytoplasmas are not known. Therefore it is not known whether a competent vector is present in New Zealand. The known vectors of ‘*Ca*. P. solani’ are *Hyalesthes obsoletus* (main vector), *Reptalus panzer, Pentastiridius leporinus, Anaceratagallia ribauti,* and *Reptalus quinquecostatus.* These insects, and other insects in these genera, are not recorded from New Zealand (NZOR 2021; PPIN 2021). However, insects in the same families are present and it is not clear whether these can vector ‘*Ca*. P. solani’ or other phytoplasmas.

Two insects in the Hemiptera superfamily Fulgoroidea have been found on avocado in New Zealand, but it is not known if these could act as a vector of the phytoplasmas found in avocado. As mentioned above, some insects in Fulgoroidea are known to be vectors of some phytoplasmas. Adults of the introduced kiwifruit pest (*Scolypopa australis* (Fulgoroidea: Ricaniidae)) (passion vine hopper) have been reported on avocado plants in the wild in New Zealand (Logan et al. 2002; PPIN 2021). There are weak associations of another species with avocado. Four live nymphs of *Siphanta acuta* (Fulgoroidea: Flatidae) were found on avocado leaves in a visual survey on a commercial site in the Bay of Plenty, and pupae and egg masses were found on avocado leaves from another sample (PHA13737). In another case *S. acuta* eggs were laid on export avocado fruit (PHA2977, PPIN 2021).

It is possible that a competent vector could enter and establish in New Zealand in the future. Many exotic psyllids have established in New Zealand previously, such as *Bactericera cockerelli* (tomato-potato psyllid), *Cardiaspina fiscella* (brown lace lerp psyllid), *Ctenarytaina eucalypti* (blue-gum psyllid) (NZFFA 2009, 2022a). There are also many adventive species of *Acizza* (NZFFA 2022b) and “about a third of the New Zealand psyllid species are adventive” (Gordon 2010).

As the phytoplasma and possible vectors involved are not known, it is not possible to say whether there will be enough suitable host plants present in New Zealand for this phytoplasma to establish. There are plenty of avocado trees present in New Zealand, but it is not clear whether the phytoplasma can establish long term in avocado trees. If it is ‘*Ca.* P. solani’, many plants could be potential hosts. More than 100 species, belonging to 40 different families and 22 orders have been recorded to be infected by ‘*Ca.* P. solani’. This includes wild plants, ornamental plants, and major and minor crops. Crops affected include grapevines, tomato, potato, capsicum, celery, carrot, parsley, and garden bean (Quaglino 2020).

Climate is not likely to be a barrier to establishment of these avocado-infecting phytoplasmas. The avocado-infecting phytoplasmas were reported from areas which have a CMI (Composite Match Index, an index of climate similarity) of ≥0.7, indicating a similar climate to the whole of New Zealand, or northern New Zealand (Auckland and Northland Crosby code regions) respectively (Phillips et al. 2018; Roigé and Phillips 2021). Climate matches range from 0 to 1, with a higher number indicating a more similar climate. Northern New Zealand is currently one of the areas with the greatest production of avocado (the other is the Bay of Plenty). The relevant areas of southern Spain (major avocado growing areas) have a climate match with all of New Zealand of 0.8-0.9 (Halloy 2021); and mostly 0.7 but some 0.6 and 0.8 with northern New Zealand. Quillón in Chile has a climate match with all of New Zealand, and with northern New Zealand, of 0.8 (Phillips et al. 2018; Roigé and Phillips 2021). Therefore the phytoplasma is present in climates in Spain and Chile which are similar to climates in New Zealand and northern New Zealand, and climates throughout New Zealand are likely to be suitable for the establishment of avocado-infecting phytoplasmas.

Given the arguments and evidence above, that is:

- Asymptomatic infected avocado plants released from PEQ will be used for grafting. Avocado trees originating from these are likely to be infected, and likely to be spread widely in avocado growing areas;
- However if phytoplasma symptoms appear in avocado trees in New Zealand the avocado industry and nurseries will likely start testing for phytoplasma and only use clean stock;
- It is uncertain whether there will be a competent vector in New Zealand. Known '*Ca.* P. solani' vectors are not known to be present, but insects in the same family are present.
- Two insects with the potential to be vectors have been reported on avocado in New Zealand;
- It is possible that another competent vector could enter and establish in New Zealand in the future, and some related species have established here;
- If there is a competent vector in New Zealand potentially other host plants (e.g. grapevine, tomato, potato, capsicum) could be infected with the phytoplasma found in avocado trees;
- Climate is not likely to be a barrier to the establishment of these avocado-infecting phytoplasmas in New Zealand;

the likelihood of avocado-infecting phytoplasmas establishing in New Zealand in avocado trees originating from imported budwood is considered low, with moderate uncertainty. The likelihood of it establishing in unrelated avocado trees and other plant species hosts is moderate with high uncertainty if a competent vector is present in New Zealand and low with high uncertainty if there is no competent vector in New Zealand. So the overall likelihood of the phytoplasma establishing in New Zealand is considered to be moderate with high uncertainty if a competent vector is present in New Zealand and low with high uncertainty if there is no competent vector in New Zealand. It is considered that there is a low-moderate likelihood of there being a competent vector present in New Zealand (with high uncertainty).

4.1.7 Impacts in New Zealand

If there is no vector, then the only plants that will be impacted are some avocado trees propagated from the infected mother plant/s which were released from PEQ, and any other avocado trees propagated from those, if the industry does not start testing for phytoplasma in graft source trees. If there is a vector, then the phytoplasma may be able to be spread to more un-related (i.e. not related through grafting) avocado trees, or to other host plant species.

Economic impacts

There is very high uncertainty in the determined level of economic consequences, for the following reasons

- It is not known which phytoplasmas were found on avocado (one was in the stolbur group, likely '*Ca.* P. solani').
- It is not known if there will be a competent insect vector in New Zealand to transmit phytoplasma from avocado plant to avocado plant or to other plant species.
- It is not known what plants may host this phytoplasma in New Zealand, and what the potential severity of disease could be caused by this phytoplasma on those hosts.

If there is no vector, then the only plants that will be impacted are some avocado trees propagated from the infected mother plant/s which were released from PEQ, and any other avocado trees propagated from those, if the industry does not start testing for phytoplasma in graft source trees. So phytoplasma impacts would be limited to the damage the disease does to these avocado trees. It is not known how badly the phytoplasma would affect the tree; if it would cause reduced yield of avocado fruit or tree decline or death. There is no evidence of avocado tree decline or death. The two published records of phytoplasma in avocado (Bäuerle Bascuñán 2002; Laviña et al. 2002) only recorded mild phytoplasma symptoms on the observed infected trees; and did not state anything about more serious effects which would likely have been mentioned if they were present. Therefore if there is no vector present in New Zealand economic consequences are considered to be very low, with moderate uncertainty.

If there is a vector, then the phytoplasma may be able to be spread to more un-related (i.e. not related through grafting) avocado trees, or to other host plant species. If the phytoplasma can be transmitted to other host plant species, then economic consequences could be quite large. As mentioned above, '*Ca.* P. solani' can affect more than 100 plant species and have serious crop losses on grapevines (as bois noir), potatoes, tomatoes, capsicum and others (Quaglino et al. 2013; Quaglino 2020), some of which are of very high economic importance to New Zealand. Wine exports for example, were worth $NZ1.91 billion in 2020; domestic and export production of potato, tomato and capsicum were worth $NZ1.07 billion, $NZ143 million and $NZ60 million respectively (Plant & Food Research 2020). It is not known how much the presence of '*Ca.* P. solani' in this crop would reduce these amounts. Therefore with a competent vector present in New Zealand economic consequences are considered to be moderate with very high uncertainty.

Given the arguments and evidence above, that is:

- There is very high uncertainty in the level of economic consequences because the phytoplasma involved is not known, the presence of a competent vector in New Zealand is not known, and the possible host plants that could be affected and the severity of disease on those plants is not known.
- If there is no competent vector in New Zealand, the likely plants infected would be limited to avocado trees grafted from infected plants from PEQ and their offspring, and possibly industry would stop grafting from infected plants.
- If there is a competent vector in New Zealand, other crop plants may be affected but it is difficult to know which without knowing the phytoplasma or vector. Some phytoplasmas such as '*Ca.* P. solani' can have major impacts on many crop plants including reduced crop yield and plant decline and death.

The economic consequences of avocado-infecting phytoplasmas to New Zealand is considered to be:

- with a competent vector in New Zealand: moderate with very high uncertainty; and
- without a competent vector in New Zealand: very low, with moderate uncertainty.

Environmental impacts

The environmental consequences have a very high uncertainty, for similar reasons as those listed in the economic consequences section above.

A different phytoplasma has already had a large environmental impact in New Zealand. One of the two phytoplasmas known from New Zealand, '*Ca.* P. australiense', has particularly affected the iconic cabbage tree, *Cordyline australis,* with many trees killed by this infection (*Cordyline* sudden decline). In New Zealand *'Ca.* P. australiense' is also associated with the diseases *Phormium* yellow leaf (affecting flax), and *Coprosma* lethal decline (affecting coprosma). '*Ca.* P. australiense' is also associated with several weed and crop plants in New Zealand (Liefting et al. 2011). Note that it is assumed that the avocado phytoplasmas reported by Laviña et al. (2002) and Bäuerle Bascuñán (2002) are not *'Ca.* P. australiense'; however, this gives an example of phytoplasma impacts on native plants in New Zealand.

There is a very low likelihood that the phytoplasma could lead to decline or death in several species of native plants and trees, so the environmental impacts are considered to be low, with high uncertainty. This requires the presence of a competent vector.

Given the arguments and evidence above, the impact on the environment from the establishment of avocado-infecting phytoplasmas in New Zealand if there is a competent vector is considered to be low with high uncertainty. If there is not a competent vector in New Zealand, the impact on the environment is considered to be negligible with low uncertainty.

Human health impacts

No evidence was found of any human health impacts for phytoplasmas, as expected because phytoplasmas are exclusively plant pathogens, found in plant phloem or insect vectors.

Given the arguments and evidence above, the human health impact of avocado-infecting phytoplasmas to New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

There is also high uncertainty in this estimation, for the same reasons – uncertain phytoplasma, uncertain vectors, uncertain hosts, uncertain impacts on those hosts.

There is a very low likelihood that some garden plants (both amenity and food plants) and street amenity trees could decline or die. '*Ca.* P. solani' can impact grapevines, potato, capsicum, tomatoes and other plants which are grown in home gardens, as well as domestic avocado trees. This could lead to low sociocultural impacts.

There is a very low likelihood that avocado-infecting phytoplasmas could have impacts on Maori taonga plant species. A phytoplasma already present in New Zealand ('*Ca.* P. australiense') causes decline and death of tī kōuka (cabbage tree) and harakeke (flax) (Liefting et al. 2011), and there is a very low likelihood that a new avocado-infecting phytoplasma could impact native taonga plant species also. This depends on the presence of a competent vector.

Given the arguments and evidence above, the sociocultural impact of avocado-infecting phytoplasmas to New Zealand if a competent vector is present is considered to be low, with high uncertainty. If no competent vector is present in New Zealand the sociocultural impact is considered to be negligible with low uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, health and society if there is a competent vector in New Zealand is considered to be moderate, with high uncertainty. The overall impact if there is not a competent vector in New Zealand is considered to be very low, with low uncertainty.

4.1.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is very low (with moderate uncertainty);
- assuming entry, the likelihood of exposure is high (with low uncertainty);
- assuming successful exposure, the likelihood of establishment is moderate (with high uncertainty) if there is a competent vector in New Zealand, and low (with high uncertainty) if there is not a competent vector;
- the overall impact on the New Zealand economy, environment, health and society is considered to be moderate (with high uncertainty) if there is a competent vector present in New Zealand, and very low (with low uncertainty) if there is no competent vector present in New Zealand.

the overall level of assessed risk to New Zealand from avocado-infecting phytoplasmas on imported *Persea americana* budwood (as in the commodity description) is considered to be moderate (with high uncertainty) if there is a competent vector present in New Zealand; and negligible (with low uncertainty) if there is no competent vector. It is considered that there is a low-moderate likelihood of a competent vector being present in New Zealand, with high uncertainty.

4.1.9 Management considerations

Symptoms

In the two known detections of phytoplasma in avocado plants, symptoms shown by infected trees were

- leaf roll, leaf veinal chlorosis with the leaves becoming small and abnormally red, and dwarfing. These symptoms were irregularly distributed on infected trees (Laviña et al. 2002).
- dwarfism, general chlorosis, short internodes, bud proliferation, deformation of new leaves were observed in a Hass avocado orchard in Quillón, VIII region [Chile] in October 1999 (Bäuerle Bascuñán 2002).

It is likely that these symptoms would be exhibited in phytoplasma infections in avocado plants in PEQ. It is possible that other generally known phytoplasma symptoms may be exhibited too, such as leaf yellowing, loss of apical dominance and witches' broom type growth, generalised stunting, general decline and possible plant death (Agrios 2005; Bertaccini and Lee 2018).

ISPM 27 (*Diagnostic protocols for regulated pests: DP 12: Phytoplasmas*) has a very useful section on phytoplasma testing which goes some way towards answering the remaining management questions (references included in this quote are not included in the reference list; FAO 2016):

> *"Polymerase chain reaction (PCR) techniques are the method of choice for phytoplasma detection. Successful molecular detection of phytoplasmas is dependent on appropriate sampling of plant tissue and reliable nucleic acid extraction methods …. Phytoplasmas can be unevenly distributed and in an uneven titre throughout a plant, particularly in woody hosts, and symptomatic tissue is optimal for phytoplasma detection …. Symptomless infection can occur in some plant hosts and if this is suspected it is important to thoroughly sample different tissues of the plant.*
>
> *Phytoplasma titre in the plant host affects the reliability of the PCR test …. Phytoplasma titre can be affected by phytoplasma strain or species, host plant species, timing of infection and climatic conditions. The timing for sampling plant tissues is important as location in the plant and titre of phytoplasmas may be affected by seasonal changes ….*
>
> *For most phytoplasma diseases, leaves with symptoms are the best sources of samples for diagnosis. Phytoplasmas reside in the phloem sieve elements of infected plants and therefore the leaf petioles and midveins, stems or inner bark are often used for DNA extraction. In some cases (e.g. X-disease phytoplasma), fruit peduncles contain the highest phytoplasma titre ….. Although phytoplasmas can be detected in roots and bark scrapings of dormant trees, generally it is best to test for phytoplasmas at the end of summer."*

Known environmental conditions conducive to symptom expression

For phytoplasma in avocado plants, environmental conditions conducive to symptom expression are not known specifically, as there are only two publications of this relationship (Bäuerle Bascuñán 2002; Laviña et al. 2002), and these papers did not go into environmental conditions. Probably summer like conditions, see discussion in "optimal season" question below.

Taking samples for testing – plant parts maximising detection

Phytoplasma distribution and titre are known to vary throughout a plant and over time. Testing symptomatic tissue (e.g. symptomatic leaves) is optimal, but where symptomless expression is suspected it is important to thoroughly sample different tissues of the plant. Phloem containing plant parts such as leaf petioles and midveins, stems or inner bark are often used (FAO 2016). Testing in only one part of the plant can lead to false negative results when the plant does in fact have a phytoplasma infection (e.g. Donkersley et al. 2019).

Laviña et al. (2002) prepared DNA for PCR from leaf petioles, midribs, and trunk phloem of symptomatic trees (and from asymptomatic as controls). They did not state which of these plant parts gave positive samples, or if they separated out the plant part types, but they did get positive results for many of their samples, so at least some if not all of these plant parts gave positive results in symptomatic trees.

Bäuerle Bascuñán (2002) used mainly the midribs of the shoot leaves for PCR testing for phytoplasmas and obtained positive results in symptomatic trees.

The optimum season for conducting detection testing

The optimum season for conducting detection testing is likely the end of summer. Symptoms were seen on infected trees in summer (1999) in Spain (Laviña et al. 2002) and October 1999 in Chile (mid-summer, Bäuerle Bascuñán 2002). Furthermore while Laviña et al. (2002) tested for phytoplasmas in May, July, and October in the year 2000, they only detected phytoplasma in samples collected in July (mid-summer).

For phytoplasmas in general, generally it is best to test for phytoplasmas at the end of summer (FAO 2016). So summer like conditions would be best for testing.

PCR tests for phytoplasmas

Level 3 PEQ does not include pre-determined PCR tests for phytoplasmas. If universal phytoplasma PCR tests were used, either conventional nested-PCR or qPCR (MPI 2018), with multiple plant parts across the plant tested (as phytoplasma titre differs across the plant), the risk of entry of this phytoplasma would be considered to be reduced to negligible.

4.1.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Likely usual phytoplasma symptoms. Infected avocados have shown symptoms of leaf roll, leaf veinal chlorosis, with small red leaves, dwarfing, general chlorosis, short internodes, bud proliferation, deformation of new leaves.

- ***What are the known environmental conditions conducive to symptom expression?***

Probably summer-like conditions.

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Phloem containing plant parts from multiple areas across the plant as the plant may have uneven phytoplasma titre throughout the plant.

- ***What is the optimum season for conducting detection testing?***

Late summer.

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4.2 *Xanthomonas campestris* and *X. axonopodis* avocado pathovars

Xanthomonas species are gram-negative bacteria that infect a wide range of crops and wild plants. Two pathovars (strains that cause the same disease on the same host range) of *Xanthomonas* associated with disease in avocado (*Persea americana*) have been reported in the literature: *X. campestris* causing bacterial canker in California; and *X. axonopodis* causing bacterial leaf spot in the Seychelles.

Because there is very little information associated with *Xanthomonas* on avocado, information for the genus will be used where information for the pathovars is lacking.

4.2.1 Taxonomic description

Scientific name: *Xanthomonas campestris*

Order/Family: Xanthomonadales/ Xanthomonadaceae

Other names include: *Bacillus campestris, Pseudomonas campestris* (EPPO 2020)

Taxonomic notes: The papers identifying the *Xanthomonas* isolated from avocado in California (Cooksey and Azad 1992; Cooksey et al. 1993) used comparisons of morphology, biochemical characteristics, isozyme analysis and RFLP to assign the isolates to *X. campestris*. These papers predate a major revision of *Xanthomonas* taxonomy based on DNA-DNA hybridisation of 183 strains of the genus, which reassigned 65 pathovars and strains of *X. campestris* to 15 different species (Vauterin et al. 1995). It is therefore unclear which species the Californian *Xanthomonas* isolates from avocado belong to.

Scientific name: *Xanthomonas axonopodis*

Order/Family: Xanthomondales/ Xanthomonadaceae

Taxonomic notes: The paper identifying the *Xanthomonas* isolated from avocado in the Seychelles used multilocus sequence analysis to assign the isolates to *X. axonopodis* (Pruvost et al. 2009) sensu Vauterin et al. (1995). The species-level identity of this pathovar is therefore likely to be correct.

4.2.2 Hazard identification

Xanthomonas campestris is present in New Zealand (NZOR 2021; Biota NZ 2022) whereas *X. axonopodis* is recorded as absent from New Zealand (NZOR 2021; Biota NZ 2022) . The *Xanthomonas campestris* and *X. axonopodis* avocado pathovars are not known to be present (i.e. not listed as present in the literature) in New Zealand (Young et al. 2010; MPI 2021; NZOR 2021; PPIN 2021). However, there are records of other *X. axonopodis* pathovars being present in New Zealand (e.g. *X. axonopodis* pv. *begonia*, *X. axonopodis* pv. *glycines*, *X. axonopodis* pv. *poinsetticola*) (Biota NZ 2022). The only record of *Xanthomonas* from avocado (*Persea americana*) in New Zealand is a specimen lodged with the National Collection of Plant Pathogenic Bacteria (NCPPB) in the UK, identified as belonging to *X. hortorum* (Parkinson et al. 2009). Because collection details for this specimen are not supplied, it is not known if this is a field collected bacterium or an isolate from a bacterial collection in New Zealand. Additionally, since details of symptoms are not supplied for this specimen it is not possible to determine if it is a representative of either of the pathovars being assessed here. Therefore, there is insufficient evidence to indicate that either the *X. campestris* or the *X. axonopodis* avocado pathovars are present in New Zealand.

The *X. campestris* and *X. axonopodis* avocado pathovars have the potential to establish (and spread) in New Zealand because:

- hosts (in the form of cultivated avocados or native and introduced members of the Lauraceae family) are available for their establishment (NZPCN 2021), and

- suitable climate is available for their establishment (Cooksey et al. 1993; Phillips et al. 2018; California Avocado Commission 2020).

The *X. campestris* and *X. axonopodis* avocado pathovars have the potential to cause harm to New Zealand because:

- the *X. campestris* avocado pathovar causes bacterial canker of avocados (Cooksey et al. 1993; Eskalen and Faber 2016),
- the *X. axonopodis* avocado pathovar causes bacterial leaf spot of avocados (Pruvost et al. 2009), and
- avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$ 50.6 million (Plant & Food Research 2020).

The *X. campestris* and *X. axonopodis* avocado pathovars are associated with *P. americana* budwood:

- *Persea americana* is a known host of both pathovars (Cooksey et al. 1993; Pruvost et al. 2009), and
- both pathovars can be associated with *P. americana* budwood either intercellularly, in the vascular system or as epiphytes (Rudolph 1993; Stall et al. 1993; Jacques et al. 2016).

Given the arguments and evidence above, both the *X. campestris* and *X. axonopodis* avocado pathovars meet the criteria to be hazards on *P. americana* budwood (as in the commodity description) imported to New Zealand.

4.2.3 Risk assessment

Biology and epidemiology of Xanthomonas avocado pathovars

Host range

The host range of *Xanthomonas* pathovars is typically narrow and generally restricted to a single species or a small number of representatives within a single family but a small number of pathovars have broader host ranges. For example, *X. campestris* pv. *campestris* is able to infect all members of the Brassicaceae family (Jacques et al. 2016). Therefore, it is likely that the host range of the *Xanthomonas* pathovars being assessed here is restricted to *P. americana* although it may include other members of the Lauraceae family. The *Xanthomonas* avocado pathovars assessed in this PRA have been recorded in California, USA (Cooksey and Azad 1992; Cooksey et al. 1993) and the Seychelles (Pruvost et al. 2009).

Symptoms

The *X. campestris* avocado pathovar causes lesions on trunks and branches. The lesions appear as sunken dark areas with a necrotic, watery pocket underneath. The lesions develop into cankers that split and ooze fluid that dries to a powdery white residue. Cankers are usually 2-10cm in diameter and usually appear at the base of trees before spreading upwards, usually in a straight line along one side of the trunk or branch. Necrotic streaks extend above and below the necrotic lesions underneath cankers (Cooksey et al. 1993; Eskalen and Faber 2016).

Genetic and pathogenic variability has been observed for the *X. campestris* avocado pathovar (Cooksey and Azad 1992). In California, 26 strains were isolated from avocado canker symptoms and some of these strains appeared to be nonpathogenic (but genetically related to pathogenic strains) (Cooksey and Azad 1992). It remains unclear whether the canker is caused by a single pathovar or a mixture of strains from other hosts (Cooksey and Azad 1992).

The *X. axonopodis* avocado pathovar causes small, black, water-soaked, angular leaf lesions on avocados. These lesions sometimes coalesce into larger lesions (Pruvost et al. 2009).

Life cycle and transmission

In nature, cells of *Xanthomonas* are deposited on plant surfaces by water splash or aerosols. They may enter a prolonged, asymptomatic, epiphytic phase, persisting on the plant surface for up to several months before entering plant cells and causing symptoms. Because xanthomonads lack active mechanisms for entering plants, they can only enter host plants through natural openings such as stomata (gas exchange pores found in plant epidermis) or hydathodes (water secreting pores along the leaf margins of some plants), or through wounds. High relative humidity favours penetration of bacteria into plants (Rudolph 1993; Stall et al. 1993; Ryan et al. 2011).

Many pathogenic xanthomonads show a high degree of tissue specificity, either invading the vascular system of hosts (after entry through hydathodes or wounds) or the mesophyll tissue (after entry through stomata) but some species or pathovars are capable of colonising both (Ryan et al. 2011). There is no information on which tissues are colonised by either the *X. campestris* or *X. axonopodis* avocado pathovar. In addition, there are some epiphytic xanthomonads that are not plant pathogens (Vauterin et al. 1996) which can interfere with diagnosis of pathogenic xanthomonads (Taylor et al. 2002).

Multiplication of bacteria within plant tissues is accompanied by the degeneration of plant tissues. Most xanthomonads are highly competent in causing infection and even a single bacterial cell can be enough to cause infection (Rudolph 1993; Stall et al. 1993).

Long distance spread of xanthomonads is generally by human movement of contaminated or infected propagative materials, which is the main pathway for international spread of xanthomonads (Stall et al. 1993). Shorter-distance spread is generally by wind-blown water (e.g. rain splash) (Stall et al. 1993). A wide range of dispersal distances is possible with this dispersal method, with studies of citrus canker in Florida recording spread distances of between 12m and 3.5km, although tornadoes and tropical storms can be associated with spread of up to 11km (Gottwald 2000).

Time to symptom development and factors influencing symptom expression

The timeline of symptom development following infection by the *X. campestris* avocado pathovar is not well described but Cooksey et al. (1993) note that, in their experiments, necrosis was visible on avocado stems four weeks after inoculation. However, in this experiment, wounded plants were inoculated with very high levels of inoculum (i.e. 1011 cfu compared to other studies which tend to use < 108 cfu, Pruvost et al. 2009) and thus, demonstrated a bias to cause infection (Cooksey et al. 1993). Detailed information on the factors affecting symptom development of the *X. campestris* avocado pathovar is unavailable, although drought stress and boron deficiency may promote symptom development (Eskalen and Faber 2016).

The timeline of symptom development following infection by the *X. axonopodis* avocado pathovar is also not well described. In inoculation experiments, where the youngest leaves of an avocado plant were infiltrated with a concentrated bacterial suspension via a needleless syringe, symptoms developed in 6-8 days (Pruvost et al. 2009). No further information on factors affecting symptom development is available for the *X. axonopodis* avocado pathovar.

Although symptom expression of other xanthomonads is affected by factors such as temperature and humidity, optimum levels of these variables for symptom expression differ between species and pathovars (Stall et al. 1993). For example, optimum *in vitro* growth temperatures can range from 20–23˚C for cool weather adapted *Xanthomonas* such as *X. populi* (Ridé and Ridé 1992) to 30 ˚C for warm weather adapted types like *X. citri* subsp. *citri* (Stall et al. 1993). Development of walnut blight caused by *X. arboricola* pv. *juglandis*, on the other hand, appears to not be influenced by temperature (CABI 2021b). Given the range of temperature and humidity optima for species and strains of *Xanthomonas* the information cannot be generalised to the avocado pathovars being assessed here.

4.2.4 Likelihood of entry

This assessment is made on the basis that the *X. campestris* and *X. axonopodis* avocado pathovars are present in the countries where avocado budwood is being imported from. The *X. campestris* avocado pathovar is only reported from the United States of America (Cooksey and Azad 1992; Cooksey et al. 1993) and the bacterial canker it causes is known to be widespread only in California (Eskalen and Faber 2016). The *X. axonopodis* avocado pathovar is only reported from the Seychelles (Pruvost et al. 2009).

In plants, *Xanthomonas* can colonise all aerial plant parts (i.e. stems, twigs, leaves, flowers, buds, roots, and seeds) either intercellularly, in the vascular system, or as epiphytes (Rudolph 1993; Stall et al. 1993; Jacques et al. 2016). Therefore the *X. campestris* and *X. axonopodis* avocado pathovars can be associated with avocado budwood.

Symptoms of bacterial canker caused by the *X. campestris* avocado pathovar can develop in a matter of weeks (Cooksey and Azad 1992), and symptoms of bacterial leaf spot caused by the *X. axonopodis* avocado pathovar can develop in a few days (Pruvost et al. 2009) under experimental conditions. However, it is possible that asymptomatic infections of both pathovars also exist because asymptomatic infections (either latent infections or epiphytic growth) have been reported from other xanthomonads. For example, experimentally infecting tissue cultured *Anthurium andraeanum* (flamingo flower) with *X. axonopodis* pv. *dieffenbachiae* (the causal agent of anthurium blight) caused latent infection in 1-7% of calli (the undifferentiated cell masses formed from the source tissue), with no symptoms or turbidity of the medium for over three months. The pathogen was also able to survive in a small percentage of shoots without producing symptoms for over a year (Norman and Alvarez 1994). Although latent infections by *Xanthomonas* have been reported, it is likely that symptoms would be expressed by most infected plants during the PEQ period.

Asymptomatic survival of xanthomonads as epiphytes for long periods is also documented. For example, *X. phaseoli* pv. *manihotis* (the causal agent of bacterial blight of cassava) can survive asymptomatically as an epiphyte on cassava leaves in the field for eight months during the dry season, only causing symptoms once the rainy season has begun (Stall et al. 1993). In addition, some xanthomonads may survive in the field, without causing symptoms, for several months in plant structures. For example, experimental contamination of field-grown peach trees with *X. arboricola* pv. *pruni* (causal agent of bacterial spot of stone fruits and almonds) showed that the pathogen was able to survive in buds and leaf scars for 6-7 months without causing symptoms (Zaccardelli et al. 1998).

It is not known how long the *X. campestris* and *X. axonopodis* avocado pathovars could survive as epiphytes on avocado budwood in PEQ without causing symptoms. Depending on the temperature and relative humidity within PEQ, and in the absence of overhead irrigation to simulate rainfall, the appropriate environmental conditions for symptom development may be absent throughout the entire PEQ period. The routine surface disinfection of budwood entering PEQ (dipping in 1% sodium hypochlorite for 2 minutes) will reduce the surface population of the bacterium but is not likely to completely eliminate it, especially if cells are found deep within crevices of buds. Therefore, it is possible that a small population of epiphytic bacteria may survive asymptomatically throughout the PEQ period and enter New Zealand.

The bud grafting process may introduce epiphytic bacteria into the rootstock, making symptom expression during PEQ likely. However, it is also possible for a small number of cells to be present on only the tip of a bud and not at the cutting site, which would leave the epiphytic population undisturbed without introducing bacteria into the plant. Again, this could result in a small population of epiphytic bacteria surviving asymptomatically throughout the PEQ period and entering New Zealand. Given that xanthomonads are highly competent in causing infection (Rudolph 1993; Stall et al. 1993), even a small population may be enough for establishment.

Given the arguments and evidence above, that is:

- the *X. campestris* and *X. axonopodis* avocado pathovars could survive as epiphytes on avocado budwood in PEQ without causing symptoms for the PEQ period (minimum of 12 months) (Rudolph 1993; Stall et al. 1993; Jacques et al. 2016), but
- in the case of internal infection symptoms are more likely to appear within a matter of days or weeks (Cooksey and Azad 1992; Pruvost et al. 2009)

the likelihood of the *X. campestris* and *X. axonopodis* avocado pathovars entering New Zealand associated with *P. americana* budwood is considered to be very low. Given that there are only three peer-reviewed publications and one extension document about these avocado pathogens, the uncertainty of this conclusion is high.

4.2.5 Likelihood of exposure

This assessment is made on the basis that the *X. campestris* and *X. axonopodis* avocado pathovars have entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for both the *X. campestris* and *X. axonopodis* avocado pathovars.

If avocado plants are infected with either the *X. campestris* or *X. axonopodis* avocado pathovar at the time of their release from PEQ and they are planted in New Zealand or used to propagate new plants for planting (see section 2.3.1 for additional information about exposure) then these pathovars will be transferred to a suitable host multiple times. Promoting the growth and survival of hosts promotes the growth and survival of associated pathogens.

Given the arguments and evidence above, the likelihood of exposure of the *X. campestris* and *X. axonopodis* avocado pathovars in New Zealand from *P. americana* budwood is considered to be high, with low uncertainty.

4.2.6 Likelihood of establishment

This assessment is made on the basis that the *X. campestris* and *X. axonopodis* avocado pathovars have been successfully exposed to a suitable host plant in the New Zealand environment.

If the host range of the two avocado pathovars is only *P. americana* then hosts in the form of cultivated avocados will be available for their establishment in the North Island (see section 2.3). If the host range includes other members of the family Lauraceae then the number of potential host species available for the establishment of these pathovars is greatly increased – there are five native and four introduced species (in addition to *P. americana*) from the Lauraceae family found in New Zealand (NZPCN 2021).

The *X. campestris* avocado pathovar is reported from San Diego, Orange, Los Angeles, Ventura and Santa Barbara counties of southern California (Cooksey et al. 1993). The avocado growing regions in these counties (California Avocado Commission 2020) have a similar climate to parts of New Zealand, including the Auckland and Northland regions, as indicated by a climate match index of ≥0.7 (Phillips et al. 2018) when these locations are compared with all of New Zealand. Seasonal climate ellipses show considerable overlap between avocado growing areas in California and New Zealand, although Californian sites have warmer winters, are hotter in summer and drier overall (this is compensated for by irrigation) (Halloy 2021). Therefore, climate suitable for the establishment of the *X. campestris* avocado pathovar is likely to occur in at least some parts of New Zealand, especially in the north of the North Island. In addition, because wetter conditions favour the establishment of xanthomonads (Stall et al. 1993), and conditions in New Zealand are wetter compared to those of southern California (Beck et al. 2018), New Zealand conditions are likely to be more favourable for the establishment of the *X. campestris* avocado pathovar. In addition, there are records of other *X. axonopodis* pathovars being present in New Zealand (e.g. *X. axonopodis* pv. *begonia*, *X. axonopodis* pv. *glycines*, *X. axonopodis* pv. *poinsetticola*) (Biota NZ 2022) further suggesting New Zealand has a favourable climate for these species.

The *X. axonopodis* avocado pathovar is reported only from the Seychelles (Pruvost et al. 2009). No climate match data are available for the Seychelles but the climate is generally warmer and wetter than that of New Zealand (World Bank Group 2021). The suitability of the New Zealand environment for establishment of this pathovar cannot be definitively ruled out because remote island distributions may not be reflective of the fundamental niche of a species but rather of geographic isolation.

Based on the currently available information, the *X. campestris* avocado pathovar has not spread beyond southern California. Rather than indicating a lack of invasive potential, this could be related to the fact that avocado in the USA is only grown over a small area (213 Km^2) in California and Florida (Ag Marketing Resource Center 2018) and that international trade in avocado nursery stock from the USA is likely to be of a high phytosanitary standard. Similarly, although the *X. axonopodis* avocado pathovar is not reported to have spread outside the Seychelles, this is most likely due to the extremely low volume of plant and plant product exports from the Seychelles (International Trade Centre 2021).

Several other xanthomonads are well known to be invasive outside their native range, including *X. citri* pv. *citri* (citrus canker), *X. vasicola* pv. *vasculorum* (bacterial leaf streak of corn) and *X. translucens* pv. *translucens* (bacterial leaf streak of barley) (Triplett and Patel 2020; CABI 2021d).

Given the arguments and evidence above, that is:

- hosts (in the form of cultivated avocados or, potentially native and introduced members of the Lauraceae family) are available for the establishment of the *X. campestris* and *X. axonopodis* avocado pathovars (NZPCN 2021),
- suitable climate is available for the establishment of the of the *X. campestris* avocado pathovar (Cooksey et al. 1993; Phillips et al. 2018; California Avocado Commission 2020),
- the wetter conditions of New Zealand compared to southern California (Beck et al. 2018) are likely to favour establishment of the *X. campestris* avocado pathovar, and
- the suitability of the New Zealand environment for establishment of the *X. axonopodis* pathovar cannot be definitively ruled out

the likelihood of the *X. campestris* and *X. axonopodis* avocado pathovars establishing in New Zealand is considered high, with moderate uncertainty (because of the lack of information specific to these pathovars).

4.2.7 Impacts in New Zealand

In California bacterial canker caused by the *X. campestris* avocado pathovar is considered to be an unimportant though widespread disease. Because wet conditions favour the penetration of xanthomonads into plants (Stall et al. 1993), and because New Zealand has a wetter climate than southern California (Beck et al. 2018) it is possible that the impacts of the *X. campestris* avocado pathovar would be more severe here than in California. The relatively high wind speeds across New Zealand[30] will also favour longer distance spread of the pathogen.

No major outbreak of bacterial leaf spot of avocado caused by the *X. axonopodis* pathovar has been reported in the Seychelles archipelago since 2003 (Pruvost et al. 2009). No further information on the importance of this disease in the Seychelles is available. Although the Seychelles have a wetter climate than New Zealand (World Bank Group 2021), New Zealand has higher wind speeds[30] which could result in more widespread impacts of the *X. axonopodis* avocado pathovar should it establish here.

There is no cure known for infections by any species or pathovar of *Xanthomonas* and control is usually achieved by use of less susceptible varieties, use of windbreaks, disinfection of tools and equipment, application of copper-based treatments, and removal of infected trees. However, none of these methods are fully effective (Vítor Rodrigues et al. 2019) and control of outbreaks can involve wide-scale removal and destruction of host plants (Gottwald 2000). The main in-field management for bacterial canker caused by the *X. campestris* avocado pathovar in California is keeping trees in good condition and removing severely diseased trees (Eskalen and Faber 2016).

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[30] Information obtained from the “Global Wind Atlas 3.0, a free, web-based application developed, owned and operated by the Technical University of Denmark (DTU). The Global Wind Atlas 3.0 is released in partnership with the World Bank Group, utilizing data provided by Vortex, using funding provided by the Energy Sector Management Assistance Program (ESMAP). For additional information: https://globalwindatlas.info”

Economic impacts

Because it is likely that avocados are the only commercially produced hosts of the *X. campestris* and *X. axonopodis* avocado pathovars in New Zealand, it is likely that economic impacts will be limited to the avocado industry. In 2020, the export values for avocado was $112.3 million and the domestic value was $50.6 million (Plant & Food Research 2020). Infection with a xanthomonad may impact yields, as is the case with infection by other xanthomonads, although yield losses vary widely between pathovars. For example, *X. arboricola* pv. *corylina* (hazelnut blight) can cause up to 10% mortality and yield losses of 1-10% in hazelnuts (CABI 2021a), whereas *X. a.* pv. *juglandis* (walnut blight) can cause up to 100% crop loss of walnuts if not controlled (CABI 2021b). Infection may also impact marketability, as is the case with plums infected by *X. a.* pv. *pruni* (bacterial canker of stone fruit) which are reduced in size and often unmarketable (CABI 2021c).

In California, trees severely affected by the *X. campestris* avocado pathovar are reported to have low yields on one branch or the whole tree (Eskalen and Faber 2016) although yield loss estimates are not provided. There is no information on the impact of the *X. axonopodis* avocado pathovar but even if this pathogen were confined to only leaf mesophyll, it could affect yields or marketability of fruits due to a decrease in the plant’s photosynthetic area.

Given that there is no cure for infection by any species or pathovar of *Xanthomonas* (Vítor Rodrigues et al. 2019) and that control of an outbreak may involve wide scale removal and destruction of host plants (Gottwald 2000), the impacts of trying to control an outbreak of the *X. campestris* or *X. axonopodis* avocado pathovars may be severe for the avocado industry. Eradication may not be possible, as has been the case with *X. citri* subsp. *citri* (citrus canker) in the USA (United States Department of Agriculture 2020).

If there were an outbreak of the *X. campestris* or *X. axonopodis* avocado pathovar in one of the major avocado-producing regions in New Zealand, it is likely that the outbreak could be contained to that region by limiting human-assisted dispersal of the pathogen. Spread within a region could, however, be quite rapid, especially if there was a storm during the outbreak.

Given the arguments and evidence above, that is:

- avocados are the only hosts of the *X. campestris* and *X. axonopodis* avocado pathovars commercially produced in New Zealand,
- avocado in an economically important domestic and export crop in New Zealand and infection of avocado trees with the *X. campestris* or *X. axonopodis* avocado pathovar could cause yield losses, and control of an outbreak could involve wide scale destruction of avocado trees (e.g. removal of all trees in one major avocado production area).

the economic impact of the *X. campestris* and *X. axonopodis* avocado pathovars to New Zealand is considered to be moderate, with high uncertainty (because there are no specific data on losses caused by these pathovars and impacts of xanthomonads vary widely).

Environmental impacts

Although the host range is likely to be restricted to *Persea americana*, the host range of the *X. campestris* and *X. axonopodis* avocado pathovars may include other species of the family Lauraceae, which may include the five native Lauraceae species found in New Zealand (NZPCN 2021). There are no reports of *Xanthomonas* causing significant disease in wild plants or natural environments (google scholar search terms [Xanthomonas AND disease and "wild plants"] and [Xanthomonas AND disease and "natural environments"]). However, a response to an outbreak of the *X. campestris* or *X. axonopodis* avocado pathovar may involve the removal of potential alternative hosts in the outbreak area. This was the case with the 2004 citrus canker outbreak in Australia, which involved the removal and destruction of 175,000 native citrus trees (*Citrus glauca*) (Senate Standing Committee on Rural and Regional Affairs and Transport 2006). This sort of environmental impact is, however, likely to be localised and short-lived.

Given the arguments and evidence above, the impact on the environment from the establishment of *X. campestris* and *X. axonopodis* avocado pathovars in New Zealand is considered to be very low, with high uncertainty.

Human health impacts

There are no known human health impacts associated with either the *X. campestris* or *X. axonopodis* avocado pathovars.

Sociocultural impacts

A response to an outbreak of the *X. campestris* or *X. axonopodis* avocado pathovars may involve the removal of infected or exposed avocado trees growing on residential properties. This was the case with the attempted eradication of citrus canker from Florida in the 1990s (Schubert et al. 2001). This is likely to cause distress to affected residents, however, this impact is likely to be localised and short-lived. In addition, if not eradicated, the pathogen may affect the appearance of trees and yield in home gardens.

Given the arguments and evidence above, the sociocultural impact of *X. campestris* and *X. axonopodis* avocado pathovars to New Zealand is considered to be very low, with high uncertainty.

Overall impact to New Zealand

The overall impact of the *X. campestris* and *X. axonopodis* avocado pathovars on the New Zealand economy, environment, health and society is considered to be moderate, with high uncertainty.

4.2.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is very low
- assuming entry, the likelihood of exposure is high
- assuming successful exposure, the likelihood of establishment is high
- the overall impact on the New Zealand economy, environment, health and society is considered to be moderate.

the overall level of assessed risk to New Zealand from the *X. campestris* and *X. axonopodis* avocado pathovars on imported *P. americana* budwood (as in the commodity description) is considered to be moderate, with high uncertainty.

4.2.9 Management considerations

There is no information on what sort of environmental conditions are likely to induce symptom expression in either The *Xanthomonas campestris* or *X. axonopodis* avocado pathovar. Although symptom expression of other xanthomonads is affected by factors such as temperature and humidity, optimum levels of these variables for symptom expression differ between pathovars (Stall et al. 1993) so the information cannot be generalised to the avocado pathovars being assessed here.

A large number of tests for the detection and identification of various *Xanthomonas* pathovars in both symptomatic and asymptomatic tissue have been developed (Catara et al. 2021). These include:

- serological methods such as: immunofluorescence; and enzyme-linked immunosorbent assay (ELISA),

- molecular methods such as: conventional and quantitative real-time PCR (qPCR); sequencing; DNA-DNA hybridisation; fingerprinting; and Clustered Regularly Interspaced Short Palindromic Repeat (CRISPR) genotyping, and

- other methods such as bioassays and biochemical tests.

These tests have been developed for the detection and identification of particular species or pathovars of *Xanthomonas* and not as generic tests. Generic PCR tests for the identification of members of the genus *Xanthomonas* have been developed (Adriko et al. 2014) but they have not been validated for either of the two pathovars being assessed here. In addition, validation (through sequencing) cannot occur as there are no reference cultures in any international culture collections.

Sequencing of the DNA gyrase subunit B (*gyrB*) gene is a useful identification tool which has been shown to work for a large number of *Xanthomonas* species (Parkinson et al. 2009). It is likely that this test could be used for a species level identification of *Xanthomonas* present in avocado budwood. In the absence of reference sequence data for the *X. campestris* or *X. axonopodis* avocado pathovar this test would not be able to provide a definitive pathovar identification. Furthermore, these two avocado pathovars have not been formally described and there are no reference cultures available. Therefore, if a *X. campestris* or *X. axonopodis* is isolated, there would be an inability to determine if these pathovars are the same as described in California (*X. campestris*) or the Seychelles (*X. axonopodis*).

There is no information on the best sampling regime (in terms of plant part or season) to identify either of these pathovars from asymptomatic tissues. However, generally spring and early summer are the best seasons for detecting bacterial pathogens including other xanthomonads (R. Taylor, pers. comm.; An et al. 2019). It is possible to have misting in PEQ to provide conducive conditions for other bacterial pathogens such as *Xylella* and *Pseudomonas* (R. Taylor, pers. comm.).

4.2.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

The *Xanthomonas campestris* avocado pathovar causes lesions on trunks and branches which develop into cankers (usually 2-10 cm in diameter) that split and ooze fluid that dries to a powdery white residue (Cooksey et al. 1993; Eskalen and Faber 2016). Lesions on budwood or young plants could be much smaller than this. The *X. axonopodis* avocado pathovar causes small, black, water-soaked, angular leaf lesions on avocados which sometimes coalesce into larger lesions (Pruvost et al. 2009).

- ***What are the known environmental conditions conducive to symptom expression?***

There is no information on what sort of temperature and humidity levels are likely to induce symptom expression in either the *X. campestris* or *X. axonopodis* avocado pathovar. Although symptom expression of other xanthomonads is affected by factors such as temperature and humidity, optimum levels of these variables for symptom expression differ between species and pathovars (Stall et al. 1993). For example, optimum *in vitro* growth temperatures can range from 20–23˚C for cool weather adapted *Xanthomonas* such as *X. populi* (Ridé and Ridé 1992) to 30 ˚C for warm weather adapted types like *X. citri* subsp. *citri* (Stall et al. 1993). Development of walnut blight caused by *X. arboricola* pv. *juglandis*, on the other hand, appears to not be influenced by temperature (CABI 2021b). Given the range of temperature and humidity optima for species and strains of *Xanthomonas* the information cannot be generalised to the avocado pathovars being assessed here. The use of overhead irrigation to simulate rainfall could induce symptom expression of epiphytic *Xanthomonas* populations, however, it would increase the risk of the disease spreading between plants.

What are the limitations to taking samples for potential testing?

- ***What* plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?**

There is no information on which tissues are colonised by *X. campestris*. Leaf tissue would be tested for leaf spotting pathogens (e.g. *X. axonopodis*) which would also detect epiphytic populations (R. Taylor, pers. comm.).

- ***What is the optimum season for conducting detection testing?***

There is no specific information on the optimum season for conducting detection testing to maximise the detection of either the *X. campestris* or *X. axonopodis* avocado pathovar in the case of asymptomatic infection. However, generally spring and early summer are the best seasons for detecting bacterial pathogens (R. Taylor, pers. comm.; An et al. 2019). It is possible to have misting in PEQ to provide conducive conditions for other bacterial pathogens such as *Xylella* and *Pseudomonas* (R. Taylor, pers. comm.).

4.2.11 References

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4.3 *Xylella fastidiosa* (bacterial leaf scorch)

Xylella fastidiosa is a fastidious xylem-limited bacterium (i.e. it has specialised nutrient needs and can be difficult to culture). It can colonise a very broad range of plants, including economically important crops and environmentally significant plants. It is a very serious pathogen of some of these plants, blocking water transport and causing wilting and death. Natural transmission is via specialist xylem-feeding insects.

4.3.1 Taxonomic description

Scientific name: *Xylella fastidiosa* Wells et al., 1987

Order/Family: Xanthomonadales/Xanthomonadaceae

Other names: There are no other scientific names. Common names for the diseases caused by this bacterium include Anaheim disease, California vine disease, citrus variegated chlorosis, leaf scorch of almond/ coffee/ oleander, olive quick decline syndrome, peach phony disease, Pierce's disease, plum leaf scald and dwarf disease of lucerne/ alfalfa (EPPO 2021).

Taxonomic notes: *Xylella fastidiosa* is a genetically diverse species that has been associated with a wide range of plant diseases. The American representatives of the species were initially divided into three subspecies[31] based on DNA-DNA hybridization data:

- *Xylella fastidiosa* subsp. *fastidiosa,* the cause of Pierce's disease of grapevines (among other diseases) (Vanhove et al. 2019);
- *Xylella fastidiosa* subsp. *multiplex*, the causal agent of several diseases in almonds, oaks, summerfruit and shade trees (Landa et al. 2020; Johnson et al. 2021); and
- *Xylella fastidiosa* subsp. *pauca*, which causes disease in citrus and coffee; and which has recently been found to cause olive quick decline syndrome in Italy (Vanhove et al. 2019).

Multilocus sequence typing (MLST)[32] sequence data has confirmed the status of these subspecies, and suggested a fourth, *X. fastidiosa* subsp. *sandyi*. Two additional subspecies, *X. fastidiosa* subsp. *morus* and *X. fastidiosa* subsp. *tashke* have also been proposed (Almeida and Nunney 2015; Sicard et al. 2018).

4.3.2 Hazard identification

Xylella fastidiosa is not known to occur in New Zealand in any of its forms

- The species is reported absent in Biota NZ (2022) and is not recorded in PPIN (2021).
- It and its subspecies are regulated pests and unwanted and notifiable organisms (ONZPR 2021).
- New Zealand has country freedom status for *X. fastidiosa* (ONZPR 2021). Country freedom in this case is based on historical freedom; no systematic surveys have been carried out to determine the presence of this bacterium in New Zealand. The possibility that it is present in one or more of its forms cannot be discounted. For example, *X. fastidiosa* is likely to have

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[31] Below the subspecies level, the further MLST classification of isolates into sequence types (unique genotypes based on the seven loci used in MLST) has provided insights about *X. fastidiosa* evolution and host specificity (Almeida and Nunney 2015). However, Landa et al. (2020) note that while MLST is particularly powerful for the taxonomy of recombinogenic taxa (*X. fastidiosa* is naturally competent), it is also possible that strains belonging to the same ST may not be phylogenetically related or share similar traits such as host plant range.

[32] MLST is a method that categorizes isolates into sequence types (STs) based on the alleles present at (usually) seven house-keeping loci (Baldi and La Porta 2017). Assessment of genetic diversity in *X. fastidiosa* is still principally based on MLST even though the number of available draft and complete genomes of *X. fastidiosa* is rapidly increasing (Saponari et al. 2019).

been in France for at least 15 years –and possibly much longer– before its first detection there (Soubeyrand et al. 2018).

Xylella fastidiosa has the potential to establish and spread in New Zealand:

- Suitable hosts (including avocado plants) are available in many areas of New Zealand. *Xylella fastidiosa* (at the species level) is able to colonise a very wide range of plant species. The current EFSA database (Delbianco et al. 2021) includes over 600 plant species reported to be infected by the bacterium. These species include important crop, ornamental and native plant species, many of which are common throughout New Zealand.
- However, the climate may limit the development and spread of diseases caused by *X. fastidiosa*. The optimal temperature range for its growth is 26–28℃ (Baldi and La Porta 2017). Diseases caused by this bacterium are more common in tropical and subtropical regions (Janse and Obradovic 2010), and its distribution, spread and/or expression may be limited in some parts of New Zealand. However, CLIMEX predictions by MPI (2020) indicated that all of the North Island is highly suitable for the establishment of *X. fastidiosa*.

Xylella fastidiosa has the potential to cause harm to New Zealand because:

- It can cause severe plant pathologies leading to huge economic losses. Some examples are Pierce's disease of grapevines, olive quick decline, oak bacterial leaf scorch, phony peach disease, citrus variegated chlorosis and almond leaf scorch (EPPO 2021). Some of these hosts, such as grapes, summerfruit and citrus, are of high economic importance to New Zealand. In addition, sentinel host plant testing overseas has shown that *X. fastidiosa* is able to infect iconic native plant species such as Kauri and Pohutukawa (Groenteman et al. 2015).
- *Xylella fastidiosa* has been reported to cause disease in avocado, an important commercial crop in New Zealand. In 2020, export sales of fresh avocados earned NZ$100.1 million and domestic sales earned NZ$50.6 million, while export sales of avocado oil earned $NZ9.3 million (Plant & Food Research 2020).

Xylella fastidiosa is associated with avocado budwood because:

- Avocado has been reported as a host of *X. fastidiosa* (CAC and CMCC 2003; Montero-Astúa et al. 2008);
- *Xylella fastidiosa* infections are systemic. The bacterium exists within the xylem tissues of its host plants (Almeida and Nunney 2015) and can therefore be associated with leafless, rootless avocado budwood cuttings.

Given the arguments and evidence above, *X. fastidiosa* meets the criteria to be a hazard on avocado budwood (as in the commodity description) imported to New Zealand.

4.3.3 Risk assessment

Biology and epidemiology of Xylella fastidiosa

Host range

At the species level, *X. fastidiosa* has a very wide "host range". The current EFSA database (Delbianco et al. 2021) includes over 600 plant species reported to be colonised by the bacterium. The concept of *X. fastidiosa* as a generalist pathogen with a wide host range is accurate in the sense that a very large number of plant species have been demonstrated to sustain infections. However, very few of these plants sustain the infections long-term and become symptomatic. Mounting evidence suggests that specific clades of *X. fastidiosa* cause disease in a limited number of plant species, while still being capable of colonizing many other species as harmless endophytes. Specific symptomatic hosts are only susceptible to isolates in one or a limited number of *X. fastidiosa* clades, with the result that specific clades (at the subspecies level and lower) of *X. fastidiosa* have a small number of symptomatic host plant species that are largely non-overlapping (Yuan et al. 2010; Almeida and Nunney 2015; Sicard et al. 2018; Nunney et al. 2019).

Association with avocado

The only published record of avocado as a host of *X. fastidiosa* is from Costa Rica (Montero-Astúa et al. 2008). Symptomatic trees were found in the field in at least two distinct geographical locations (Alajuela and San José provinces) during the 1990s. Culture attempts were not successful[33] but *Xylella fastidiosa* infection was detected using DAS-ELISA[34] (in 188 of 227 trees tested) and PCR (from two grafted seedlings and 7 field trees). The PCR method followed Pooler and Hartung (1995). The specificity of this test has been validated over a number of years and it is still listed in the IPPC diagnostic protocol for *Xylella fastidiosa* (FAO 2018). Seedlings that were grafted with ELISA-positive budwood developed scorch symptoms. GenBank sequences EU021997 to EU022000 showed 99 to 100% sequence identity to a Pierce's disease strain from California (Temecula1) and 94 to 95% to a citrus variegated chlorosis strain from Brazil (Found-5). EFSA (2015) report this record, citing *X. fastidiosa* subsp. *fastidiosa* as the “putative subspecies”, while Delbianco et al. (2021) reports “subspecies unknown”. In North America, *X. fastidiosa* subsp. *fastidiosa* is primarily associated with Pierce’s disease of grapevine but it has a number of other hosts, notably almond (Nunney et al. 2019).

In California, tests for *X. fastidiosa* were reported to “come up positive on avocado in some areas” by the California Avocado Commission (CAC and CMCC 2003). Test methods were not reported and no identification below species level was provided. No direct symptoms were identified or linked to positive *Xylella* test results in these detections.

No other reports of *X. fastidiosa* infecting avocado have been found in searches of the scientific literature, databases (including the most current EFSA host plant database (Delbianco et al. 2021)), Google and Google Scholar searches or in the MPI Emerging Risk System (all searches carried out in English language in December 2021).

Geographical distribution

Xylella fastidiosa is native to the Americas, where it exists as a number of subspecies, strains and serotypes. Sampling and MLST analysis have indicated that the component subspecies evolved in geographical isolation and have distinct geographical and host ranges (Almeida and Nunney 2015; Sicard et al. 2018; Landa et al. 2020).

The first observed detection of *X. fastidiosa* outside the Americas was from Taiwan in 2002 (Su et al. 2013). Since that time, it has spread to Italy (Denancé et al. 2017; Cunty et al. 2020); France (Denancé et al. 2017); Germany (eradicated, Jeger et al. 2018); Spain (Cunty et al. 2020) and Portugal (EPPO 2021).

For regulatory purposes, the following countries are not recognised by MPI as free from *X. fastidiosa*: all countries in Europe, the Americas and the Caribbean; India, Taiwan and Iran[35] (as at 22 January 2022).

Symptoms in avocado

Field infections were observed in avocado trees in at least two localities in Costa Rica. Symptoms were reported to include chlorotic mottling, marginal scorch, deformation of leaves, defoliation, shortening of internodes, and branch dieback. These symptoms were not uniformly distributed in the tree, and the results of ELISA tests of individual trees also varied with the season and branches tested (Montero-Astúa et al. 2008).

Avocado is reported to be asymptomatically infected with *X. fastidiosa*:

- In Costa Rica, symptoms in infected trees were not uniformly distributed in the tree; some branches were symptomatic while others were not (Montero-Astúa et al. 2008).

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[33] However, it is known that *Xylella* cannot be cultured from all hosts (Rapicavoli et al. 2018)
[34] ELISA tests have been known to generate false positives (R. Taylor, pers.comm.).
[35] https://www.mpi.govt.nz/dmsdocument/1152/direct, https://www.mpi.govt.nz/dmsdocument/15655

- In California, no symptoms were associated with *X. fastidiosa* infections (CAC and CMCC 2003).

Reproduction and transmission

Xylella fastidiosa colonises the xylem network of plants, where it can move up- and downstream. In susceptible hosts, where the bacteria are able to multiply, they can form high populations within the plant, leading to high frequencies of blocked xylem vessels, which are usually correlated with disease symptom development and severity (EFSA 2015).

Natural spread of the bacterium is through transmission by its insect vectors, specialist xylem-feeding hemipterans (mostly sharpshooters and spittlebugs; Almeida and Nunney 2015). Invasion of new regions is accomplished only by long-distance dispersal of infected vectors or by the movement of infected plant material (Almeida and Nunney 2015). The bacterium is graft transmitted. Montero-Astúa et al. (2008) demonstrated graft transmission in avocado. Fifteen greenhouse-grown, ELISA-negative avocado seedlings were grafted with budwood from an ELISA-positive tree. Eight of the 15 seedlings developed scorch symptoms and one also showed chlorotic mottling and deformation, though the time to development of these symptoms was not reported. In two of the grafted seedlings, positive PCR results were obtained. *Xylella fastidiosa* is not known to be seed transmitted (EFSA 2015; Parkinson and Malumphy 2020).

Vectors

Known or potential vectors are currently considered to include (and to be limited to) three groups of hemipterans:

- all sharpshooters (species in the subfamily Cicadellinae, superfamily Membracoidea);
- all species in the superfamily Cercopoidea (spittlebugs and or froghoppers), and
- all cicada species (superfamily Cicadoidea) (EFSA 2013, 2015; Cornara et al. 2019).

Sharpshooters are not known to be present in New Zealand, so potential vectors here are limited to spittlebugs and cicadas:

- Spittlebugs: within the superfamily Cercopoidea, only the family Aphrophridae[36] is known to be present in New Zealand, with a fauna comprised of 16 species. The only known vector is the introduced European meadow spittlebug *Philaenus spumarius*, which is widespread and has a broad host range. Fifteen other species are known, 14 of which are native (Lariviere et al. 2010).
- Over 30 cicada species are known, all are endemic (Lariviere et al. 2010). However, there is very little evidence of *X. fastidiosa* transmission by cicadas (Sicard et al. 2018; Cornara et al. 2019).

There is no evidence of *X. fastidiosa* genotype-vector specificity, i.e., all insect vectors are thought to be capable of transmitting all *X. fastidiosa* genotypes. This lack of specificity increases the likelihood that *X. fastidiosa* isolates introduced into a novel environment will be transmitted by endemic vector species (Almeida and Nunney 2015; Sicard et al. 2018). However, transmission efficiency is highly variable among vectors and their hosts. It may differ for different vector species on the same host plant, or the same vector species feeding on different tissues of the same plant (Almeida and Nunney 2015). Thus, the impact of an introduction will depend on the interaction of which subspecies of pathogen is introduced, the plants susceptible to this strain, and the vectors which use the plant as hosts.

Asymptomatically infected plants are likely to harbour lower bacterial populations, and this is likely to affect acquisition by the vector. Plants that harbour larger bacterial populations distributed throughout the plant are more likely to result in vector infection than plants with low bacterial populations, which usually do not become systemic or induce severe disease symptoms (EFSA 2015). However

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[36] Sometimes regarded as Aphrophorinae, a subfamily of the Cercopidae (e.g. by ONZPR 2021). This classification follows Lariviere et al. (2010), EPPO (2021) and the Arthemis database (INRAE-CBGP 2020).

asymptomatic plants are potentially infectious. Their importance appears to vary among pathosystems, for example, they appear to be important for Pierce's disease in Northern California but not for citrus variegated chlorosis (CVC) in Brazil (Sicard et al. 2018; White et al. 2020).

Time to symptom development

There is no information available regarding the time to symptom development for *X. fastidiosa* in avocado plants. Montero-Astúa et al. (2008) demonstrated graft transmission in avocado but did not report time to symptom development. In the only other report of *X. fastidiosa* infecting avocado (CAC and CMCC 2003), infections were reported to be asymptomatic.

In other plant species colonised by *X. fastidiosa*, the timing and extent of symptom development vary greatly (EPPO 2018). In some hosts, the plant or tree can be killed rapidly. Some plants may remain asymptomatic for varying periods (latent period) before the bacterium begins to replicate within the host and colonizes large parts of the plant xylem. Variation in the asymptomatic period is significant for different host and pathogen subspecies combinations, for example from a median of up to 1 month in ornamental plants and up to 10 months in olive, for subsp. pauca (Bragard et al. 2019) or several years for other *Xylella* subspecies in other plants (Parkinson and Malumphy 2020). It is clear that *X. fastidiosa* has one of the longest symptomless periods amongst some of the most dangerous plant diseases (White et al. 2020). Additionally, New Zealand's relatively cool temperatures could result in latent periods becoming even longer (Parkinson and Malumphy 2020 for the UK).

However, under natural conditions, many *X. fastidiosa* hosts are always asymptomatic (EFSA 2013; Sicard et al. 2018). In these hosts, the bacterium does not usually move systemically within the plant and vectors are less likely, but still able, to acquire it (Hopkins and Purcell 2002; Parkinson and Malumphy 2014; Sicard et al. 2018).

4.3.4 Likelihood of entry

This assessment is made on the basis that *X. fastidiosa* is present in the countries from which avocado budwood is being imported. However, there is a high level of uncertainty associated with assessing the likelihood of entry. *Xylella fastidiosa* infection of avocado has currently only been reported once in the published scientific literature, from Costa Rica. There is also an unpublished report from California. These infections therefore appear to be very rare. However, *X. fastidiosa* infections can be asymptomatic, or can cause symptoms that may be mistaken for other conditions, such as physiological stress. It is possible that in Costa Rica and California, *X. fastidiosa* infections of avocado are more widespread than are reported. Similarly, it is possible that in other countries where *X. fastidiosa* is present, the bacterium may colonise avocado without having yet been reported.

Xylella fastidiosa infections are systemic. The bacterium exists within the xylem tissues of its host plants and may therefore be associated with leafless, rootless avocado budwood cuttings, and would not be affected by surface sterilisation of budwood cuttings in dilute sodium hypochlorite.

It is unlikely that symptomatic avocado budwood material would be used for germplasm. However, infections are reported to be asymptomatic in avocado. They may also have long latent periods, as is reported in other hosts (the time to symptom development in avocado is not reported). For these reasons, infected material may not be detected during preparation for export or in pre-export phytosanitary inspections or on arrival in New Zealand.

As part of the commodity description, all imported avocado germplasm, will undergo a growing period of active growth with a minimum of three distinct phenological growing events ('shoot flush') in 3A PEQ (minimum of one year). Since infections in many hosts are reported to be asymptomatic or may have very long latent periods, it is likely that the period spent in 3A PEQ is not sufficient to manage the risk of *X. fastidiosa* being present in imported avocado budwood that is released from PEQ.

It is likely that the subspecies reported from avocado in Costa Rica is *X. fastidiosa* subsp. *fastidiosa*, but otherwise it is unknown which bacterial genotypes can colonise avocado. Other genotypes may be more likely to colonise avocado as endophytes that do not produce symptoms. These genotypes still represent a risk to New Zealand because if asymptomatic plants are exposed to vectors in New Zealand it is still possible for the bacterium to be acquired and spread.

The bacterium also exists within the bodies of its insect vectors, so *X. fastidiosa* may also be introduced through the entry of viruliferous vectors, which are able to feed on and infect susceptible hosts. However, it is assumed that the mandatory pesticide treatment (Chapter 2) will be effective in managing the risk of introducing any live insect vectors or their eggs[37] that may be associated with imported avocado budwood. In addition, Level 3A PEQ has physical and operational measures in place to manage the risks associated with arthropods, wastewater, waste and accidental transfer, therefore insect vectors carrying *X. fastidiosa* are highly unlikely to escape level 3A PEQ to infect a suitable host.

Given the arguments and evidence above, that is:

- infection of avocado by *X. fastidiosa* appears to be very rare (it has only ever been documented once in the published literature); however
- infections of avocado and many other hosts are reported to be asymptomatic. Where infections are symptomatic the latent period is not known but may be long. Therefore established *X. fastidiosa* infections may not be detected before release from PEQ, and
- for the above reasons, *X. fastidiosa* infections of avocado may also be more common than is reported;

the likelihood of *X. fastidiosa* entering New Zealand (being released from PEQ[38]) associated with avocado budwood is considered to be **very low**, with **high** uncertainty.

4.3.5 Likelihood of exposure

This assessment is made on the basis that *X. fastidiosa* has entered New Zealand undetected.

When a plant pest or pathogen arrives in a new area, it usually needs to encounter a growing host plant in order to establish. The process of moving from or with the commodity into a suitable environment to allow establishment is termed exposure.

On some pathways, contaminating organisms need to actively or passively leave the import pathway (i.e., move from the pathways) in order to find a growing host plant. However imported nursery stock, in particular high value nursery stock, is itself intended to generate plants for planting in New Zealand. Plants released from PEQ will be grown with care, promoting the survival of both the plants and their exotic bacterial populations. Many plants may eventually be derived from the released material, so that multiple populations of the bacterium may also be present in New Zealand. This is particularly true in the case of pathogens such as *X. fastidiosa*, which can infect plants asymptomatically.

Successful exposure in this context is thus considered to mean that plants derived from imported avocado budwood infected with *X. fastidiosa* are growing successfully in the New Zealand environment[39], either as plants being multiplied in nurseries or planted in New Zealand avocado orchards. Avocado plants are perennial. The average range of productive life in California has been estimated at 20 to 30 years, with an upper estimate of 40 years under favourable conditions (Goodall et al. 1971), while trees growing in the wild in Mexico have been known to live as long as 400 years[40]. Infected trees may therefore remain present as sources of bacterial inoculum for many years, until their removal or death.

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[37] Note that *X. fastidiosa* is not passed from adult hemipterans to their eggs (Almeida and Nunney 2015), so the accidental importation of live egg masses via any pathway is not in itself capable of introducing the bacterium. However, such incursions may give rise to the establishment of competent exotic vector species.

[38] In the form of plants grown from imported budwood that has been grafted onto New Zealand sources avocado rootstock

[39] This definition may not necessarily apply in other situations.

[40] http://ucavo.ucr.edu/general/answers.html

Given the arguments and evidence above, the likelihood of exposure of *X. fastidiosa* in New Zealand from avocado budwood is considered to be high, with low uncertainty.

4.3.6 Likelihood of establishment

This assessment is made on the basis that *X. fastidiosa* has been successfully exposed to a suitable host plant/s in the New Zealand environment.

To establish a permanent or persistent population, the organism must generally be able to move to new hosts, and the environment must be favourable for its survival. Natural spread of *X. fastidiosa* is through transmission by its insect vectors. Unless a competent vector is present, it can only move to new hosts by human-mediated means such as grafting to other avocado plants. It is likely that imported budwood will be widely propagated, since it is of high value. It is uncertain to what degree this is likely to spread the bacterium to healthy rootstock. The only information available for avocado is from one small trial: Montero-Astúa et al. (2008) found that around 50% (8/15) of healthy seedlings developed symptoms after being grafted with infected budwood, and around 13% (2/15) tested positive for *X. fastidiosa* by PCR. It is possible that *X. fastidiosa* populations could persist in avocado orchards for an indefinite period even in the absence of a vector, since infections can be asymptomatic and trees are long-lived. Symptomatic trees are unlikely to be propagated from and may be removed, so that eventually infections that are transmitted solely by propagation to avocados may die out.

If competent vectors are present, the bacterium has the potential to spread naturally to other plants (including other, “native” avocado plants that are New Zealand grown), depending on the host range of the vector and the susceptibility of those plants to the bacterium. This means the host range of the bacterium is dependent on the host ranges of its vectors.

The establishment and spread of *X. fastidiosa* therefore depends on a combination of several factors: *i*) the presence of competent vectors, *ii*) the presence of suitable host plants and *iii*) the suitability of the environment.

Vectors

At least one known vector species, the introduced European meadow spittlebug *Philaenus spumarius*, is present in New Zealand. This spittlebug has an extremely wide host range and is the main vector of olive quick decline syndrome (*X. fastidiosa* subsp. *pauca*) in Italy (Cornara et al. 2017). It has been collected throughout much of New Zealand and from very many common crop, weed and amenity plants (Lariviere et al. 2010), including avocado (PPIN 2021). It therefore has the potential to feed on infected avocado plants (in nurseries or orchards) and to transmit the bacterium to other plants in the managed estate. It has also been collected on a number of native plants (see *Environmental Impacts* section below). This means this vector is also capable of moving the bacterium from the managed to the natural estate.

Other potential vectors are present in New Zealand. Fifteen additional spittlebug species have been reported: the introduced Australian *Bathyllus albicinctus* and 14 native species. Their host ranges include crop and amenity plants in addition to many native plants (Lariviere et al. 2010). Around 30 native cicada species are known, however the role of cicadas in *X. fastidiosa* transmission is unclear.

The bacterium therefore has the potential to spread naturally both within and between the managed and natural estates in New Zealand.

Host plants

A very large number of plant species (over 600) have been demonstrated to sustain *X. fastidiosa* infections, including many important crop, ornamental and native plants common throughout New Zealand.

Each bacterial subspecies is thought to have a largely non-overlapping set of symptomatic host plants (Yuan et al. 2010; Nunney et al. 2019), so the range of hosts that will be impacted upon in an incursion of any particular genotype is likely to be limited. For example, the putative subspecies associated with the Costa Rican disease of avocado is *X. fastidiosa* subsp*. fastidiosa*. In North America, this subspecies is primarily associated with grapevine (commonly grown in New Zealand), though it has a number of other hosts including almond (Nunney et al. 2019). Nevertheless, non-symptomatic hosts are colonised by the bacterium and they may act as reservoirs of infection (Sicard et al. 2018). Therefore, it is unlikely that the presence of suitable host plants will be a limiting factor to the establishment of the bacterium.

Suitability of the New Zealand environment

At the species level, *X. fastidiosa* survival is limited by cold temperatures. The optimal growing temperature of the pathogen is 26–28°C. It is usually limited to the tropical and subtropical areas of North and South America and in general diseases caused by the bacterium are not reported from areas with low winter temperatures or at high altitudes (Hopkins and Purcell 2002; Baldi and La Porta 2017; Godefroid et al. 2019).

However, the responses to climate for each subspecies may vary. Some subspecies have been found in much colder countries, such as Canada (Baker 2017). Baker (2017) suggested that *X. fastidiosa* subsp. *multiplex* provides the greatest likelihood of establishment in the United Kingdom, since none of the other subspecies have yet been found in temperate climates. For New Zealand, CLIMEX predictions by Hoddle (2004) indicated that cold stress would exclude *X. fastidiosa* (Pierce's disease) from most of New Zealand's wine production areas. However, CLIMEX predictions by MPI (2020) indicated that all of the North Island is highly suitable for the establishment of *X. fastidiosa*, while the South Island is overall less suitable.

There is a high level of uncertainty in assessing the impact of climate on the establishment of *X. fastidiosa*:

- It is not known which *X. fastidiosa* genotype could be introduced into New Zealand with avocado budwood. It is likely that the subspecies infecting avocado in Costa Rica is *X. fastidiosa* subsp. *fastidiosa*, in which case its distribution, host range, spread and/or expression may be limited in this country (Hoddle 2004). Symptomatic infections of avocado by *X. fastidiosa* have only been reported from two localities in Costa Rica: Alajuela province, with a CMI (Composite or Climate Matching Index) of 0.4 and San José province, with CMIs varying from 0.4 to 0.6 (match is with all of New Zealand; Phillips et al. 2018), meaning that the New Zealand climate may not be highly suitable for disease development, at least not in avocado. If other, yet unreported genotypes, are associated with avocado, particularly subsp. *multiplex*, distribution and spread are likely to be wider and/or expression more severe.
- *Xylella fastidiosa* is likely to be more widely distributed than current records suggest because it is difficult to identify on the basis of symptoms (Baker 2017).
- Due to the lack of data on impacts from areas with cooler summer temperatures, it is not known what the direct impact of *X. fastidiosa* may be in temperate regions such as the U.K. (Parkinson and Malumphy 2020) or New Zealand.
- The severity of *X. fastidiosa*-induced diseases has recently increased, possibly due to global warming (Godefroid et al. 2019).

Given the arguments and evidence above, that is:

- plants suitable for colonisation by *X. fastidiosa* are widely grown throughout New Zealand;
- some parts of New Zealand are likely to be suitable for the establishment and expression of diseases caused by *X. fastidiosa*;
- at least one competent vector species is present in New Zealand, it is widely distributed and has a broad host range which includes avocado as well as other economically important and environmentally significant plants;

the likelihood of *X. fastidiosa* establishing in New Zealand is considered to be moderate, with moderate uncertainty.

4.3.7 Impacts in New Zealand

Xylella fastidiosa is an obligatory plant and insect vector colonizer that can only spread by the movement of infected plant material or through the movement of its vectors. It is genetically diverse at the species level, and evidence indicates that specific clades cause disease in a small number of host plants that are largely non-overlapping, while colonising many other plant species as a harmless endophyte. The impact of any invasion will depend on the bacterial genotype that is introduced, the plants susceptible to this genotype and the vectors which feed on those plants. Abiotic variables such as climate are also important, and responses to climate by different genotypes may also be variable. Thus the epidemiology and dynamics of each pathosystem are distinct and depend on complex biological and ecological interactions between the plant, pathogen, and vector over short and long time periods (Yuan et al. 2010; Almeida and Nunney 2015; Baker 2017; Sicard et al. 2018; Nunney et al. 2019).

There is therefore a high level of uncertainty in assessing the potential impacts of an incursion or the establishment of *X. fastidiosa* in New Zealand.

Economic impacts

Xylella fastidiosa can cause severe plant pathologies such as Pierce's disease of grapevines, olive quick decline, oak bacterial leaf scorch, phony peach disease, citrus variegated chlorosis and almond leaf scorch. These can lead to huge economic losses, for example:

- costs incurred by the Pierce's disease strain (*X. fastidiosa* subsp. *fastidiosa*) on the grape industry in California alone are estimated to amount to hundreds of millions of dollars annually (Tumber et al. 2014);
- the CoDiRo strain of *X. fastidiosa* subsp. *pauca*, which is associated with olive quick decline syndrome (OQDS) in southern Italy, has killed millions of olive trees in Apulia, causing unprecedented socio-economic issues (Godefroid et al. 2019);
- Phony peach disease (PPD) has caused severe yield loss in Georgia and elsewhere in the southeastern United States, with millions of PPD-infected trees being removed from peach orchards over the last century. The disease remains a production constraint, and management options are few (Johnson et al. 2021).

Some of the bacterium's highly susceptible hosts, such as grapes, summerfruit and citrus, are of very high economic importance to New Zealand. Wine exports for example, were worth $NZ1.8 billion in 2019; domestic and export production of summerfruit and citrus were worth almost $NZ120 million and $NZ70 million respectively (Plant & Food Research 2019). The putative subspecies associated with avocado in Costa Rica is subsp. *fastidiosa*. The establishment of *X. fastidiosa* subsp. *fastidiosa* in New Zealand would be likely to impose the greatest losses on the economically important wine industry, since wine grapes are highly susceptible and wine production is concentrated in warmer areas. Subspecies *multiplex* appears to have the widest host range in terms of plant species expressing disease symptoms (EFSA 2015) and is thought to be more tolerant of temperate climates (Baker 2017), so impacts of this genotype could be more severe depending on its realised host range in New Zealand.

Xylella fastidiosa is known to colonise the native *Leptospermun scoparium* (manuka; Groenteman et al. 2015), the source of the valuable manuka honey industry. New Zealand's pure honey exports in 2018/19 were worth $355 million, slightly over half of which was monofloral manuka honey and a further significant proportion was multifloral manuka honey (MPI 2019). Little is known about the potential impacts of *X. fastidiosa* on manuka but, given the value of the honey export industry, even a low level of impact could be significant.

Nurseries could have significant financial losses from impacts on susceptible plants and also due to movement restrictions. New Zealand's plant production (nursery) industry is worth an estimated $500 million per year (NZPPI 2019), including significant native plant production. Some native plants are known hosts.

There is little information on which to assess impacts on the avocado industry. Infection of avocado is very rarely reported, and impacts have not been quantified. However, the CMIs for the localities where the only symptomatic infections have been reported with all of New Zealand are low (0.4–0.6) (Phillips et al. 2018), suggesting that climate conditions in avocado growing areas in New Zealand may not be ideal for symptom expression.

Nielsen et al. (2019) assessed the economic impact of *Xylella* in New Zealand. These authors concluded that although economic impacts assessments have been carried out for Australia and for California, it is not possible to extrapolate these to the New Zealand situation for reasons discussed at the beginning of this section. However, they concluded that horticultural production worth between $301 million per year and $1.7 billion per year would be at risk if *X. fastidiosa* became established in New Zealand.

The introduction of *X. fastidiosa* would very likely result in a costly response and potentially in trade restrictions for nursery stock commodities and cut flowers and foliage. Restrictions for *X. fastidiosa* are not generally imposed on fresh produce or seeds since these pathways are considered unlikely to introduce the bacterium (EFSA 2015). Australia, New Zealand's most important trading partner, is considered to be free of *X. fastidiosa* (EPPO 2021). The establishment, or even an incursion of *X. fastidiosa* is likely to result in trade disruption since the bacterium is considered to be their highest priority plant pest[41].

Given the arguments and evidence above, that is:

- *Xylella fastidiosa* can cause serious damage to crops and plant-based industries that are of high economic importance to New Zealand, for example, grapes, summerfruit, citrus;
- the introduction of *X. fastidiosa* may result in market access restrictions for nursery stock and cut flowers, particularly to Australia which is New Zealand's main trading partner;
- the costs of responding to an incursion of *X. fastidiosa* and/or long-term management costs resulting from establishment if the bacterium are likely to be very high;
- but considering that impacts on some crops may be limited by New Zealand's temperate climate (depending on the bacterial genotype introduced);

the economic impact of *X. fastidiosa* to New Zealand is considered to be very high, with high uncertainty.

Environmental impacts

Sentinel host plant testing overseas has shown that *X. fastidiosa* is able to infect a range of native plants, including cabbage tree (*Cordyline australis*), kauri (*Agathis australis*), manuka (*Leptospermum scoparium*), pōhutukawa (*Metrosideros* spp.) and several *Coprosma* and *Pittosporum* species (Groenteman et al. 2015). In this study, although plants displaying typical *Xylella* symptoms were preferentially sampled, a definitive link between symptoms and infection has not yet been established[42]. However, it is highly likely that infection with *X. fastidiosa* will cause disease symptoms on at least some native plants in at least some areas of New Zealand.

The bacterium has the potential to spread from the managed estate to the natural estate and within the natural estate. One known vector species, the spittlebug *P. spumarius,* is widespread in New Zealand and has a host range that includes native and crop species. It has been collected feeding on avocado (PPIN 2021) and on a number of native plants. Examples include Pōhutukawa (*Metrosideros excelsa*) (Plant-SyNZ 2022), *Nothofagus* sp. (e.g. NZAC04158227; NZAC 2021), *Phormium* (e.g. NZAC04158089; NZAC 2021), *Pseudopanax arboreus* (as *Neopanax arboretum*) (e.g. NZAC04171451; NZAC 2021), *Olearia nummulariifolia* and *O. virgata* (e.g. NZAC04171172 and NZAC04171473 respectively; NZAC 2021), *Veronica salicifolia* (Lariviere et al. 2010) and *Melicytus ramiflorus* (Sandanayaka and Charles 2006).

In addition, there are a number of native spittlebugs which are potential vectors. Some of these species, e.g. *Carystoterpa fingens* and *C. vagans*, also have indicative host ranges that include introduced and native plants (Lariviere et al. 2010).

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[41] https://www.awe.gov.au/biosecurity-trade/pests-diseases-weeds/plant/national-priority-plant-pests-2019.
[42] research is currently being carried out to address this in the Better Border Biosecurity (B3) collaboration.

There is uncertainty associated with predicting the level of environmental impacts that could result from an incursion of *X. fastidiosa*. Although it is likely that infection would produce disease in some native plants, it is not known how likely this is to have an impact at the population or ecosystem level, and the climate suitability will vary with the genotype of *X. fastidiosa* that is introduced.

However, given the arguments and evidence above, the impact on the New Zealand environment from the establishment of *X. fastidiosa* is considered to be moderate, with high uncertainty.

Health impacts

Xylella fastidiosa is not known to pose risks to human health, and no evidence was found of this bacterium causing human health issues. Searches using the term '*Xylella fastidiosa*' in Pubmed, Google Scholar, CAB abstracts and Google found no mention of adverse effects on humans and animals.

Given the arguments and evidence above, the health impact of *X. fastidiosa* to New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

As with other impacts, the sociocultural impacts of *X. fastidiosa* establishment are expected to be variable depending on the particular genotype involved. Very high socio-cultural impacts could be expected if Pierce's disease (subsp. *fastidiosa*) established and the climate was suitable for disease expression. Impacts could potentially be similar to those that resulted from New Zealand's Psa (bacterial canker of kiwifruit or *Pseudomonas syringae* pv. *actinidiae*) outbreak. Job and income losses impacted not only on growers but on whole communities that supported the industry (Vanneste 2017).

Xylella fastidiosa can infect amenity plants that are valued in modified environments, e. g. oaks (*Quercus* spp.), elms (*Ulmus* spp.) and poplars (*Populus* spp.) (Delbianco et al. 2021). However, it is uncertain to what extent such amenity trees might be affected in the New Zealand climate. The bacterium is also able to infect a range of native species of high cultural significance, such as kauri, cabbage tree, pōhutukawa and manuka. These impacts are discussed under "*Environmental impacts*" above.

Nurseries could have significant financial losses from impacts on plants and also due to restrictions on movement of many plants, possibly leading to job losses or even closure of some businesses (Parkinson and Malumphy 2020). New Zealand's nursery industry employs approximately 4000 people (NZPPI 2019).

Given the arguments and evidence above, the sociocultural impact of *X. fastidiosa* to New Zealand is considered to be moderate to high, with high uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, health and society is considered to be very high, with high uncertainty.

4.3.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of *X. fastidiosa* entering New Zealand associated with avocado budwood is considered to be very low (with high uncertainty);
- assuming entry, the likelihood of exposure is considered to be high (with low uncertainty);
- assuming successful exposure, the likelihood of establishment is considered to be moderate, with moderate uncertainty;
- the overall impact on the New Zealand economy, environment, health and society is considered to be very high (with high uncertainty)

the overall level of risk to New Zealand from *X. fastidiosa* on imported avocado budwood is assessed as high, with moderate uncertainty.

4.3.9 Management considerations

Diseases caused by *X. fastidiosa* are often characterized by leaf scorch, defoliation, foliage wilt and a general decline in vigour (FAO 2018). However, symptoms are not a good diagnostic indicator for *X. fastidiosa*. Their expression is heterogeneous, depending on the host plant species, *X. fastidiosa* genotype and the climatic conditions (FAO 2018). When present they can mimic symptoms of water stress or physiological disorders (FAO 2018). More importantly, asymptomatic infections of *X. fastidiosa* occur frequently, with colonised plants either exhibiting very long latent periods or remaining symptomless (EFSA 2013; Sicard et al. 2018).

Temperature and stress are important factors for symptom expression, with bacterial growth generally faster at higher temperatures, however the optimal temperature may depend on the bacterial genotypes that are introduced (Parkinson and Malumphy 2020).

The distribution and concentration of *X. fastidiosa* within the plant can vary seasonally. In general, sampling should be performed during the period of active growth and the best plant material to test is the leaf petiole and the midrib of mature leaves from either asymptomatic or symptomatic plant material.

Xylella fastidiosa is a fastidious bacterium and its strains are difficult to isolate, even from symptomatic plants, and difficult to grow in axenic (pure) culture. Detection is based on serological methods (ELIZA, etc), and a variety of molecular methods including:

- DNA extraction using a number of commercially available kits
- conventional and real time polymerase chain reaction (PCR) using a variety of primers and probes,
- loop-mediated isothermal amplification (LAMP).

A multilocus sequence typing (MLST) approach has been described for the identification of *X. fastidiosa* subspecies.

4.3.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Diseases caused by *X. fastidiosa* are often characterized by leaf scorch, defoliation, foliage wilt and a general decline in vigour (FAO 2018). Symptoms reported in avocado include chlorotic mottling, marginal scorch, deformation of leaves, defoliation, shortening of internodes, and branch dieback (Montero-Astúa et al. 2008).

However, symptoms are not a good diagnostic indicator for *X. fastidiosa*. Their expression is heterogeneous, depending on the host plant species, *X. fastidiosa* genotype and the climatic conditions (FAO 2018). When present they can mimic symptoms of water stress or physiological disorders (FAO 2018).

More importantly, asymptomatic infections of *X. fastidiosa* occur frequently, with colonised plants either exhibiting very long latent periods or remaining symptomless (EFSA 2013; Sicard et al. 2018). Asymptomatic infections are reported to occur in avocado, and even where infections are symptomatic, the symptoms may not be uniformly expressed in the plant. Montero-Astúa et al. (2008) observed that some branches showed symptoms while others in the same avocado plant did not.

- ***What are the known environmental conditions conducive to symptom expression?***

Many *X. fastidiosa* infections are asymptomatic, including some of those reported in avocado. Where symptoms are expressed, they are usually more pronounced in stressed plants, particularly those that are water deficient (FAO 2018; Bragard et al. 2019). Temperature is also an important factor for symptom expression. Diseases caused by *X. fastidiosa* are more common in tropical and subtropical regions (Janse and Obradovic 2010). Feil and Purcell (2001) determined that the optimal growth rate (*in vitro*) of the bacterium causing Pierce's disease of grapevine (*X. fastidiosa* subsp. *fastidiosa*) was 28°C. They postulated that temperatures between 25 and 32°C may be critical for disease development because of the rapid bacterial growth rate at these temperatures, whereas temperatures below 12 to 17°C and above 34°C may affect the survival of *X. fastidiosa* in plants. Other subspecies are likely to have different temperature responses, for example *X. fastidiosa* subsp. *multiplex* is considered to be more cold-tolerant due to the more temperate distribution of the diseases it causes (Baker 2017). However, symptom expression is likely to be reduced at lower temperatures for all bacterial genotypes (Parkinson and Malumphy 2020).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Samples should be taken from parts close to the symptoms and preferably consist of stems that have mature symptomatic leaves with petioles and woody twigs. Individual leaves with petioles can also be sampled. The best plant material to test for the presence of *X. fastidiosa* is the leaf petiole and the midrib of mature leaves from either asymptomatic or symptomatic plant material. Samples of necrotic and dead tissue or sections of the plant at an advanced stage of infection are not suitable for diagnosis, as saprophytes quickly colonize this tissue, interfering with the isolation or detection of the pathogen (FAO 2018). A diagnostic protocol is provided by FAO (2018) as an annex to ISPM 27.

- ***What is the optimum season for conducting detection testing?***

The distribution and concentration of *X. fastidiosa* within the plant can vary seasonally. In general, sampling should be performed during the period of active growth of the plants (Hopkins 1980). This is usually from late spring to autumn in temperate zones. For asymptomatic plants, sampling is also possible during the period of active growth. In temperate regions or in deciduous plants, sampling after warm periods (e.g., late summer–early autumn) increases the probability of accurate bacterial detection because the bacteria do not move into the new season's growth until the middle of summer. However, for tropical plant species grown indoors, sampling may be performed all year round when plants are exhibiting periods of active growth (FAO 2018).

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5. Pest risk assessments: Fungi

5.1 *Akaropeltopsis* sp. (Sooty blotch of avocado)

Akaropeltopsis sp. is a fungus that causes sooty blotch on the branches, stems, leaf veins and fruit of *Persea americana* (avocado).

5.1.1 Taxonomic description

Scientific name: *Akaropeltopsis* sp. Batista & Peres, 1966

Order/Family: Microthyriales/Micropeltidaceae

Other names include: *Stomiopeltis citri* Bitancourt (sooty blotch of citrus) has been described in the literature as the causative agent of sooty blotch of avocado (Kotze and Theron 1979). However, morphological analysis suggests that *Stomiopeltis citri* is not synonymous *Akaropeltopsis* sp., and that causative agent of sooty blotch of avocado is in fact *Akaropeltopsis* sp. (Theron et al. 1981).

Taxonomic notes: *Akaropeltopsis* is similar to *Stomiopeltis* (Theiss.) and other genera in the family Micropeltidaceae. This family is characterised by genera that grow superficially on the cuticle of plants. *Akaropeltopsis* sp. resembles *Stomiopeltis citri* (sooty blotch of citrus) morphologically but differs by having 16-spored asci. Their similarity has resulted in misidentifications and a degree of uncertainty regarding the causative agent of sooty blotch on avocado is reflected in the literature (Kotze and Theron 1979; Theron et al. 1981).

5.1.2 Hazard identification

Akaropeltopsis sp. is not known to be present in New Zealand:

- *Akaropeltopsis* sp. is not listed in either Biota NZ (2022) or PPIN (2021).
- The regulatory status of *Akaropeltopsis* sp. has not been assessed (ONZPR 2021).

Akaropeltopsis sp. has the potential to establish (and spread) in New Zealand because:

- The reported host of *Akaropeltopsis* sp. is *P. americana*. Avocado plants are commonly grown in New Zealand, in home gardens and commercial production, mainly in the north of the North Island.
- The climate is unlikely to limit its development and spread: *Akaropeltopsis* sp. is found in South Africa which has a Climate Match Index (CMI) ≥ 0.7 indicating a similar climate to the whole of New Zealand (Phillips et al. 2018).

Akaropeltopsis sp. has the potential to cause harm to New Zealand because:

- It does not parasitise the host and the blotches are only on the surface, but they reduce the marketability and value of avocado fruit (Theron et al. 1981; Smith et al. 1985; Smith et al. 1987; Coates et al. 2001),
- Avocados are an important commercial crop in New Zealand. In the 2018–2019 financial year, export sales of fresh avocados earned NZ$110 million and domestic sales earned NZ$ 56.5 million (Plant & Food Research 2019).

Akaropeltopsis sp. is associated with *P. americana* budwood because:

- *Persea americana* is a known host of *Akaropeltopsis* sp. (Smith et al. 1985; Smith et al. 1987)
- *Akaropeltopsis* sp. occurs on the branches and stems of avocado plants (Smith et al. 1985).

Given the arguments and evidence above, *Akaropeltopsis* sp. meets the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.1.3 Risk assessment

Biology and epidemiology of Akaropeltopsis sp.

Akaropeltopsis sp. is an unspecified species of fungi from the Micropeltidaceae family, that causes sooty blotch on avocado. The mycelium grows superficially on the cuticle of the host tissue. Although the organism does not parasitise the host, it produces blotches that spoil the appearance of the fruit and decreases its market value.

Host range

Similarities between *Akaropeltopsis* sp. found on avocado and *A. marchaeriifolii* found on leaves of a *Machaerium* sp. in Brazil have been reported but these species have not been confirmed to be conspecific (Theron et al. 1981; Smith et al. 1987). No other records of *Akaropeltopsis* sp. causing sooty blotch on other hosts were found in the literature, suggesting that *Akaropeltopsis* sp. is limited to *P. americana*.

Distribution

There are multiple, reliable records of *Akaropeltopsis* sp. causing sooty blotch on avocado in South Africa (Theron et al. 1981; Smith et al. 1985; Smith et al. 1987; Korsten et al. 1997).

There is little information on the distribution of *Akaropeltopsis* sp. elsewhere in the world, and the records of its distribution are largely unsubstantiated or unconfirmed (Theron et al. 1981; Smith et al. 1987; Queensland Government 2001), as follows:

- Sooty blotch on avocado in Australia has been reported to be caused by a range of causative agents, including *Stomiopeltis* sp. (Coates et al. 2001; Willingham 2002; Cooke et al. 2009), *Akaropeltopsis* spp./*Stomiopeltis* spp. complex (Queensland Government 2001), and multiple non-specified fungi (Allen 2004). Given that the causative agents of sooty blotch on avocado have been confused in the past due to the high level of morphological similarity between *Akaropeltopsis* and *Stomiopeltis* (Theron et al. 1981; Smith et al. 1987), and that there were no records found describing *Akaropeltopsis* sp. on avocado in Australia, there is uncertainty regarding the presence of *Akaropeltopsis* sp. in Australia.
- *Akaropeltopsis* sp. has been reported to be present and associated with avocado in Colombia, but this is based on international literature compilation and not from direct observation. According to the USDA (2016), the presence of *Akaropeltopsis* sp. in Colombia is 'doubtful'.
- *Akaropeltopsis* sp. appears to be similar to *A. marchaeriifolii* found on leaves of a *Machaerium* sp. in Brazil, but these species have not been confirmed to be conspecific (Theron et al. 1981; Smith et al. 1987). No other records were found in the literature of *Akaropeltopsis* sp. causing sooty blotch in Brazil.

Table 5-1. The potential geographic distribution of *Akaropeltopsis* sp. causing Sooty blotch on avocado.

Country	CMI[43] range	Notes	References
Australia	0.4 - 1.0	Presence is uncertain	Coates et al. (2001); Queensland Government (2001); Willingham (2002); Cooke et al. (2009)
Brazil	0.3 - 0.9	Presence is uncertain	Theron et al. (1981); Smith et al. (1987)
Columbia	0.3 - 0.8	Presence is uncertain	USDA (2016)
South Africa	0.5 - 0.9	Presence is confirmed	Theron et al. (1981); Smith et al. (1985); Smith et al. (1987)

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[43] Climate Match Index (CMI) with all of New Zealand (Phillips et al. 2018).

Symptoms

Avocado sooty blotch is characterised by sooty blotches on the branches, stems, leaf veins and fruit (Smith et al. 1985). The blotchy appearance is due to irregularly shaped mycelial mats that grow on the plant surface. Mycelium originates from ascospores or mycelial fragments and initially appear as small light brown spots on the plant surface (Smith et al. 1987). Over the summer season, the mycelium spreads over the plant, turns to a charcoal black colour and becomes thick and uneven, resulting in the characteristic sooty blotched appearance (Queensland Government 2001; Allen 2004). When new flushes emerge in spring they contrast in colour to the darkened twigs from the previous season. Mycelium usually takes at least two seasons to colonise the leaves. *Akaropeltopsis* sp. penetrates the wax layer of the plant but not the cuticle or the epidermal cells, and therefore does not cause rotting or damage of plant tissues. Haustoria have not been observed (Kotze and Theron 1979; Theron et al. 1981; Smith et al. 1985; Smith et al. 1987; Coates et al. 2001).

Optimal conditions for growth and symptom development

Mycelial growth is responsible for the sooty blotched appearance on the surface of the plant, and is promoted by surface moisture (dew, rain, high humidity), low airflow, shade and warm temperatures (Allen 2004). Mycelium grows and spreads over the plant surface during summer (Smith et al. 1987).

Reproduction and transmission

The mycelium of *Akaropeltopsis* sp. produces and releases large amounts of airborne spores at the onset of summer rains (Cooke et al. 2009). These spores are the primary source of spread and infection, and germinate readily in a film of moisture on the surface of a host plant (Smith et al. 1985). Ascospores are 14-16 µm x 4-5 µm (Smith et al. 1985).

5.1.4 Likelihood of entry

There is potential for *Akaropeltopsis* sp. to be imported into post-entry quarantine on avocado budwood, in which case it would be likely to produce visible symptoms in PEQ:

- *Akaropeltopsis* sp. appears to be restricted to South Africa. There are several unconfirmed/unreliable records of its presence in Australia, Brazil and Colombia.
- it can be associated with leafless avocado budwood because it colonises the surface of branches, stems, leaves and fruit (Smith et al. 1985).
- budwood/leafless cuttings are dipped in 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility, which is considered to reduce the risk of importing *Akaropeltopsis* sp. into PEQ to very low, with low uncertainty.
- if *Akaropeltopsis* sp. is imported into PEQ, it is highly likely to produce visible symptoms in PEQ because it is reported to grow and spread under warm and wet conditions.
- airborne spores are 14-16 µm x 4-5 µm (Smith et al. 1985), indicating they may escape from because the mesh screen over the level 3A PEQ vents is required to have a maximum opening size of 0.04 mm^2 (MPI 2021).
- however, the fungus is slow growing and takes months to produce spores (Smith et al. 1985) so it would be detected by plant inspections well before spores are produced. Furthermore, spore production is promoted by rain (Cooke et al. 2009), suggesting that conditions in level 3A PEQ would not be conducive to spore production because there is no overhead irrigation (MPI 2021).

Given the arguments and evidence above, that is:

- *Akaropeltopsis* sp. has a limited global distribution, with high uncertainty.
- it can be present on the surface of budwood (Smith et al. 1985), but such infections are highly likely to be manged by 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility.
- if *Akaropeltopsis* sp. is imported into PEQ then it is highly likely to produce visible symptoms in level 3A PEQ.

- the fungus is unlikely to sporulate in level 3A PEQ because it takes months to produce spores and PEQ conditions may not be conducive to spore production.

the likelihood of *Akaropeltopsis* sp. entering New Zealand associated with *P. americana* (avocado) budwood is considered to be **negligible**, with **low** uncertainty.

5.1.5 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of *Akaropeltopsis* sp. entering New Zealand associated with *P. americana* budwood is considered to be negligible with low uncertainty. Therefore, continuing the assessment is considered unnecessary; and

the overall level of risk to New Zealand from *Akaropeltopsis* sp. on imported *P. americana* budwood is considered to be negligible, with low uncertainty.

5.1.6 Management considerations

Avocado sooty blotch is characterised by sooty blotches on the branches, stems, leaf veins and fruit (Smith et al. 1985). The blotchy appearance is due to mycelial mats that grow on the plant surface during the summer (Smith et al. 1987; Queensland Government 2001; Allen 2004). Growth and spore production is promoted by surface moisture (dew, rain, high humidity), low airflow, shade and warm temperatures (Allen 2004; Cooke et al. 2009). *Akaropeltopsis* sp. does not parasitise the host and the blotches are only on the surface of the plant (Theron et al. 1981; Smith et al. 1985; Smith et al. 1987; Coates et al. 2001).

There were no reports found in the literature of asymptomatic *Akaropeltopsis* sp. infections on avocado. In case the sodium hypochlorite dip treatment fails to penetrate into creases and folded plant surfaces, plant parts such as buds could be tested to maximise the chances of detection of the pest in PEQ.

Sooty blotch is effectively controlled with petroleum-based oil sprays and copper sprays (Coates et al. 2001). Fungicides used to control anthracnose are considered to be effective against sooty blotch (Queensland Government 2001; Aglave 2018). These include azoxystrobin and copper hydroxide (Eskalen and Faber 2016).

5.1.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Avocado sooty blotch is characterised by sooty blotches on the branches, stems, leaf veins and fruit (Smith et al. 1985). The blotchy appearance is due to mycelial mats that grow on the plant surface. Mycelia originates from ascospores or mycelial fragments. *Akaropeltopsis* sp. infection initially appears as small light brown spots on the plant surface (Smith et al. 1987). Over the summer season, mycelia spread over the plant, turns to a charcoal black colour and becomes thick and uneven, resulting in the characteristic sooty blotched appearance (Queensland Government 2001; Allen 2004). When new flushes emerge in spring, they contrast in colour to the darkened twigs from the previous season. Mycelium usually takes two seasons to colonise the leaves. Haustoria44 have not been observed (Smith et al. 1985; Smith et al. 1987). *Akaropeltopsis* sp. does not parasitise the host

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[44] Haustoria are specialised fungal hypha that penetrate the tissues of a host and absorbs nutrients and water.

and the blotches are only on the surface of the plant (Theron et al. 1981; Smith et al. 1985; Smith et al. 1987; Coates et al. 2001).

- ***What are the known environmental conditions conducive to symptom expression?***

Mycelial growth is responsible for the sooty blotched appearance on the surface of the plant, and is promoted by surface moisture (dew, rain, high humidity), low airflow, shade and warm temperatures (Allen 2004). Mycelium grows and spreads over the plant surface during summer (Smith et al. 1987).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

There were no reports found in the literature of asymptomatic *Akaropeltopsis* sp. infections on avocado. In case the sodium hypochlorite dip treatment fails to penetrate into creases and folded plant surfaces, plant parts such as buds could be tested to maximise the chances of detection of the pest in PEQ

- ***What is the optimum season for conducting detection testing?***

Mycelial growth and sporulation is promoted by moisture (dew, rain, high humidity), low airflow, shade and warm temperatures (Allen 2004). Mycelium grows and spreads over the plant surface during summer (Smith et al. 1987), and large amounts of airborne spores are produced and released at the onset of summer rains (Cooke et al. 2009), indicating that summer is the optimum season for conducting detection testing.

5.1.8 References

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5.2 Ambrosia fungi of the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp. (vectored by beetles of the *Euwallacea fornicatus* species complex)

Ambrosia fungi of the Ambrosia *Fusarium* Clade (AFC), *Graphium* and *Paracremonium* are symbionts associated with small (1–2 mm) ambrosia beetles in the genus *Euwallacea*. These fungi supply essential diet components for the ambrosia beetles and in return rely on the beetles to carry them to a new host. By cultivating their fungal symbionts in the xylem of plants, some *Euwallacea* species have managed to expand their host range and become invasive in new areas of the world. In recent years, for example, the *Euwallacea fornicatus* complex, together with their symbiotic fungi, have become a concern to the avocado industry in California, Florida, Australia, Israel and South Africa.

5.2.1 Taxonomic description

Taxonomic scope: The taxonomic scope of this PRA are ambrosia fungi (Table 5-2 and Table 5-3) vectored by beetles in the *Euwallacea fornicatus* species complex (Coleoptera: Scolytinae) (Table 5-4), known to attack living avocado trees and causing dieback.

The Ambrosia *Fusarium* Clade: The primary symbionts associated with *Euwallacea* are fusaria that form a monophyletic group within Clade 3 of the *Fusarium solani* species complex (FSSC), known as the Ambrosia *Fusarium* Clade (AFC, Kasson et al. (2013)). Recently, it was proposed that species in the FSSC, including the AFC species, be placed in the genus *Neocosmospora* (Sandoval-Denis et al. 2019). However, this change has not been widely accepted and new AFC species are still being described as *Fusarium* (Aoki et al. 2021; Lynn et al. 2021). To date, at least 23 AFC species are known, some of which are yet to be described (Table 5-2). The list of AFC fungi is currently being updated and is expected to keep expanding, as is the list of their associated beetle species (Aoki et al. 2021; Jiang et al. 2021; Lynn et al. 2021).

Table 5-2. Species of the Ambrosia *Fusarium* Clade (AFC) and associated beetle species, assembled from Kasson et al. (2013), Owens et al. (2018), Carrillo et al. (2019), Aoki et al. (2021), Lynn et al. (2021) and Jiang et al. (2021). *Fusarium* species in bold **indicate isolates from avocado. Associated *Euwallacea* beetle vector is provided where known.**

Scientific name	AFC species	Associated beetle species
Fusarium ambrosium	AF-1	*Euwallacea perbrevis* (TSHBa)
Fusarium euwallaceae	**AF-2**	*E. fornicatus* (PSHB)
Fusarium floridanum	AF-3	*E. interjectus* (formerly *Xyleborus interjectus*)
Fusarium oligoseptatum	AF-4	*E. validus* (formerly *Xyleborus validus*)
Fusarium tuaranense	AF-5	unknown
***Fusarium* sp.**	**AF-6**	*E. perbrevis* (TSHBa)
Fusarium obliquiseptatum	**AF-7**	*Euwallacea* sp.
Fusarium duplospermum	**AF-8**	*E. perbrevis* (TSHBa)
Fusarium sp.	AF-9	*Xyleborus ferrugineus*
Fusarium drepaniforme	AF-10	unknown
Fusarium papillatum	AF-11	*E. perbrevis* (TSHBa)
Fusarium kuroshium	**AF-12**	*E. kuroshio* (KSHB), *E. fornicatus* (PSHB), *E. interjectus*
***Fusarium* sp.**	**AF-13**	*E. fornicatus* (PSHB), *E. perbrevis* (TSHBa)
***Fusarium* sp.**	**AF-14**	*E. fornicatus* (PSHB), *E. perbrevis* (TSHBa)
***Fusarium* sp.**	**AF-15**	*E. fornicatus* (PSHB)
***Fusarium* sp.**	**AF-16**	*E. fornicatus* (PSHB), *E. perbrevis* (TSHBa)

Scientific name	AFC species	Associated beetle species
***Fusarium* sp.**	**AF-17**	*E. fornicatus* (PSHB), *E. perbrevis* (TSHBa)
***Fusarium* sp.**	**AF-18**	*E. fornicatus* (PSHB), *E. kuroshio* (KSHB), *E. perbrevis* (TSHBa)
Fusarium rekanum	AF-19	*E. perbrevis* (TSHBa), *E. similis*
Fusarium akasia	AF-20	*E. perbrevis* (TSHBa), *E. similis*
Fusarium awan[45]	AF-21	*E. similis*
Fusarium mekan	AF-22	*E. similis*
Fusarium variasi	AF-23	unknown
Fusarium warna	AF-24	*E. perbrevis* (TSHBa)

Graphium spp. and *Paracremonium* spp. ambrosia fungi:

Auxillary (or secondary) fungi of *Euwallacea* ambrosia beetles, such as *Graphium* spp. and *Paracremonium* spp. are also isolated from other ambrosia beetle genera, such as *Scolytodes, Megaplatypus* or *Xyleborus* (Baker and Norris 1968; Kolařík and Hulcr 2009; Ceriani-Nakamurakare et al. 2016). Although some of them have recently gained attention because they were commonly associated with *Euwallacea fornicatus* disease complex on avocado (Table 5-3), most of them have not been described.

Table 5-3. *Graphium* spp. and *Paracremonium* spp. ambrosia fungi isolated from avocado (bold), assembled from Na et al. (2018), Lynch et al. (2016) and Carrillo et al. (2019). Associated *Euwallacea* beetle vector is provided where known.

Scientific name	Associated beetle species
Graphium euwallaceae	*E. fornicatus* (PSHB)
Graphium sp.	*Euwallacea* sp. (TSHB, unknown clade)
Graphium sp.	*E. fornicatus* (PSHB)
Graphium sp.	*E. fornicatus* (PSHB)
Graphium sp.	*E. fornicatus* (PSHB)
Graphium kuroshium	*E. fornicatus* (PSHB), *E. kuroshio* (KSHB)
Paracremonium sp.	*E. fornicatus* (PSHB), *Euwallacea* sp. (TSHB, unknown clade)
Paracremonium pembeum	*E. fornicatus* (PSHB), *E. kuroshio* (KSHB)

The *Euwallacea fornicatus* species complex

The *Euwallacea fornicatus* species complex (Table 5-4) and their vectored symbiotic fungi have recently become invasive outside their native range, South East Asia, and a concern for the avocado industry in various regions of the world (Carrillo et al. 2019; Freeman et al. 2019).

Table 5-4. The *Euwallacea fornicatus* species complex (Smith et al. 2019; Lynn et al. 2021).

Scientific name	Other names
Euwallacea fornicatus (Eichhoff, 1868) (= *E. tapatapaoensis* (Schedl, 1951); = *E. whitfordiodendrus* (Schedl, 1942))	PSHB; Polyphagous Shot Hole Borer
Euwallacea fornicatior (Eggers, 1923) (= *E. schultzei* (Schedl, 1951))	TSHBb; Tea Shot Hole Borer Clade b
Euwallacea perbrevis (Schedl, 1951)	TSHBa; Tea Shot Hole Borer Clade a
Euwallacea kuroshio Gomez & Hulcr, 2018	KSHB; Kuroshio Shot Hole Borer

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[45] *Fusarium awan* is the only ambrosia fungus isolated from the mycangia of an *Euwallacea* species that does not group phylogenetically within the Ambrosia *Fusarium* Clade (AFC), but still within the *Fusarium solani* species complex (FSSC) (Lynn et al. 2021).

Note: The main ambrosia fungi associated with the *Euwallacea fornicatus* species complex of concern to the avocado industry, particularly in invaded areas, such as California, Israel, Australia and South Africa, are *Fusarium euwallaceae*, *F. kuroshium*, *Graphium euwallaceae, G. kuroshium* and *Paracremonium pembeum* (Lynch et al. 2016; Na et al. 2018; Freeman et al. 2019). However, recent evidence suggests a non-specific relationship between the ambrosia fungi associated with the *Euwallacea fornicatus* complex and potential for symbiont switching within the *Euwallacea* genus (Carrillo et al. 2019; Carrillo et al. 2020a; Jiang et al. 2021; Lynn et al. 2021). Hence, different fungi-beetle combinations (Table 5-2 and Table 5-3) may be associated with *Euwallacea* beetle galleries in avocado budwood.

5.2.2 Hazard identification

The ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) are not known to be present in New Zealand:

- *Fusarium euwallaceae, F. kuroshium, F. oligoseptatum, F. tuaranense, F. obliquiseptatum, F. duplospermum, F. drepaniforme, F. papillatum, F. rekanum, F. akasia, F. awan, F. mekan, F. variasi, F. warna, G. euwallaceae, G. kuroshium* and *P. pembeum* are not listed in either Biota NZ (2022), NZOR (2021), PPIN (2021) or ONZPR (2021).

The ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) can be associated with *Persea americana* budwood because:

- *Persea americana* is a known host of some of the ambrosia fungi (AF-2, AF-6 to 8, AF-12 to 18, Table 5-2) associated with the *Euwallacea fornicatus* species complex (Carrillo et al. 2019; Freeman et al. 2019);
- the beetle vectors in the *Euwallacea fornicatus* species complex infect preferably small live branches and stems of *P. americana* (Mendel et al. 2017) used for budwood.

The ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) have the potential to establish (and spread) in New Zealand because:

- *Persea americana* (avocado) is a known host (Lynch et al. 2016; Na et al. 2018; Freeman et al. 2019) of ambrosia fungi of the *Euwallacea fornicatus* species complex (AFC, *Graphium* spp. and *Paracremonium* spp.);
- multiple potential host plants and reproductive hosts of the *Euwallacea fornicatus* species complex, such as *Acer negundo, Ricinus communis, Quercus robur, Persea americana* (Eskalen et al. 2013; Lynch et al. 2016; Na et al. 2018) are widely grown in New Zealand (NZPCN 2021) in home gardens, landscaping and commercial production;
- ambrosia fungi of the *Euwallacea fornicatus* species complex (AFC, *Graphium* spp. and *Paracremonium* spp.) have been isolated from avocado trees in areas with a climate match index (CMI) ≥ 0.7 indicating a similar climate to the whole of New Zealand (Phillips et al. 2018; Carrillo et al. 2020a);
- There are no *Euwallacea* vectors reported to be present in New Zealand (NZOR 2021). Other species (>25) of shot hole borers are reported from New Zealand and it is not known whether they could serve as vectors of AFC fungi. However, AFC fungi isolated from avocado have so far only been reported to be vectored by *Euwallacea* species (Table 5-2) and the scenario of symbiont switching to ambrosia beetles outside the genus *Euwallacea*, is considered unlikely with current available information (Kasson et al. 2013; Aoki et al. 2021).

The ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) have the potential to cause harm to New Zealand because:

- the ambrosia fungi and their associated ambrosia beetles can cause dieback and death of economically important hosts, such as *Citrus* spp. and avocado (Aoki et al. 2019), and

- the ambrosia fungi of the *Euwallacea fornicatus* species complex and their associated ambrosia beetles are not host specific (>75 host plant families) and can severely affect natural ecosystems (Eskalen et al. 2013; Coleman et al. 2019).

Given the arguments and evidence above, the ambrosia fungi of the *Euwallacea fornicatus* species complex (AFC, *Graphium* spp. and *Paracremonium* spp.) meet the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.2.3 Risk assessment

Biology and epidemiology of the ambrosia fungi of the* Euwallacea fornicatus *species complex (the Ambrosia* Fusarium *Clade (AFC),* Graphium *spp. and* Paracremonium *spp.)

Ambrosia symbiosis background

Ambrosia symbioses consist of reciprocally obligate mutualisms between ambrosia beetles, a group of specialized wood-boring weevils (Scolytinae and Platypodinae), and their symbiotic ambrosia fungi (Joseph and Keyhani 2021). Ambrosia beetle females carry their fungal symbionts in special organs, called mycangia, to a host tree where they burrow into the sapwood and construct galleries to house their brood and fungal gardens (Joseph and Keyhani 2021). Ambrosia fungi are a polyphyletic group and at least 16 independent evolutionary origins of fungus farming by ambrosia beetles have been reported. One of them are fungi from the Ambrosial Fusarium Complex (AFC) farmed by Euwallacea species (Aoki et al. 2019; Mendel et al. 2021).

The genus *Euwallacea*

Euwallacea is a genus of over 40 ambrosia beetle species within the tribe Xyleborini (Freeman et al. 2016). Euwallacea species have pre-oral mycangia which harbor fungal communities from the Ambrosial Fusarium Complex (AFC) (order Hypocreales), which are their primary (obligate) symbionts (Carrillo et al. 2019). In addition, they are sometimes associated with fungi in the orders Microascales and Ophiostomatlaes, which are their secondary or auxiliary symbionts (Carrillo et al. 2019). Euwallacea species are engaged in obligate mutualism with at least 23 AFC fungi (Table 5-2). In addition, at least six Graphium species and two Paracremonium species (Table 5-3) have been reported to be associated with Euwallacea species (Carrillo et al. 2019). These symbiotic fungi have co-evolved with the beetles in mutualisms providing the beetles with nutritional supplement from low-nutrient xylem tissue, while the fungi receive reliable dispersion and direct inoculation into plant hosts (Carrillo et al. 2019). Because species of Euwallacea do not feed directly on their host plant but on their xylem inhabiting fungal symbionts, their host range is usually wide and likely depends on whether their primary symbionts can be cultivated in the host (Mendel et al. 2021).

The *Euwallacea fornicatus* species complex

Members of the *Euwallacea fornicatus* species complex and their symbiotic fungi have recently invaded areas of the United States, Australia, Costa Rica, Guatemala, Israel, Panama and South Africa, where they have become a concern to the avocado industry, urban areas and natural ecosystems (Carrillo et al. 2019). At least 4 different beetle species (Table 5-4), all morphologicaly similar, are associated with different suites of ambrosia fungi (Table 5-2 and Table 5-3). Whereas the vast majority of ambrosia beetle species attack dying or dead trees, the Euwallacea fornicatus species complex and their symbionts have a wide host range and are known to attack live and healthy trees, including avocado (Hulcr et al. 2017).

Host range and preference

Ambrosia beetles from the Euwallacea fornicatus species complex have a vast host range, including over 400 plant species in 75 families, many of which are reproductive hosts (e.g. Acer negundo, Ricinus communis, Quercus robur, Persea americana) (Eskalen et al. 2013; Gomez et al. 2019). The known host range continues to grow and is frequently updated. A recent study showed that out of 412 tree species attacked by Euwallacea fornicatus (PSHB) approximately 20% were suitable for reproduction of the beetle (Mendel et al. 2021). It was hypothesised that the AFC fungal species plays a major role in which tree species is suitable for reproduction of the beetle (Mendel et al. 2021). Mendel et al. (2017) found that although all examined avocado cultivars were attacked by the beetles there seem to be differences in preference. In general, the variety Hass was much more susceptible than Ettinger to beetle attack although the varieties Reed, Nabal and Pinkerton were also markedly affected (Mendel et al. 2017). The increased beetle attacks of the Hass variety is reflected in approximately 30% of the Hass trees displaying branch wilting compared to only 2% of the trees of Ettinger (Mendel et al. 2017).

Symptoms on avocado

On avocado, the *Euwallacea fornicatus* beetle-disease complex causes branch wilting, dieback and tree mortality (Mendel et al. 2017). Typical signs of beetle attack at junctions of small and mid-size shaded branches often show the presence of white sugar volcanoes (Carrillo et al. 2016). *Fusarium euwallaceae, F. kuroshium, Graphium euwallaceae*, *G. kuroshium* and *Paracremonium pembeum* have been shown to be weak pathogens causing lesions (necrotic tissue) associated with the sugar exudation reactions when inoculated on young healthy avocado (Lynch et al. 2016; Na et al. 2018; Freeman et al. 2019). Usually, it is the combined action of the mechanical beetle burrowing and localized pathogenicity of the ambrosia fungi (Lynch et al. 2016) that together cause cambial necrosis, branch dieback and tree mortality on avocado (Mendel et al. 2012). The occurrence and severity of wilting and dieback symptoms are related to the degree of beetle infestation in the wood (Mendel et al. 2021). Large and medium diameter avocado branches were more resistant to beetle attack, compared to thin branches (Mendel et al. 2017). Effectively, gallery density increased as branch diameter decreased (Mendel et al. 2017). Concomitantly, in large and medium diameter branches, extensive sugar exudation occurred and beetle attack rarely progressed to the formation of natal galleries, whereas minimal sugar exudation was observed in thin branches (Mendel et al. 2017). This was more evident in the branches that were weakened by repeated attacks followed by successful beetle colonization. For example, *Euwallacea fornicatus* (referred to as *E. whitfordiodendrus,* PSHB) preferred and successfully colonized branches that had been previously attacked by its conspecifics, and reproduction was much higher in these branches, as opposed to initial attacks (Mendel et al. 2017). Thus, small avocado branches, such as budwood cuttings, could have beetle galleries with visible entry holes of about 0.85 mm in diameter (Carrillo et al. 2020b) or be potentially inoculated with ambrosia fungi from failed beetle attacks (Mendel et al. 2021).

Survival of primary and secondary ambrosia fungi in live and in dead xylem of avocado

An inoculation study on avocado host trees of cultivars 'Hass' and 'Ettinger' showed that *G. euwallaceae* and *P. pembeum* could not be recovered from live xylem after 3–4 months or 2 months, respectively (Freeman et al. 2019). Similar survival patterns were obtained from other tested plant hosts: inoculation of the fungi into healthy xylem indicated that both *G. euwallaceae* and *P. pembeum* disappeared rapidly, whereas *F. euwallaceae* survived for extended periods, while the xylem tissue remained viable (Freeman et al. 2019). Auxilary fungal symbionts *Graphium* and *Paracremonium* remained viable and continued to thrive in dry and dead galleries, perhaps serving as food for the late-emerging adults and therefore appearing not to be involved in pathogenicity (Freeman et al. 2019). These findings strongly suggest that survival of AFC primary symbionts may last for extended periods in the live tissue whereas auxilary fungi (*Graphium* spp. and *Paracremonium* spp.) may persist in their aposymbiotic phase in dying or dead xylem. Hence, in case of inoculated live avocado budwood *Graphium* spp. and *Paracremonium* spp. would likely disappear after 2 months without their symbiotic beetles.

Different roles of the ambrosia fungi *Fusarium*, *Graphium* and *Paracremonium* associated with *Euwallacea* species in avocado

Mounting evidence suggests that *Euwallacea* spp. need their *Fusarium* symbionts (AFC) to survive and reproduce (Freeman et al. 2019), whereas their Graphium spp. and Paracremonium spp. symbionts are seen as auxiliary fungi (Carrillo et al. 2020a). For example, diet experiments showed that Euwallacea spp. were able to reproduce with their Fusarium and Graphium associates but could not survive or reproduce on P. pembeum alone (Carrillo et al. 2019; Freeman et al. 2019). Similarly, Carrillo et al. (2020a) showed that when solely reared on auxilary fungi, beetles had no or only very few male offspring. Although the exact role of the ambrosia fungi is not yet fully understood, it appears that Fusarium (AFC) is required for the process of establishment in a living tree, i.e. establishment of beetle galleries and first food source for female beetles in avocado (Freeman et al. 2016). During the colonization process, Graphium spp. and Paracremonium spp. appear to be the main food source for development of the larvae (Freeman et al. 2016; Freeman et al. 2019). Freeman et al. (2019) found that in invaded areas mainly the species Fusarium euwallaceae was used as the food source for the adult beetles and hatching larvae, whereas G. euwallaceae served predominantly as food for the establishment of beetle larvae and callow adults.

Vectors

Members of the *Euwallacea fornicatus* species complex have been reported as pests of avocado (Carrillo et al. 2019) and as vectors of xylem-inhabiting Fusarium (AFC), Graphium spp. and Paracremonium spp. (Aoki et al. 2019; Carrillo et al. 2019). Currently, there are no *Euwallacea* spp. reported from New Zealand (NZOR 2021; PPIN 2021). However, there are at least 25 other ambrosia beetles present in New Zealand, including xyleborine ambrosia beetles (Brockerhoff et al. 2003; NZOR 2021) and polyphagous ambrosia beetles that have been reported from avocado, such as Xylosandrus spp. (Virgen et al. 2021). Lateral transfer of fungal symbionts has been reported between sympatric ambrosia beetle species, possibly facilitated by the beetles colonizing the same host (Kolařík and Hulcr 2009; Carrillo et al. 2014; Ceriani-Nakamurakare et al. 2016; Carrillo et al. 2019). However, outside the genus Euwallacea, only one other ambrosia beetle species (Xyleborus ferrugineus) has been reported to have an association with an AFC species (i.e., AF-9, so far not isolated from avocado), and with Graphium and Acremonium-like species (Baker and Norris 1968; Paciura et al. 2010; Kasson et al. 2013). Xyleborus ferrugineus is not reported to occur in New Zealand (NZOR 2021). The gall wasp Quadrastichus erythrinae was also hypothesised to be an alternative vector of two AFC fungi (isolates it-1 and it-2) on Erythrina trees in Japan (Takashina et al. 2020). The potentially alternative vector Q. erythrinae is not reported to be present in New Zealand (NZOR 2021). Thus, there are no known vectors of AFC fungi from avocado (Table 5-2) currently reported from New Zealand (Bateman et al. 2016; Joseph and Keyhani 2021; NZOR 2021).

5.2.4 Likelihood of entry

The ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) are associated with ambrosia beetle infestation and beetle galleries in avocado (Lynch et al. 2016; Carrillo et al. 2019; Freeman et al. 2019). Small branches are preferred over bigger stems for beetle galleries in the host avocado (Freeman et al. 2019). Therefore, ambrosia fungi and their beetles could be associated with leafless, rootless avocado budwood cuttings.

The routine surface disinfection of budwood entering PEQ (dipping in 1% sodium hypochlorite for 2 minutes) will most likely not eliminate infection, given the fungus will be associated with beetle galleries where the surface disinfection may not reach. However, if avocado budwood cuttings have an established infection, it is likely that beetle galleries will be detected, and infected material removed or destroyed, during phytosanitary inspections on arrival in New Zealand.

The results of Freeman et al. (2019) suggest that *Graphium euwallaceae, G. kuroshium and Paracremonium pembeum* would only survive PEQ on dead xylem. In live xylem (as in fresh budwood cuttings) the fungi would likely disappear after 2 months (Freeman et al. 2019). Primary ambrosia fungi from the AFC, such as *F. euwallaceae* were shown to survive in live xylem for more than 25 months (Freeman et al. 2019). This infection, however, remains localized near the beetle entrance holes and galleries, typically at the base of a branch, and results in wilt and dieback of individual branches (Freeman et al. 2019; Kendra et al. 2020; Mendel et al. 2021).

As part of the commodity description, all imported avocado germplasm will undergo a growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ. This period in 3A PEQ will impact on likelihood of entry in the following manner:

- ambrosia beetle galleries (i.e. entry holes and sugar exudates) will be visible during inspection and thus infected budwood would be detected and destroyed before plants are released from PEQ, and

- budwood will be treated with miticides and insecticides, which are unlikely to affect ambrosia beetles residing internally (see section 2.2). However, Level 3A PEQ has measures in place to manage the risks associated with insects, wastewater, waste and accidental transfer (see section 2.2), therefore ambrosia beetles and their fungal symbionts are highly unlikely to escape level 3A PEQ to infect a suitable host.

Therefore, the residual biosecurity risks are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ.

Given the arguments and evidence above, that is:

- established infections and beetle galleries are highly likely to be detected, and infected material removed or destroyed, during preparation for export, in phytosanitary inspections pre-export or on arrival in New Zealand;
- residual biosecurity risks from ambrosia entry points or recent infections on the surface of avocado budwood are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ

the likelihood of ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia Fusarium Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) entering New Zealand associated with *Persea americana* budwood is considered to be very low, with low uncertainty.

5.2.5 Likelihood of exposure

This assessment is made on the basis that ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia Fusarium Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) have entered New Zealand on avocado budwood as in the commodity description undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand.

If avocado plants are infected with ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) at the time of their release from Post Entry Quarantine (PEQ) they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Primary ambrosia fungi of the *Euwallacea fornicatus* species complex live in obligate mutualism with *Euwallacea* ambrosia beetles and depend on their beetle vectors to carry and inoculate them into the xylem of a new host (Lynn et al. 2021). No known vectors are reported to be present in New Zealand (see section Vectors, NZOR (2021)).

The ambrosia fungi associated with the *Euwallacea fornicatus* species complex on avocado have so far been shown to have localised pathogenicity (<10 cm spread of beetle entry or inoculation point) in avocado and no systemic infection by these fungi is currently known (Mendel et al. 2017; Freeman et al. 2019). Thus, the here assessed ambrosia fungi would remain localised, in the individual avocado branch.

Given the arguments and evidence above, the likelihood of ambrosia fungi of the *Euwallacea fornicatus* species complex transferring from *Persea americana* budwood onto a host in New Zealand is considered to be very low, with moderate uncertainty.

Therefore, further assessment is considered unnecessary at this time, and likelihoods of establishment and impacts to New Zealand were not assessed.

5.2.6 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood above, that:

- the likelihood of ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia Fusarium Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) entering New Zealand associated with *Persea americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ) is considered to be very low, with low uncertainty;
- the likelihood of transferring from the imported commodity onto a suitable host is considered to be very low, with moderate uncertainty;
- therefore, continuing the assessment was considered unnecessary; and

the overall level of risk to New Zealand from ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) on imported *Persea americana* budwood (as in the commodity description) is considered to be negligible, with low uncertainty.

5.2.7 Management considerations

Pesticides and trapping

Infections with ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia Fusarium Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) and their beetles are inside of hardwood. Thus, the fungi and the beetles are difficult to control with systemic insecticides, contact insecticide, fungicides or biological control (Eatough Jones et al. 2017). No pesticide combination provided substantial control over time (Eatough Jones et al. 2017). Trapping of female beetles of the *Euwallacea fornicatus* species complex might reduce damage. For example, α-copaene was shown to be equal in attraction to quercivorol, a food-based attractant and the standard lure for *E*. nr. *fornicatus* (Kendra et al. 2020); moreover, a combination of α-copaene and quercivorol lures resulted in synergistic attraction and improved pest detection (Kendra et al. 2020). This two-component lure has field longevity of 3 months and a sampling range of 30–35 m (Kendra et al. 2020).

Molecular identification

Carrillo et al. (2020b) developed multiplex real-time quantitative PCR assays using hydrolysis probes targeting the β-tubulin gene to detect, distinguish, and quantify ambrosia fungi associated with the polyphagous shot hole borer (PSHB; *Euwallacea fornicatus*, *Fusarium euwallaceae*, *Graphium euwallaceae* and *Paracremonium pembeum*) as well as the Kuroshio shot hole borer (KSHB; *Euwallacea kuroshio*, *Fusarium kuroshium* and *Graphium kuroshium*).

5.2.8 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Symptom expression with beetle galleries: Infection with the ambrosia fungi of the *Euwallacea fornicatus* species complex (the Ambrosia *Fusarium* Clade (AFC), *Graphium* spp. and *Paracremonium* spp.) is usually associated with ambrosia beetles or their galleries (Carrillo et al. 2020b; Mendel et al. 2021). External symptoms associated with beetle galleries are visible (small holes of about 1 mm and sugar exudates) (Carrillo et al. 2020b). White powdery exudate on the outer bark is associated with beetle holes and wood under exit holes of galleries will reveal brown necrosis (Mendel et al. 2017). Older plants also show black pitch symptoms (i.e. black exudate found on branches) and branch dieback and wilting (Mendel et al. 2017). The ambrosia fungi associated with the *Euwallacea fornicatus* species complex on avocado have so far been shown to have localised pathogenicity (<10 cm spread of beetle entry or inoculation point) in avocado and no systemic infection by these fungi is currently known (Mendel et al. 2017; Freeman et al. 2019), as opposed to other ambrosia fungi, such as in the genus *Raffaelea* (Joseph and Keyhani 2021). Thus, symptom expression of the ambrosia fungi of the *Euwallacea fornicatus* species complex (AFC, *Graphium* spp. and *Paracremonium* spp.) is expected to be in close proximity to beetle galleries in the host avocado (from entry point, Mendel et al. (2017)) and external symptoms associated with beetle galleries are expected to be visible.

Symptom expression without beetle galleries: Inoculation of avocado branches with ambrosia fungi of the *Euwallacea fornicatus* species complex (AFC, *Graphium* spp. and *Paracremonium* spp.) could result from beetle attacks without the beetle establishing galleries or colonising the xylem. Inoculation of 30 cm detached healthy 2.54 cm woody shoots of avocado and seedlings with *Fusarium kuroshium*, *F. euwallaceae*, *Graphium euwallaceae, G. kuroshium* and *Paracremonium pembeum* resulted in visible lesions (necrotic tissue) with lengths that varied between 4 and 14 cm from 5 days to 4 weeks of incubation at 25℃ (Lynch et al. 2016; Na et al. 2018). *Paracremonium pembeum* produced longer lesions than *G. euwallaceae* (Lynch et al. 2016). Plants infected with *G. euwallaceae* showed lesions with an average length of 6.4 ± 2.8 cm, and those infected with *G. kuroshium* showed lesions with an average length of 7.6 ± 4.1 cm (Na et al. 2018). Avocado seedlings inoculated with *F. kuroshium* produced lesions with an average length of 3.15 ± 1.8 cm, and seedlings inoculated with *F. euwallaceae* produced lesions with an average length of 6.89 ± 2.1 cm (Na et al. 2018). Although not all fungi associated with the *Euwallacea fornicatus* species complex have been described and their pathogenicity studied, generally, fungi from the AFC isolated from avocado and other hosts show very similar symptoms of lesions and dieback (Kasson et al. 2013; Carrillo et al. 2016; Mendel et al. 2017; Aoki et al. 2019; Freeman et al. 2019; Aoki et al. 2021; Lynn et al. 2021). Thus, it is assumed that symptom expression would likely be visible in case of successful infection, especially during the first months of PEQ.

Survival of ambrosia fungi in healthy young avocado branches and seedlings: A long-term inoculation study of avocado branches with *Graphium euwallaceae* and *P. pembeum* showed that both fungi could not be recovered from live avocado tissue after 3–4 months or 2 months, respectively (Freeman et al. 2019). However, Freeman et al. (2019) recovered *G. euwallaceae* and *P. pembeum* from dead xylem of avocado and concluded that the fungi could survive long-term in dead xylem as opposed to live avocado tissue. Thus, after active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in Level 3A PEQ it is highly likely that the fungi *G. euwallaceae, G. kuroshium* and *Paracremonium pembeum* would not survive in live avocado tissue. This is in contrast to *F. euwallaceae* which could be recovered from live tissue after 25 months of inoculation (Freeman et al. 2019). However, in both natural infection and pathogenicity tests in avocado, *F. euwallaceae* and *F. kuroshium* could only be isolated from discoloured (light brown) xylem tissues or beetle galleries but not from adjacent healthy (white) xylem tissue close to the stained point (Na et al. 2018; Freeman et al. 2019). This suggests that only the primary ambrosia fungi of *Euwallacea* (AFC) could survive the PEQ period in live tissue of avocado, however, they would likely remain localised in discoloured (light brown) xylem tissues in close proximity of inoculation.

- ***What are the known environmental conditions conducive to symptom expression?***

Graphium euwallaceae has been shown to have a growth temperature range from 20–35℃ with an optimum growth at 25℃ (Lynch et al. 2016). *Paracremonium pembeum* has been shown to have a growth temperature range from 15–35℃ with an optimum growth at 30–35℃ (Lynch et al. 2016). Similarly, Na et al. (2018) showed a growth temperature range from 10–40℃ with optimal growth at 30℃ for *G. euwallaceae* and *G. kuroshium*. Similarly, *F. euwallaceae* and *F. kuroshium* had an optimal growth at 25℃ (Na et al. 2018; Freeman et al. 2019). Therefore, high temperatures > 25℃ would likely be conducive to symptom expression.

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

There is no evidence that *Fusarium euwallaceae*, *F. kuroshium, Graphium euwallaceae, G. kuroshium* and *Paracremonium pembeum* can cause asymptomatic infections and thus, there is no information on which plant part should be tested to maximise the detection in the case of asymptomatic infection. The fungi would likely be in close proximity of beetle galleries and beetle galleries have been found more frequently in small branches of avocado and close to the base of a branch, up to 30 cm from the branching points (Mendel et al. 2017). In both natural infection and pathogenicity tests in avocado, *F. euwallaceae* and *F. kuroshium* could only be isolated from discoloured (light brown) xylem tissues or beetle galleries but not from adjacent healthy (white) xylem tissue close to the stained point (Na et al. 2018; Freeman et al. 2019). Some uncertainty arises from the fact that not all AFC fungi (Table 5-2) isolated from avocado have been tested for pathogenicity.

- ***What is the optimum season for conducting detection testing?***

In Israel, beetle attacks and lesion frequency on avocado were highest during late summer, July to September (Mendel et al. 2017). Moreover, the growth of *Fusarium euwallaceae*, *F. kuroshium, Graphium euwallaceae, G. kuroshium* and *Paracremonium pembeum* has been shown highest at warmer temperatures > 25℃ (Lynch et al. 2016; Na et al. 2018). Thus, warm seasons have a higher potential for detecting the fungi.

5.2.9 References

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5.3 *Colletotrichum* spp.

This PRA assesses species associated with avocado found in two species complexes: the *Colletotrichum gloeosporioides* species complex and the *Colletotrichum acutatum* species complex. The following species will be assessed: *Colletotrichum aenigma*, *C. chrysophilum*, *C. endophyticum*, *C. jiangxiense*, *C. queenslandicum* and *C. tropicale* are in the *C. gloeosporioides* species complex and *C. nymphaceae* is in the *C. acutatum* species complex.

Colletotrichum species in both the *gloeosporioides* and *acutatum* species complexes are fungi with a wide host and environment range. They are important plant pathogens but can have endophytic and saprotrophic lifestyles. In avocado, *Colletotrichum* species cause anthracnose in fruit and sometimes stem dieback.

5.3.1 Taxonomic description

Scientific name: *Colletotrichum aenigma* B. S. Weir & P. R. Johnston (2012)

Taxonomic notes: *Colletotrichum aenigma* is part of the *gloeosporioides* species complex which is defined genetically on the basis of multi-gene phylogenies (Weir et al. 2012).

Scientific name: *Colletotrichum chrysophilum* W.A.S. Vieira, W. G. Lima, M. P. S. Câmara & V.P. Doyle (2017)

Taxonomic notes: *Colletotrichum chrysophilum* is part of the *gloeosporioides* species complex (Vieira et al. 2017) which is defined genetically on the basis of multi-gene phylogenies (Weir et al. 2012).

Scientific name: *Colletotrichum endophyticum* Manamgoda, Udayanga, L. Cai & K. D. Hyde (2013)

Other names include: *Colletotrichum endophytica* Manamgoda, Udayanga, L. Cai & K. D. Hyde (2013)

Taxonomic notes: *Colletotrichum endophyticum* is part of the *gloeosporioides* species complex (Manamgoda et al. 2013) which is defined genetically on the basis of multi-gene phylogenies (Weir et al. 2012).

Scientific name: *Colletotrichum jiangxiense* F. Liu and L. Cai (2015)

Taxonomic notes: *Colletotrichum jiangxiense* is part of the *gloeosporioides* species complex (Liu et al. 2015) which is defined genetically on the basis of multi-gene phylogenies (Weir et al. 2012).

Scientific name: *Colletotrichum nymphaeae* (Passerini) Aa (1978)

Other names include: *Colletotrichum mahoniae* Fabric. 1950 (Damm et al. 2012), *Ascochyta nymphaeae* Passerini 1876 (Damm et al. 2012), *Ramularia nymphaeae* Bres. (1894) (Damm et al. 2012), *Phyllosticta nymphaeae* (Passerini) Passerini (1888) (EPPO 2020)

Taxonomic notes: *Colletotrichum nymphaeae* is part of the *acutatum* species complex which includes destructive pathogens on fruits such as strawberry, citrus, apple, olive, cranberry and blueberry (Damm et al. 2012). A morphological feature of species within the *C. acutatum* species complex include the ‘acute ends’ of the conidia (Damm et al. 2012).

Scientific name: *Colletotrichum queenslandicum* B. S. Weir & P. R. Johnston (2012)

Other names include: *Colletotrichum gloeosporioides* var. *minus* Simmonds (1968) (EPPO 2020)

Taxonomic notes: *Colletotrichum queenslandicum* is part of the *gloeosporioides* species complex which is defined genetically on the basis of multi-gene phylogenies (Weir et al. 2012).

Scientific name: *Colletotrichum tropicale* Rojas, Rehner & Samuels (2010)

Taxonomic notes: *Colletotrichum tropicale* is part of the *gloeosporioides* species complex (Rojas et al. 2010) which is defined genetically on the basis of multi-gene phylogenies (Weir et al. 2012).

Order/Family: Glomerellales/Glomerellaceae

5.3.2 Hazard identification

Colletotrichum spp. (*C. aenigma, C. chrysophilum, C. endophyticum, C. jiangxiense, C. nymphaeae, C. queenslandicum, C. tropicale*) are not known to be present in New Zealand:

- *Colletotrichum chrysophilum*, *C. endophyticum* and *C. jiangxiense* are not listed in either Biota NZ (2022), NZOR (2021) or PPIN (2021)
- *Colletotrichum aenigma*, *C. nupharicola*, *C. queenslandicum* and *C. tropicale* are recorded as absent from New Zealand in Biota NZ (2022), NZOR (2021) and are not listed in PPIN (2021)
    - *Colletotrichum nymphaeae* has an organism report in PPIN (2021) stating it is not present in New Zealand
- *Colletotrichum aenigma, C. chrysophilum, C. endophyticum, C. jiangxiense, C. nymphaeae, C. queenslandicum, C. tropicale* are not listed in ONZPR (2021)

Colletotrichum spp. have the potential to establish (and spread) in New Zealand because:

- *Persea americana* (avocado) is a known host (Weir et al. 2012; Sharma et al. 2017; Fuentes-Aragón et al. 2020) that is widely grown in New Zealand, in home gardens and commercial production, mainly in the north of the North Island.
- *Colletotrichum* spp. cause anthracnose in avocado in regions around the world (e.g. Israel, Mexico) (Sharma et al. 2017; Fuentes-Aragón et al. 2020) that have a climate match index (CMI) ≥ 0.7 indicating a similar climate to the whole of New Zealand (Phillips et al. 2018).

Colletotrichum spp. have the potential to cause harm to New Zealand because:

- *Colletotrichum* spp. cause anthracnose in avocado which causes sunken necrotic black lesions on the fruit (Sharma et al. 2017). The fungus survives between fruiting cycles on leaves and twigs on trees (Sharma et al. 2017). Lesions would reduce marketability of fruits.
- avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$ 50.6 million (Plant & Food Research 2020)

Colletotrichum spp. is associated with *P. americana* budwood because:

- *Persea americana* is a known host of *Colletotrichum* spp. (Weir et al. 2012; Sharma et al. 2017; Fuentes-Aragón et al. 2020);
- *Colletotrichum* spp. have been isolated from branches and stems of *P. americana* (Sharma et al. 2017).

Given the arguments and evidence above, *Colletrotrichum* spp. meet the criteria to be hazards on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.3.3 Risk assessment

Biology and epidemiology of Colletotrichum spp.

Hosts and geographical distribution

The *Colletotrichum* spp. assessed in this pest risk analysis have a wide geographic and host range (Table 5-6).

Symptoms

Fruit anthracnose is the most common symptom of *Colletotrichum* infection (Dowling et al. 2020; Fuentes-Aragón et al. 2020). However, many species in the *C. glorosporioides* species complex infect leaves, crowns, stems, twigs and petioles (Sharma et al. 2017; Dowling et al. 2020). Infections of leaves, stems and twigs are commonly observed in apples and strawberries (Dowling et al. 2020). Symptom severity can vary among species within a species complex as well as among species complexes (Dowling et al. 2020).

In *P. americana* (avocado), the *Colletotrichum* spp. assessed in this PRA commonly cause fruit anthracnose (Sharma et al. 2017; Fuentes-Aragón et al. 2020). Initial symptoms include small necrotic black spots (< 1mm) on the exocarp (i.e., outer layer of skin of fruit) (Velázquez-del Valle et al. 2016; Fuentes-Aragón et al. 2020). These black spots become larger and more sunken over time (Fuentes-Aragón et al. 2020). In some cases, an orange, pink conidial mass will form on the surface of the lesions (Sharma et al. 2017). In severe infections, fruit drop occurs in early fruit development stages (Fuentes-Aragón et al. 2020). Symptoms on larger fruit reduce the quality of the fruit which are not suitable for export (Fuentes-Aragón et al. 2020).

In Israel, a study was conducted to determine *Colletotrichum* species associated with avocado trees (Sharma et al. 2017). Six orchards were surveyed from which 5 fruits, 5 twigs and 5 leaves were collected from each of 5 trees (450 samples). From these samples, 576 isolates of *Colletotrichum* species were collected. Of these isolates, 94.88% were isolated from fruit, 19.87% were isolated from green leaves, 18% from dry leaves and 10.93% from green twigs (Sharma et al. 2017). Low infection values of 0.9% were isolated from dry and dead twig tissues (Sharma et al. 2017). Collected plant samples (fruit, fresh leaves, twigs) produced anthracnose symptoms after 7–10 days when maintained under humid conditions (Sharma et al. 2017).

Inoculated avocado fruits produced necrotic lesions at 4-days post inoculation (dpi) and the quality of the fruits was visibly affected at 7 dpi (Fuentes-Aragón et al. 2020; Ayvar-Serna et al. 2021). Members of the *C. gloeosporioides* species complex appear to be more virulent than members in other species complexes (i.e., the *acutatum* species complex), producing lesions between 29.1 mm to 34.6 mm in length (Fuentes-Aragón et al. 2020). Species in the *gloeosporioides* complex tend to show high sporulation at 7 dpi in comparison to members in other complexes (Fuentes-Aragón et al. 2020).

The majority of the *Colletotrichum* species assessed in this analysis have been recorded to cause fruit rot in avocado (i.e., *C. nymphaceae*, *C. tropicale*, *C. chrysophilum*, *C. endophyticum*, *C. jiangxiense*) (Shivas et al. 2016; Sharma et al. 2017; Vieira et al. 2017; Fuentes-Aragón et al. 2020). Two species, *C. aenigma* (Sharma et al. 2017) and *C. queenslandicum* (Shivas et al. 2016) have been recorded on stems, however, description of symptoms on stems is lacking. *Colletotrichum aenigma* has also been recorded producing leaf spots (Sharma et al. 2017).

Latency of symptoms

Symptoms do not always appear when plants are colonised with *Colletotrichum* species. For example, *C. acutatum* has been observed germinating and sporulating on symptomless strawberry and blueberry leaves (Dowling et al. 2020). Symptoms can be delayed as survival of inoculum of the pathogen in the form of quiescent germinated appressoria on leaves and twigs may occur, especially during dry, summer seasons (Sharma et al. 2017). Quiescent infections on immature fruit are also common (Dowling et al. 2020). Spores can remain dormant on fruit surface until conditions that favour infection (Dowling et al. 2020).

Some *Colletotrichum* spp. have the ability to be endophytic (e.g. *C. endophyticum* and *C. tropicale*) (Rojas et al. 2010; Manamgoda et al. 2013), however there is debate whether stem-end rot pathogens (i.e. anthracnose disease causing organisms) are endophytes living in the stems, branches and pedicels of the avocado canopy which grow into the fruits (Hartill et al. 2002). It is most likely anthracnose pathogens are symptomless phellophytes (i.e. fungi growing in bark and phellem tissue) (Hartill et al. 2002). Endophytes would have the ability to grow down xylem elements whereas phellophytes would require to break through cell barriers before entering the xylem (Hartill et al. 2002). In New Zealand, treatment of avocado fruit (with their pedicel) with a systemic fungicide (Benlate) reduced fruit rot in comparison to fruit treated with a contact fungicide (Captan), suggesting a possibility that infections may be endophytic (Hartill et al. 2002).

Reproduction and transmission

Conidia of *Colletotrichum* species are transmitted via water-splash (Cannon et al. 2012; Dowling et al. 2020) and can be produced on all host tissues (Ploetz 2003). Infection occurs via a germinating spore on the surface of a plant producing an appressorium and germination is favoured by warm and moist conditions (Dowling et al. 2020). The appressorium penetrates the plant cuticle and sometimes epidermal cells of the plant (Cannon et al. 2012). For example, fruits are usually the main plant part that becomes infected, as recorded for *C. acutatum* (*acutatum* species complex) and infection usually occur after ripening when the appressorium penetrates the host (Ploetz 2003). New leaf flushes also tend to be most susceptible (Ploetz 2003).

There are slight differences between *C. acutatum* (within the *acutatum* species complex) and *C. gloeosporioides* (within the *gloeosporioides* species complex) on their optimal growth temperatures, infection, reproduction and overwintering (Table 5-5). These species are used as representative of their species complexes (Dowling et al. 2020).

Table 5-5: A comparison between *Colletotrichum gloeosporioides* and *C. acutatum* based on key characteristics of their life cycle and life stages (adapted from Dowling et al. 2020).

Species	*Colletotrichum gloeosporioides*	*Colletotrichum acutatum*
Spread	Rainsplash and wind	Rainsplash
Optimal temperature	30°C	25°C
Infection	Best known for infecting vegetative tissue but can also infect fruit	Best known for infecting fruit but can also infect vegetative tissue
Reproduction	Both sexual and asexual stages found in the field	Only asexual stage found in the field
Overwintering	On dead wood, plant debris and alternate hosts	On mummified fruit, infected twigs and symptomless transplants

In addition to overwintering on plant debris, conidia may also lie dormant in the soil (EPPO 2021). It is the sexual stage of *Colletotrichum* that occurs more frequently on plant debris (Dowling et al. 2020). Dead leaves within a tree canopy can be a main source of inoculum, as described for *C. gloeosporioides* (within the *gloeosporioides* species complex) (Jeffries et al. 1990).

The majority of *Colletotrichum* species have a hemibiotrophic infection strategy: the fungus develops biotrophic hyphae inside the host which later transitions to necrotrophic secondary mycelia (Lu et al. 2018). The biotrophic phase results in infected tissue remaining externally symptomless (Cannon et al. 2012) and symptoms may only appear after harvest.

5.3.4 Likelihood of entry

Colletotrichum species have a wide distribution (Table 5-6). Species have been recorded in areas where avocado is produced including Australia (Weir et al. 2012; Shivas et al. 2016), Mexico (Fuentes-Aragón et al. 2020), Israel (Sharma et al. 2017) and Sri Lanka (Adikaram and Yakandawala 2020). In these regions, *Colletotrichum* species cause fruit anthracnose. In Australia, *C. queenslandicum* has caused stem dieback (Shivas et al. 2016).

Infection of *Colletotrichum* in avocado is mainly associated with fruit lesions and necrosis (Fuentes-Aragón et al. 2020), but has been isolated from leaves, twigs and stems (Sharma et al. 2017; MPI 2021). Therefore, *Colletotrichum* species could be associated with leafless, rootless avocado budwood cuttings.

Symptoms can be delayed as survival of inoculum of the pathogen in the form of quiescent germinated appressoria on leaves and twigs may occur, especially during dry, summer seasons. Rain and elevated humidity increase levels of rot symptoms (Sharma et al. 2017). For example, samples from different parts of avocado trees in orchards (fruits, twigs and leaves) were collected and maintained in a moist chamber at room temperature (20–25℃); anthracnose symptoms were observed after 7–10 days under these humid conditions (Sharma et al. 2017).

It is not known if any of the *Colletotrichum* species listed here could survive as endophytes on avocado budwood in PEQ without causing symptoms. Some of the species assessed here have been recorded as endophytes in other plant species. *Colletotrichum tropicale* has been recorded as an endophyte from a leaf of *Theobroma cacao* (Rojas et al. 2010). *Colletotrichum endophyticum* has also been recorded as an endophyte in the grass species *Pennisetum purpureum* (Manamgoda et al. 2013). Given the endophytic abilities of these *Colletotrichum* species on plants hosts other than avocado, there is some uncertainty regarding their endophytic capabilities in avocado budwood. Furthermore, depending on the temperature and relative humidity within PEQ, and in the absence of overhead irrigation to simulate rainfall, the appropriate environmental conditions for symptom development may be absent throughout the entire PEQ period. However, it has been recorded that new leaf flushes tend to be the most susceptible to infection (Ploetz 2003).

The routine surface disinfection of budwood entering PEQ (dipping in 1% sodium hypochlorite for 2 minutes) will reduce the surface population of the *Colletotrichum* species but is not likely to completely eliminate it, especially if fungi are located deep within crevices of buds. Furthermore, some *Colletotrichum* species seem to only express symptoms when fruits begin to ripen on plants (Sharma et al. 2017). Therefore, it is possible that a small population of fungi may survive asymptomatically throughout the PEQ period and enter New Zealand.

Given the arguments and evidence above, that is:

- *Colletotrichum* infection under the right conditions (in a moist chamber at room temperature 20–25℃) can produce symptoms in a few days (Sharma et al. 2017), but
- uncertainty around the endophytic capabilities of some *Colletotrichum* species in *P. americana* budwood may allow asymptomatic *Colletotrichum* infection during the PEQ period,

the likelihood of *Colletotrichum* species entering New Zealand associated with *P. americana* budwood is considered to be low, with moderate uncertainty.

5.3.5 Likelihood of exposure

This assessment is made on the basis that *Colletotrichum* species have entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *Colletotrichum* species.

If avocado plants are infected with *Colletotrichum* at the time of their release from PEQ they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *Colletotrichum* in New Zealand from *P. americana* budwood is considered to be high, with low uncertainty.

5.3.6 Likelihood of establishment

This assessment is made on the basis that *Colletotrichum* species have been successfully exposed to a suitable host plant in the New Zealand environment.

Colletotrichum has a broad host range (Table 5-6) and many of its hosts are either grown commercially in New Zealand (e.g., kiwifruit, apples, strawberries, grapes) or as backyard plants in many home gardens. Therefore, hosts for the establishment of *Colletotrichum* are readily available.

Colletotrichum species are also found across a broad geographic range from countries (Table 5-6) that have a similar climate to the whole of New Zealand, as well as to the Auckland and Northland regions, as indicated by a climate match index of ≥0.7 (Phillips et al. 2018). For example, seasonal climate ellipses show considerable overlap between avocado growing areas in Mexico, Israel and New Zealand, although Mexico sites have overall warmer average monthly temperatures (Halloy 2021). Therefore, climate suitable for the establishment of the *Colletotrichum* is readily available, especially in the north of the North Island. In addition, because wetter conditions favour the establishment of *Colletotrichum* species (Sharma et al. 2017), and conditions in New Zealand are wetter, on average, compared to those of Mexico and Israel (Halloy 2021), New Zealand conditions are likely to be more favourable for the establishment of the *Colletotrichum* species.

Given the arguments and evidence above, that is:

- hosts (in the form of cultivated avocados and other cultivated crops) are available for the establishment of *Colletotrichum* species
- suitable climate (Table 5-6) is available for the establishment of *Colletotrichum* (Phillips et al. 2018)

the likelihood of *Colletotrichum* establishing in New Zealand is considered to be high, with low uncertainty.

Table 5-6: Geographic and host range of *Colletotrichum* spp. Climate match index (CMI) ≥ 0.7 indicates countries with a similar climate to the whole of the New Zealand (Phillips et al. 2018).

Region	Country	CMI	Species complex	Species	Host	Reference
Africa	Kenya	0.4–0.8	*acutatum*	*C. nymphaeae*	*Fragaria vesca*	Damm et al. (2012)
	South Africa	0.5–0.9	*acutatum*	*C. nymphaeae*	*Fragaria x ananassa*	Damm et al. (2012)
					Protea cynaroides	Damm et al. (2012)
					Protea magnifica	Damm et al. (2012)
					Protea repens	Damm et al. (2012)
	Zimbabwe	0.5–0.7	*acutatum*	*C. nymphaeae*	*Capsicum annuum*	Damm et al. (2012)
Asia	China	0.4–0.9	*acutatum*	*C. nymphaeae*	*Fragaria* sp.	Dowling et al. (2020)
			gloeosporioides	*C. aenigma*	*Malus* sp.	Dowling et al. (2020)
					Fragaria sp.	Dowling et al. (2020)
					Vitis sp.	Dowling et al. (2020)
				C. tropicale	*Coffea arabica*	Cao et al. (2019)
					Areca catechu	Cao et al. (2020)
				C. endophyticum	*Camellia sinensis*	Wang et al. (2016)
					Mangifera indica	Li et al. (2019)
				C. jiangxiense	*Camellia sinensis*	Liu et al. (2015)
	India	0.2–0.5	*acutatum*	*C. nymphaeae*	*Pelargonium graveolens*	Damm et al. (2012)
	Indonesia	0.3–0.6	*acutatum*	*C. nymphaeae*	*Capsicum* sp.	Damm et al. (2012)
	Japan	0.6–0.8	*gloeosporioides*	*C. aenigma*	*Pyrus pyrifolia*	Weir et al. (2012)

Region	Country	CMI	Species complex	Species	Host	Reference
					Malus sp.	Dowling et al. (2020)
					Fragaria sp.	Dowling et al. (2020)
				C. tropicale	*Litchi chinensis*	Weir et al. (2012)
			acutatum	*C. nymphaeae*	*Malus* sp.	Dowling et al. (2020)
					Vaccinium corymbosum	Dowling et al. (2020)
	Korea	0.6–0.7	*acutatum*	*C. nymphaeae*	*Malus* sp.	Dowling et al. (2020)
	Philippines	0.3–0.6	*gloeosporioides*	*C. tropicale*	*Mangifera indica*	Dela Cueva et al. (2021)
	Sri Lanka	0.4–0.6	*acutatum*	*C. nymphaeae*	*Hevea brasiliensis*	Adikaram and Yakandawala (2020)
			gloeosporioides	*C. endophyticum*	*Persea americana*	Adikaram and Yakandawala (2020)
	Thailand	0.3–0.5	*acutatum*	*C. nymphaeae*	Leaf litter	Damm et al. (2012)
			gloeosporioides	*C. endophyticum*	*Pennisetum purpureum*	Manamgoda et al. (2013)
Central America	Costa Rica	0.3–0.6	*acutatum*	*C. nymphaeae*	*Fragaria* sp.	Dowling et al. (2020)
	Cuba	0.5	*gloeosporioides*	*C. tropicale*	*Annona cherimola*	Garcia and Manzano (2017)
	Panama	0.4–0.6	*gloeosporioides*	*C. tropicale*	*Theobroma cacao*	Rojas et al. (2010)
					Trichilia tuberculata	Rojas et al. (2010)
					Viola surinamensis	Rojas et al. (2010)
					Cordia aliodora	Rojas et al. (2010)
					Annona muricata	Rojas et al. (2010)
				C. chrysophilum	*Genipa americana*	Vieira et al. (2017)
					Theobroma sp.	Vieira et al. (2017)
	Puerto Rico	0.5–0.6	*gloeosporioides*	*C. chrysophilum*	*Mycopteris taxifolia*	Vieira et al. (2017)
Europe	Belgium	0.9	*acutatum*	*C. nymphaeae*	*Malus* sp.	Dowling et al. (2020)
					Fragaria sp.	Dowling et al. (2020)
	Bulgaria	0.8–0.9	*acutatum*	*C. nymphaeae*	*Fragaria x ananassa*	Damm et al. (2012)
	France	0.9	*acutatum*	*C. nymphaeae*	*Fragaria x ananassa*	Damm et al. (2012)
	Italy	0.7–0.9	*acutatum*	*C. nymphaeae*	*Anemone* sp.	Damm et al. (2012)
					Fragaria x ananassa	Damm et al. (2012)
					Mahonia aquifolium	Damm et al. (2012)
	Netherlands	0.9	*acutatum*	*C. nymphaeae*	*Anemone* sp.	Damm et al. (2012)
					Anemone coronaria	Damm et al. (2012)
					Fragaria x ananassa	Damm et al. (2012)
					Nuphar luteum	Damm et al. (2012)
					Nymphaea alba	Damm et al. (2012)
					Oenothera sp.	Damm et al. (2012)
					Phaseolus sp.	Damm et al. (2012)
	Portugal	0.8–0.9	*acutatum*	*C. nymphaeae*	*Olea europaea*	Damm et al. (2012)
					Fragaria sp.	Dowling et al. (2020)
	Spain	0.7–0.9	*acutatum*	*C. nymphaeae*	*Fragaria x ananassa*	Damm et al. (2012)

Region	Country	CMI	Species complex	Species	Host	Reference
	Switzerland	0.7–0.9	*acutatum*	*C. nymphaeae*	*Fragaria* sp.	Dowling et al. (2020)
	United Kingdom	0.8–0.9	*acutatum*	*C. nymphaeae*	*Fragaria vesca*	Damm et al. (2012)
					Photinia sp.	Damm et al. (2012)
			gloeosporioides	*C. aenigma*	*Fragaria* sp.	Dowling et al. (2020)
Middle East	Iran	0.4–0.8	*acutatum*	*C. nymphaeae*	*Fragaria* sp.	Dowling et al. (2020)
	Israel	0.5–0.7	*acutatum*	*C. nymphaeae*	*Anemone* sp.	Damm et al. (2012)
					Fragaria x ananassa	Damm et al. (2012)
			gloeosporioides	*C. aenigma*	*Persea americana*	Sharma et al. (2017)
				C. chrysophilum	*Persea americana*	Sharma et al. (2017); Khodadadi et al. (2020)
				C. nymphaeae	*Fragaria x ananassa*	Damm et al. (2012)
North America	Canada	0.2–0.9	*acutatum*	*C. nymphaeae*	*Leucaena* sp.	Damm et al. (2012)
	Mexico	0.3–0.8	*acutatum*	*C. nymphaeae*	*Persea americana*	Fuentes-Aragón et al. (2020)
				C. tropicale	*Origanum vulgare*	Ayvar-Serna et al. (2020)
			gloeosporioides	*C. tropicale*	*Persea americana*	Fuentes-Aragón et al. (2020)
				C. chrysophilum	*Persea americana*	Fuentes-Aragón et al. (2020)
				C. jiangxiense	*Persea americana*	Fuentes-Aragón et al. (2020)
				C. nymphaeae	*Fragaria* sp.	Damm et al. (2012)
	USA	0.5–0.8	*acutatum*	*C. nymphaeae*	*Malus* sp.	Dowling et al. (2020)
					Prunus persica	Dowling et al. (2020)
					Fragaria sp.	Dowling et al. (2020)
			gloeosporioides	*C. chrysophilum*	*Malus domestica*	Khodadadi et al. (2020)
				C. tropicale	*Malus* sp.	Dowling et al. (2020)
				C. nymphaeae	*Protea* sp.	Damm et al. (2012)
				C. nymphaeae	*Fragaria* sp.	Shivas and Tan (2009)
			gloeosporioides	*C. queenslandicum*	*Carica papaya*	Weir et al. (2012)
					Carica sp.	Weir et al. (2012)
					Persea americana	Weir et al. (2012)
	Fiji	0.4	*acutatum*	*C. tropicale*	*Annona* sp.	Costa et al. (2019)
			gloeosporioides	*C. queenslandicum*	*Coffea* sp.	Weir et al. (2012)
South America	Brazil	0.4–0.9	*gloeosporioides*	*C. chrysophilum*	*Musa* sp.	Vieira et al. (2017)
			acutatum	*C. nymphaeae*	*Vitis* sp.	Dowling et al. (2020)
					Malus pumila	Damm et al. (2012)
	Colombia	0.3–0.8	*acutatum*	*C. nymphaeae*	*Fragaria* sp.	Dowling et al. (2020)

5.3.7 Impacts in New Zealand

Many *Colletotrichum* species in the *acutatum*, *boninense* and *gloeosporioides* species complexes are already present in New Zealand (e.g., *C. alienum*, *C. aotearoa*, *C. cigarro*, *C. fioriniae*, *C. fructicola*, *C. karstii*, *C. perseae* and *C. siamense*) (Hofer et al. 2021). These species have been isolated from avocado producing fruit staining symptoms known as "tannin stain" (Hofer et al. 2021). In addition, *C. acutatum* and *C. glorosporioides* have been known to occur in avocado orchards since the 1980s causing anthracnose in fruit (Hartill 1991). These *Colletotrichum* species are also successful postharvest pathogens because of latent infections; the majority of infection occurs while fruit is stored or is on market shelves (Dean et al. 2012).

Management of *Colletotrichum* in avocado orchards are in place, however, species have different sensitivities to fungicides (Hartill 1991; Hofer et al. 2021). For example, benomyl is effective against *C. gloeosporioides* but not *C. acutatum* (Hartill 1991; Everett and Korsten 1998). In New Zealand, a combination of copper fungicide sprays and periodic removal of dead branches and twigs within the canopy of orchards decreases the overall incidence of postharvest rots of avocado (Hartill 1991).

Economic impacts

Given the broad host range of the *Colletotrichum* species in this assessment, it is likely that avocados would not be the only commercially produced hosts affected by these pathogens (Table 5-6). Outbreaks of these species can be devastating. For example, bitter-rot of apple caused by *C. acutatum* and *C. gloeosporioides* species complexes can result in the loss of up to 50% of crops in optimal conditions (González and Sutton 2004; Dowling et al. 2020). In some cases, greater than 75% tree defoliation is observed by harvest time which weakens trees and reduces yield (González and Sutton 2004). Before the use of synthetic fungicides, a bitter-rot outbreak in the United States resulted in losses of $400 million (in today's currency) (Dowling et al. 2020). Similarly, epidemics of *C. acutatum* causing anthracnose in strawberries has been observed in Florida and North Carolina (Delp 1980), Israel (Freeman and Katan 1997), Brazil (Henz 1991) and Bulgaria (Jelev et al. 2008). In Bulgaria, disease incidence reached 90% in some regions and percentage of diseased plants averaged 50% in some fields (Jelev et al. 2008). Some of these outbreaks have occurred despite the use of fungicide control measures (Dowling et al. 2020).

Colletotrichum acutatum is present in New Zealand (Biota NZ 2022) but other species in the *acutatum* species complex that are assessed here have also been recorded on strawberries and are not present in New Zealand (e.g. *C. nymphaeae*). Some *Colletotrichum* species are resistant to specific fungicides and in some cases, fungicide resistance limits chemical management options for diseases caused by *Colletotrichum* species (Dowling et al. 2020); this fungicide resistance has mainly been observed in the USA with *C. acutatum* (EPPO 2021). However, there is variability among species even within the same species complex. For example, *C. nymphaeae* (*C. acutatum* species complex) show moderate sensitivity to benomyl fungicide in culture (i.e., percentage of mycelial growth inhibition ranged from 33–71%) whereas species within the *C. gloeosporioides* species complex (not those assessed in this PRA) show high sensitivity to benomyl fungicide (i.e., percentage of mycelial growth inhibition is > 94%) (Moral et al. 2021). A captan-benomyl mixture has been used in strawberry crops in New Zealand (EPPO 2021) indicating that some species of *Colletotrichum* assessed in this PRA may have varying responses to fungicides used in New Zealand.

Specifically in avocados, 90% of avocado trees in backyards in the Leonardo Bravo municipality in Guerrero, Mexico had symptoms of fruit anthracnose caused by *C. jiangxiense* (Ayvar-Serna et al. 2021). Anthracnose outbreaks occur annually in Mexico, mainly in areas of high relative humidity and temperatures (Fuentes-Aragón et al. 2020).

In 2020, the export value for apples was $876.3 million with a much smaller proportion sold domestically (Plant & Food Research 2020). The export values for avocado was $112.3 million and the domestic value was $50.6 million (Plant & Food Research 2020). Strawberries had an export value of $6.2 million and a domestic value of $27.5 million (Plant & Food Research 2020). The *Colletotrichum* species assessed in this document are not listed as a quarantine pest according to the importing countries phytosanitary requirements (ICPR) and thus there would be no immediate impacts on export pathways.

Given the arguments and evidence above, that is:

- some species in both *C. acutatum* and *C. gloeosporioides* species complexes are already present in New Zealand and cause anthracnose symptoms (Hofer et al. 2021),
- appropriate in-field fungicides and management options are used in New Zealand to control for anthracnose symptoms (Hartill 1991),
- fungicide resistance has been reported for some *Colletotrichum* species (Dowling et al. 2020; Hofer et al. 2021) and *C. nymphaceae* demonstrate moderate sensitivity to benomyl fungicide,
- the host range of *Colletotrichum* indicates not only the avocado but also the apple and strawberry industry could be affected which together are valued at $1.07 billion (sum of both domestic and export),

the economic impact of *Colletotrichum* to New Zealand is considered to be moderate, with low uncertainty.

Environmental impacts

It is unknown if any of the *Colletotrichum* species assessed here have known native New Zealand plant hosts. However, other species in the *acutatum* and *gloeosporioides* species complexes are known to associate with native New Zealand plant species. For example, *C. acutatum* has been isolated from *Coprosma lucida* and *Coprosma macrocarpa* (Biota NZ 2022). *Colletotrichum gloeosporioides* has been isolated from *Kunzea ericoides* as a leaf endophyte (Biota NZ 2022).

Given the possibility of *Colletotrichum* species living as pathogens, saprotrophs and endophytes on plants and in the soil, this group of fungi could have a wide range of environmental impacts. Living as endophytes in native hosts may allow *Colletotrichum* to survive before spilling over into economically important crop species or vice versa (Power and Mitchell 2004).

Given the arguments and evidence above, the impact on the environment from the establishment of *Colletotrichum* in New Zealand is considered to be low, with moderate uncertainty.

Human health impacts

There has been one record of a *Colletotrichum* species in the *gloeosporioides* species complex with the potential to cause human health impacts. The study demonstrated, in mice, that *Camellia sinensis* infected with the endophyte *Colletotrichum gloeosporioides* damaged renal function and that increased attention is needed in tea processing (Li et al. 2017). However, there is no evidence that the *Colletotrichum* species assessed here cause human health impacts.

Given the arguments and evidence above, the human health impact of *Colletotrichum* to New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

Anthracnose diseases caused by *Colletotrichum* species are mostly evident after harvest and in storage and on shelves in markets (Dean et al. 2012). If left unmanaged *Colletotrichum* infected fruits could cause declines in edible fruit in markets. Avocado has become popular and is widely consumed in New Zealand. It is commonly grown in home gardens in some parts of the country. A similar impact could be seen in apples and strawberries.

Given the arguments and evidence above, the sociocultural impact of *Colletotrichum* to New Zealand is considered to be low, with low uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, human health and society is considered to be moderate, with moderate uncertainty.

5.3.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is low
- assuming entry, the likelihood of exposure is high
- assuming successful exposure, the likelihood of establishment is high
- the overall impact on the New Zealand economy, environment, human health and society is considered to be moderate

the overall level of assessed risk to New Zealand from *Colletotrichum* on imported *Persea americana* budwood (as in the commodity description) is considered to be moderate, with moderate uncertainty.

5.3.9 Management considerations

Colletotrichum spp. cause fruit anthracnose in *P.a americana* (avocado). Initial symptoms include small necrotic black spots (< 1mm) on the exocarp (i.e., outer layer of skin of fruit) (Velázquez-del Valle et al. 2016; Fuentes-Aragón et al. 2020). These black spots become larger and more sunken over time (Fuentes-Aragón et al. 2020). In some cases, an orange, pink conidial mass will form on the surface of the lesions (Sharma et al. 2017). In severe infections, fruit drop occurs in early fruit development stages (Fuentes-Aragón et al. 2020). Symptoms on larger fruit reduce the quality of the fruit which are not suitable for export (Fuentes-Aragón et al. 2020). *Colletotrichum aenigma* has also been recorded producing leaf spots (Sharma et al. 2017).

Many species in the *C. glorosporioides* species complex infect leaves, crowns, stems, twigs and petioles (Dowling et al. 2020) and thus, lesions may appear on these plant parts; however, there is limited evidence of these symptoms on avocado.

Rain and elevated humidity increase levels of rot symptoms (Sharma et al. 2017). High temperatures (i.e. 25 – 30℃) are also optimal for *Colletotrichum* growth (Dowling et al. 2020).

PCR tests are readily available for *Colletotrichum* species. The ITS gene region, commonly used for fungal identification, can distinguish among *Colletotrichum* species complexes; however, specific species identification is difficult and requires a multigene sequencing approach (Shivas et al. 2016; Dowling et al. 2020). Shivas et al. (2016) highlights specific primers that cover four gene regions used to identify individual *Colletotrichum* species.

5.3.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Many species in the *C. glorosporioides* species complex infect leaves, crowns, stems, twigs and petioles (Dowling et al. 2020) and thus, lesions may appear on these plant parts; however, there is limited evidence of these symptoms on avocado. *Colletotrichum aenigma* has been recorded producing leaf spots (Sharma et al. 2017). Symptoms are most likely to appear of fruit, which are unlikely to be present on plants in PEQ.

- ***What are the known environmental conditions conducive to symptom expression?***

Rain and elevated humidity increase levels of rot symptoms (Sharma et al. 2017). High temperatures (i.e. 25 – 30℃) are also optimal for *Colletotrichum* growth (Dowling et al. 2020).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Fruit is most commonly associated with *Colletotrichum* infections; however, this pathogen has been isolated from leaves, twigs and stems of avocado (Shivas et al. 2016; Sharma et al. 2017). Sharma et al. (2017) collected samples from these areas of the plant, many of which were asymptomatic. These samples were placed in a moist chamber at room temperature where they were inspected for the appearance of anthracnose symptoms (Sharma et al. 2017). Anthracnose symptoms were observed after 7–10 days in the collected fruits, leaves and twigs maintained under these humid conditions (Sharma et al. 2017).

- ***What is the optimum season for conducting detection testing?***

Optimum season would be during warm (i.e., 25–30℃), humid periods which provide the optimal conditions for the growth of *Colletotrichum*

5.3.11 References

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5.4 *Clonostachys pseudochroleuca* (avocado branch dieback and wilting)

Clonostachys pseudochroleuca is a fungus that is associated with a wide range of plants and environments. It is commonly found as a saprotroph or endophyte but has been recorded as a pathogen in avocado, vectored by ambrosia beetles, causing avocado branch dieback and wilting.

5.4.1 Taxonomic description

Scientific name: *Clonostachys pseudochroleuca* Schroers (2001)

Order/Family: Hypocreales/Bionectriaceae

Other names include: *Bionectria pseudochroleuca* Schroers & Samuels (2001)

Taxonomic notes: *Bionectria pseudochroleuca* is the teleomorph and *Clonostachys pseudochroleuca* is the anamorph stage. *Clonostachys pseudochroleuca* is the preferred name (Robert et al. 2005; Rossman et al. 2013).

5.4.2 Hazard identification

Clonostachys pseudochroleuca is not known to be present in New Zealand:

- *Clonostachys pseudochroleuca* is not listed in either Biota NZ (2022), NZOR (2021) or PPIN (2021).
- *Clonostachys pseudochroleuca* is not listed in ONZPR (2021).

Clonostachys pseudochroleuca has the potential to establish (and spread) in New Zealand because:

- *Persea americana* (avocado) is a known host (Burbano-Figueroa et al. 2018) that is widely grown in New Zealand, in home gardens and commercial production, mainly in the north of the North Island.
- *Clonostachys pseudochroleuca* causes branch dieback and wilting in tropical and subtropical regions in Columbia (Burbano-Figueroa et al. 2018). Regions in Columbia have a climate match index (CMI) ≥ 0.7 indicating a similar climate to the whole of New Zealand (Phillips et al. 2018).
- *Clonostachys pseudochroleuca* is vectored by *Xylosandrus morigerus*, *Xyleborus volvulus*, and *Premnobius cavipennis* (Burbano-Figueroa et al. 2020), none of which are present in New Zealand. However, other species in *Xylosandrus* are present in New Zealand, such as the recently established *X. crassiusculus* (Ministry for Primary Industries 2020). However, it is not known whether *X. crassiusculus* is a vector for *C. pseudochroleuca* as this ambrosia beetle has not been recovered from infected trees.

Clonostachys pseudochroleuca has the potential to cause harm to New Zealand because:

- *Clonostachys pseudochroleuca* and their associated ambrosia beetles cause avocado branch dieback and wilting (Burbano-Figueroa et al. 2018). Infection by *C. pseudochroleuca* is associated with ambrosia beetle entry points and galleries which result in damage and discolouration on branches and trunks of avocado trees and wilting of trees.
- avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$50.6 million (Plant & Food Research 2020).

Clonostachys pseudochroleuca is associated with *Persea americana* budwood because:

- *Persea americana* is a known host of *Clonostachys pseudochroleuca* (Burbano-Figueroa et al. 2018);
- *Clonostachys pseudochroleuca* infects branches and stems of *P. americana*.

Given the arguments and evidence above, *Clonostachys pseudochroleuca* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.4.3 Risk assessment

Biology and epidemiology of Clonostachys pseudochroleuca

Host range

Species of *Clonostachys* are common soil inhabitants, plant decomposers and endophytes found in tropical and subtropical regions (Abreu et al. 2014). Some members are also antagonists of other fungi and oomycetes including *Botrytis cinerea*, *Fusarium oxysporum*, *Rhizoctonia solani*, *Phytophthora palmivora* and *Pythium ultimum* (Abreu et al. 2014). Selected strains of *Clonostachys* have been developed as biocontrol agents on a commercial scale (Abreu et al. 2014).

Clonostachys pseudochroleuca (teleomorph: *Bionectria pseudochroleuca*) is a common soil fungus but has been isolated as a pathogen (Burbano-Figueroa et al. 2018), endophyte (da Silva et al. 2021), saprotroph (Schroers 2001; Moreira et al. 2016; Huanraluek et al. 2020) and mycoparasite (da Silva et al. 2021) on a variety of hosts (Huanraluek et al. 2020) (Table 5-7).

Burbano-Figueroa et al. (2018) recorded *Clonostachys pseudochroleuca* as pathogenic on avocado (West-Indian race, *Persea americana* var. *americana*) causing avocado branch dieback and wilting disease in association with ambrosia beetles. This fungus has been recorded as a pathogen on avocado only (Burbano-Figueroa et al. 2018); it is more commonly attributed to saprotrophic or endophytic habitats in other hosts (Schroers 2001) (Table 5-7).

Table 5-7: Known host range and geographic distribution of Clonostachys pseudochroleuca

Continent	Country	Host	Plant part	CMI	Reference
Asia	Thailand	*Prunus* sp.	Dead branch	0.3-0.5	Huanraluek et al. (2020)
South America	Brazil	Soil	-	0.3-0.9	Moreira et al. (2016)
	Brazil	*Phoradendron perrottetii*		0.3-0.9	da Silva et al. (2021)
	Brazil	*Lychnophora pinaster*		0.3-0.9	da Silva et al. (2021)
	Brazil	*Solanum tuberosum*		0.3-0.9	Abreu et al. (2014)
	Brazil	*Saccharum officinarum*		0.3-0.9	Abreu et al. (2014)
	Colombia	*Persea americana*	Trunk and branches	0.3-0.8	Burbano-Figueroa et al. (2018)
	French Guiana	Palm tree	Palm sheathing base	0.4	Schroers (2001)
	French Guiana	Decaying wood		0.4	Schroers (2001)
	French Guiana	*Hypoxylon* sp.		0.4	Schroers (2001)
	Guyana	*Mauritia* sp.	Decaying trunk	0.4-0.6	Schroers (2001)
	Peru	*Vitis vinifera*	Rootstock	0.4-0.8	Torcato et al. (2020)
	Peru	*Vaccinium corymbosum*	Twigs and stems	0.4-0.8	Torcato et al. (2020)
	Venezuela	Palm tree	Sheath	0.3-0.8	Schroers (2001)
Oceania	Papua New Guinea	Soil		0.3-0.7	Schroers (2001)
	Australia	-	-	0.4-1	Schroers (2001)
Central America	Puerto Rico	*Guarea* sp	Bark of dead tree	0.5-0.6	Schroers (2001)
	Puerto Rico	*Nectriopsis* sp.	Bark	0.5-0.6	Schroers (2001)

Vectors

Clonostachys pseudochroleuca infection in avocado is associated with and vectored by ambrosia beetles including *Xylosandrus morigerus*, *Xyleborus volvulus*, and *Premnobius cavipennis* (Burbano-Figueroa et al. 2020). These ambrosia beetles are commonly associated with forests and agroforestry habitats in the Neotropics (Burbano-Figueroa et al. 2020) and are not present in New Zealand (NZOR 2021; PPIN 2021; Biota NZ 2022). *Xylosandrus morigerus* is regulated and an unwanted pest in New Zealand (ONZPR 2021). Other species in *Xylosandrus* are present in New Zealand, such as the recently established *X. crassiusculus* (Ministry for Primary Industries 2020). However, it is not known whether *X. crassiusculus* is a vector for *C. pseudochroleuca* as this ambrosia beetle has not been recovered from infected trees.

Symptoms

Clonostachys pseudochroleuca causes avocado branch dieback and wilting (Figure 5-1). Avocado trees exhibit signs of damage by boring insects in trunks and branches, in addition to branch dieback and wilting (Burbano-Figueroa et al. 2020). White powdery exudate is observed on the outer bark associated with beetle holes on the trunk and main branches of trees (Burbano-Figueroa et al. 2018). In old trees (> 15 m height), black gum pitchout is observed: black exudate covers entire branches and sometimes entire trunks (Burbano-Figueroa et al. 2018; Burbano-Figueroa et al. 2020). Older trees also demonstrate brown discoloration of bark surrounding ambrosia beetle galleries (Burbano-Figueroa et al. 2020). Wood under exit holes of galleries reveal brown necrosis (Burbano-Figueroa et al. 2018). Both adults and larvae of the above mentioned ambrosia beetles are found inside the borer galleries (Burbano-Figueroa et al. 2018).

Figure 5-1: Symptoms associated with avocado branch dieback and wilting. A) Multiple ambrosia beetle entry points and white exudate (perseitol production) in a trunk. B) Black pitch (black exudate) on trunk of tree which is indicative of multiple exposures to beetles and fungal infection. C, D, E) Production of perseitol and evidence of ambrosia entry points in young trees. Photo is sourced and permission granted from Oscar Burbano-Figueroa (Burbano-Figueroa et al. 2020).

Burbano-Figueroa et al. (2018) re-inoculated 6-month old avocado seedlings with *C. pseudochroleuca* isolated from diseased trees in their initial surveys by wounding seedlings and placing mycelial plugs into the wounds (Burbano-Figueroa et al. 2018). Expanding lesions were observed within a month on all inoculated seedlings (Burbano-Figueroa et al. 2018). It is most likely the combined effects of the fungus and ambrosia beetles that cause dieback and wilting in avocado plants. The ambrosia beetles listed above (especially *X. morigerus*) are significant pests of crop and ornamental trees (CABI 2021). *Xylosandrus volvulus* and *P. cavipennis* have been associated with wilting and dieback in plantations of *Tectona grandis* (teak) (Burbano-Figueroa et al. 2020).

Reproduction and transmission

Clonostachys pseudochroleuca produces ascospores (i.e. sexual spores of Ascomycota) and conidia (i.e. asexual spores) (Schroers 2001). Conidiophores (i.e. structure producing conidia) were not observed on natural substratum but have been observed in culture; the anamorph (i.e. asexual stage) is mainly associated with soil substrates (Schroers 2001). In comparison, teleomorph specimens (i.e., sexual stage) are found on natural substrates such as decaying plant material or bark of dead trees.

There is no evidence of *C. pseudochroleuca* producing chlamydospores (i.e., thick-walled long lived spores able to survive adverse environmental conditions). The most widely studied species, *Clonostachys rosea*, is a species in this genus that is known to produce chlamydospores under harsh environments such as unfavourable pH and low temperatures (Sun et al. 2020). There was no evidence in the literature on whether other species in this genus can produce chlamydospores.

The main mode of transmission is through ambrosia beetles as described above (Burbano-Figueroa et al. 2020). *Clonostachys pseudochroleuca* is an ambrosia fungus and infection in avocado has not been found without the presence of ambrosia beetle galleries (Burbano-Figueroa, pers. comm).

In culture, optimum growth temperatures for *C. pseudochroleuca* is between 24-30℃ (Schroers 2001). Environmental models indicate disease expression is favoured when the mean diurnal range in temperature is higher than 10℃ and temperatures in the wettest quarter of the year are 22.5℃ (Burbano-Figueroa et al. 2020). Disease suitability is also highest in areas with variations in monthly precipitation of 70%, low precipitation (200 mm) in the wettest month and no precipitation in the driest quarter of the year (Burbano-Figueroa et al. 2020). Disease suitability decreases sharply when precipitation increases in either the wettest month or driest quarter (Burbano-Figueroa et al. 2020). Disease does not occur in areas with precipitation in the wettest month and driest quarter is greater than 600 mm (Burbano-Figueroa et al. 2020). It is likely that severe droughts would trigger disease events; drought events provoke water related-stress in trees which favour attacks of pathogens and pests, including ambrosia beetles (Burbano-Figueroa et al. 2020; Reverchon et al. 2021).

5.4.4 Likelihood of entry

Clonostachys pseudochroleuca has a wide distribution in subtropical and tropical areas where avocado is produced (Table 1). However, the avocado branch dieback and wilting disease it causes has only been recorded in Colombia (Burbano-Figueroa et al. 2018).

Infection of *C. pseudochroleuca* is associated with ambrosia beetle infestation and is associated with beetle galleries (Burbano-Figueroa et al. 2018). *Clonostachys pseudochroleuca* has been isolated from branches and trunks of avocado trees inside these galleries (Figure 1). Avocado seedlings inoculated with *C. pseudochroleuca* through wounds also resulted in lesions on branches (Burbano-Figueroa et al. 2018). Therefore, the fungus could be associated with leafless, rootless avocado budwood cuttings.

Clonostachys pseudochroleuca is an ambrosia fungus and infection in avocado has not been found without the presence of ambrosia beetle galleries (Burbano-Figueroa, pers. comm). Ambrosia fungi depend on and are cultivated by ambrosia beetles (Reverchon et al. 2021). Therefore, if avocado budwood cuttings have an established *C. pseudochroleuca* infection, it is likely that beetle galleries will be detected, and infected material removed or destroyed, during phytosanitary inspections on arrival in New Zealand.

The routine surface disinfection of budwood entering PEQ (dipping in 1% sodium hypochlorite for 2 minutes) will most likely not eliminate infection, given the fungus will be associated with beetle galleries where the surface disinfection may not reach.

As part of the commodity description, all imported avocado germplasm will undergo a growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ. This period in 3A PEQ will impact on likelihood of entry in the following manner:

- ambrosia beetle galleries will be visible during inspection and thus infected budwood would be detected and destroyed before plants are released from PEQ.
- *Clonostachys pseudochroleuca* is spread by ambrosia beetles (Burbano-Figueroa et al. 2018). Budwood will be treated with miticides and insecticides (see section 2.2). Furthermore, Level 3A PEQ has measures in place to manage the risks associated with insects, wastewater, waste and accidental transfer (see section 2.3), therefore ambrosia beetles carrying *C. pseudochroleuca* are highly unlikely to escape level 3A PEQ to infect a suitable host.

Therefore, if *C. pseudochroleuca* is imported on avocado, the residual biosecurity risks are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ.

Given the arguments and evidence above, that is:

- established *C. pseudochroleuca* infections are highly likely to be detected, and infected material removed or destroyed, during preparation for export, in phytosanitary inspections pre-export or on arrival in New Zealand;
- residual biosecurity risks from ambrosia entry points or recent infections on the surface of avocado budwood are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ
    - symptoms in inoculated plants are observed within one month after inoculation;

the likelihood of *Clonostachys pseudochroleuca* entering New Zealand associated with *Persea americana* budwood is considered to be negligible, with low uncertainty.

Therefore, further assessment is considered unnecessary at this time, and likelihoods of exposure and establishment, and impacts to New Zealand were not assessed.

5.4.5 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood above, that:

- the likelihood of *C. pseudochroleuca* entering New Zealand associated with *Persea americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ) is considered to be negligible with low uncertainty;
- therefore, continuing the assessment was considered unnecessary; and

the overall level of risk to New Zealand from *C. pseudochroleuca* on imported *Persea americana* budwood (as in the commodity description) is considered to be negligible, with low uncertainty.

5.4.6 Management considerations

Clonostachys pseudochroleuca and their vectors, ambrosia beetles, cause visible damage on stems of avocado plants. Infection by *C. pseudochroleuca* is unlikely without a vector (Burbano-Figueroa, per. comm). Therefore, *C. pseudochroleuca* is highly unlikely to persist as an asymptomatic infection on susceptible tissues.

Ambrosia beetle and their associated fungi commonly infest stressed trees due to drought events (Burbano-Figueroa et al. 2020; Reverchon et al. 2021) and thus development of symptoms would be more common under high temperatures and dry environments. Ambrosia beetles are unlikely to attack healthy plants (Reverchon et al. 2021).

Clonostachys spp. can be detected with specific primers for PCR targeting the β-tubulin gene region (Abreu et al. 2014) if symptoms are detected in PEQ.

5.4.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Clonostachys pseudochroleuca infection in avocado is only associated with ambrosia beetle galleries (Burbano-Figueroa et al. 2018) and causes avocado branch dieback and wilting (Figure 1). Symptoms include signs of damage by boring insects in trunks and branches. White powdery exudate on the outer bark is associated with beetle holes and wood under exit holes of galleries will reveal brown necrosis (Burbano-Figueroa et al. 2018). Older plants also show black pitch symptoms (i.e. black exudate found on branches) (Burbano-Figueroa et al. 2018).

- ***What are the known environmental conditions conducive to symptom expression?***

Clonostachys pseudochroleuca is vectored by ambrosia beetles and beetle attacks are unlikely to be associated with healthy trees (Reverchon et al. 2021). Drought events that increase stress in tree hosts are more likely to increase ambrosia beetle infestation and subsequent infection by *C. psuedochrolecua* (Burbano-Figueroa et al. 2020). Therefore, high temperatures and dry environments would be conducive to symptom expression.

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

There is no evidence *C. pseudochroleuca* causes asymptomatic infections and thus, there is no information on which plant part should be tested to maximise the detection of *C. pseudochroleuca* in the case of asymptomatic infection.

- ***What is the optimum season for conducting detection testing?***

Drought conditions exacerbate avocado branch dieback and wilting disease caused by *C. pseudochroleuca* and ambrosia beetles (Burbano-Figueroa et al. 2020). Black pitch symptoms grow during the dry season as well (Burbano-Figueroa, pers. comm). However, black pitch symptoms are commonly found in older trees and after multiple infestations by ambrosia beetles (Burbano-Figueroa et al. 2020).

5.4.8 References

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5.5 *Cophinforma tumefaciens*

Cophinforma tumefaciens is a pathogenic ascomycete fungus that causes stem galls/tumours in young and old avocado plants. It is known to cause galls/tumours in *Citrus* spp. as well as in other host plants.

5.5.1 Taxonomic description

Scientific name: *Cophinforma tumefaciens* (Hedges) F. Liu, Crous & L. Cai (2021)

Order/Family: Botryosphaeriales/Botryoshaeriaceae

Other names include: *Sphaeropsis tumefaciens* Hedges (1911)*, Sphaeropsis tumefaciens* var. citri N.D. Sharma (1974).

Taxonomic notes: *Sphaeropsis tumefaciens* was recently named *Cophinforma tumefaciens* due to recent taxonomic revision based on DNA phylogeny (Zhao et al. 2021; Index Fungorum 2022).

5.5.2 Hazard identification

Cophinforma tumefaciens is not known to be present in New Zealand:

- *Cophinforma tumefaciens* is not listed in either Biota NZ (2022), (PPIN 2021) or (NZOR 2021).
- *Cophinforma tumefaciens* is listed as regulated and an unwanted organism in ONZPR (2021).

Cophinforma tumefaciens has the potential to establish (and spread) in New Zealand because:

- host plants such as avocado and *Citrus* spp. are grown commercially and in home gardens, in northern and southern parts of New Zealand,
- *Cophinforma tumefaciens* can spread by human movement of infected nursery stock to avocado and citrus growing areas,
- climate is unlikely to be a barrier to its establishment and spread in New Zealand because it is reported to occur in places (Table 1), which have a similar climate (CMI ≥ 7) to New Zealand.

Cophinforma tumefaciens has the potential to cause harm to New Zealand because:

- *Cophinforma tumefaciens* has been associated with and shown to cause stem tumours in young and old avocado plants in Mexico (Fucikovsky 1992).
- Diseases caused by *C. tumefaciens* can potentially reduce the productivity of avocado and citrus trees as well as some ornamental plants in New Zealand. Avocado is an important commercial crop in New Zealand worth up to $110 million in domestic sales and up to $56.6 million in international sales (Plant & Food Research 2020). In 2019, limes, lemons and mandarins had a domestic value of NZ$41 million, with lemons and mandarins having export values of NZ$8 million (Plant & Food Research 2020).

Cophinforma tumefaciens is associated with *Persea americana* budwood because:

- *Persea americana* is a known host of *C. tumefaciens* (Fucikovsky 1992; Baker et al. 2008);
- *Cophinforma tumefaciens* can cause stem tumours on avocado stems, twigs and branches (Fucikovsky 1992; Baker et al. 2008).

Given the arguments and evidence above, *C. tumefaciens* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.5.3 Risk assessment

Biology and epidemiology of **Cophinforma tumefaciens**

General biology and host range of *Cophinforma tumefaciens*

Cophinforma tumefaciens is an ascomycete fungus that can cause stem knot disease in woody plant species (Nazerian et al. 2006; Dale et al. 2021). It was first isolated from *Citrus* spp. that were affected by knot disease in Jamaica (Naylor 1963). It is known to infect woody plant species by causing galls/tumours on stems, branches, and trunks. It has caused diseases in *Citrus* spp. and avocado as well as several ornamental species such as *Callistemon* spp. and *Illex* spp (Table 5-8).

Disease caused by *C. tumefaciens* can be related to water stress. Dieback symptoms and plant death have been observed to be more severe in warm and wet conditions followed by a period of drought (Dale et al. 2021). The disease can be controlled by cutting and destroying infected branches, treating pruning wounds with protective paint. Pruning tools used to should be disinfected using disinfectant such as 10% Clorox after use to prevent spreading the disease to uninfected plants (Ridings and Marlatt 1975).

Table 5-8: The distribution, host range and diseases caused by *Cophinforma tumefaciens*.

Continent	Country	State/Region	Host plants	Symptoms/disease	CMI*	Reference
Asia	Iran	Mazandran	*Citrus sinensis* (Tomson navel)	Stem knot disease	0.7-0.8	Nazerian et al. (2006)
North America	Jamaica		*Citrus* spp.	Knot disease	0.5-0.6	Naylor (1963)
	Mexico	Villa Guerrero, Veracruz	*Persea americana* (avocado)	Stem tumours	0.4-0.6	Fucikovsky (1992)
	United States	Adjuntas Puerto Rico	*Citrus* spp.	Citrus knot disease	0.6	(Hedges and Tenny 1912; Rodriguez et al. 1985a)
		Florida	*Carissa macrocarpa*, *Nerium oleander*	Galls	0.5-0.6	Farr and Rossman (2021)
		Florida	*Schinus terebinthifolius*	Stem galls	0.5-0.6	(Marlatt and Ridings 1979; Farr and Rossman 2021)
		Florida (DeSoto,Glades, Highlands and Polk)	*Hypericum edisonianum* (Edison's St. John's-wort)	Stem tumours (galls)	0.5	Van de Kerckhove et al. (2002)
		Florida	*Illex* spp. (hollies)	Stem gall and witches broom disease	0.5-0.6	Brown et al. (2012)
		Florida (Dade County)	*Callistemon viminalis* (weeping bottlebrush)	Stem galls and cankers	0.5	Marlatt and Ridings (1974)
		-	*Eucalyptus cinerea*	Stem galls and cankers		(Farr et al. 1989; Yuan 1998)

***(Phillips et al. 2018)**

Symptoms

Cophinforma tumefaciens causes galls (swelling and growth) that occurs on stems, branches, and trunks of infected plants (Naylor 1963; Fucikovsky 1992; Fucikovsky 1994; Brown et al. 2012). Galls can occur in large numbers and can lead to girdling and eventual death of affected twigs (Fucikovsky 1994; Baker et al. 2008). It can lead to death of entire plants in severe cases (Fucikovsky 1994).

Infections by *C. tumefaciens* causes galls beneath the bark of the plant which eventually cracks revealing pycnidia (fungal fruiting structures). The pathogen can be spread wind and by water splash, and infect plants through wounds (Brown et al. 2012; Dale et al. 2021).

Hard woody galls appear as knots which are either rounded or elongated and occur along the stem. Knots occur smooth on young branches and new growth and may become broken, sunken and cankerous on old infection (Naylor 1963). They can be also be found on older parts of the stem and even close to the rootstock (Naylor 1963; Blazquez and Hunt 1967). Knots (1-4cm diameter on stems) can girdle twigs and branches causing dieback and “witches broom effect” (deformed of mass of twigs and branches) (Marlatt and Ridings 1974; Nazerian et al. 2006).

Cophinforma tumefaciens caused tumours of different sizes (up to 8cm) and shapes on branches and young stems of avocados in Mexico (Fucikovsky 1992). Fucikovsky (1994) observed knots in up to 80% of grafted avocado plants that were placed under trees infected with *C. tumefaciens* (Baker et al. 2008).

Transmissions and spread

Cophinforma tumefaciens causes galls beneath the bark of the plant which eventually cracks revealing pycnidia (fungal fruiting structures). It can be spread by water splash, and infect plants through wounds (Brown et al. 2012; Dale et al. 2021). Pruning provides opportunity for the pathogen to infect wounds and spread throughout an orchard on contaminated equipment (Fucikovsky 1994; Brown et al. 2012; Popenoe et al. 2019). *Cophinforma tumefaciens* infected wounded avocado nursey plants placed under trees infected with the pathogen*,* while unwounded plants were not infected (Fucikovsky 1994). This suggests that wounds are necessary for infections to occur. *Cophinforma tumefaciens* can also be spread by the movement of infected plants (Blazquez and Hunt 1967) and contaminated pruning tools to new locations (Ridings and Marlatt 1975).

Time to symptom development

During pathogenicity tests, *C. tumefaciens* caused disease symptoms on different host plants (avocado, citrus and weeping bottlebrush), between about 2 – 24 weeks after inoculations (Marlatt and Ridings 1974; Rodriguez et al. 1985a; Rodriguez et al. 1985b; Fucikovsky 1992). It caused swelling on at inoculation areas of *Citrus* spp. during pathogenicity tests after 12 days and they exhibited dieback symptoms two months after (Rodriguez et al. 1985a). Knots were observed on *Citrus medica* 25 days after inoculation and decline of affected branch occurred after 60 days with mature pycnidia at the outer layer of the stem (epidermis cortex) and the vascular tissues (Rodriguez et al. 1985b).

Early gall formations were observed in *Callistemon viminalis* from 35 days of artificial inoculation. Galls increased rapidly in size between 42-126 days after inoculation. On naturally infected trees, galls varied in size with older galls developing a “witches broom effect” (deformed of mass of twigs and branches) (Marlatt and Ridings 1974).

Factors that influence symptom expression

Cophinforma tumefaciens causes diseases symptoms such as galls that appears to be related to water (Dale et al. 2021). Galls can generally girdle branches and interrupt the flow of nutrient and water (University of Minnesota 2019). This makes the plant more susceptible to drought stress and can lead dieback symptoms (University of Minnesota 2019). Dieback symptoms and death of woody ornamental plant species infected by *C. tumefaciens* have been observed to be more in severe, warm and wet conditions followed by a period of drought (Dale et al. 2021).

5.5.4 Likelihood of entry

Cophinforma tumefaciens has been reported to be present in Iran, Mexico, Jamaica, and the United States of America (Table 5-8). It caused galls on branches and young stems of avocados (Fucikovsky 1992; Fucikovsky 1994), and can be spread by the movement of infected plant stock (Blazquez and Hunt 1967). The fact that *C. tumefaciens* causes galls on branches of infected avocado plants (Fucikovsky 1994) suggests that it is unlikely that budwood for nursery stock will be harvested from infected plant material. However, infections of plant material by *C. tumefaciens* can occur through the use of contaminated pruning tools (Ridings and Marlatt 1975) which means that avocado nursery stock (budwood) can be infected from pruning tools during preparation for export. As such, infected budwood might not be detected by visual inspection during preparation for export or during phytosanitary inspections on arrival to New Zealand.

As part of commodity description, all imported avocado germplasm, will undergo a growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ. This period in 3A PEQ will impact the likelihood of entry in the following manner:

- Avocado seedings derived from infected avocado budwood are likely to exhibit symptoms during the time spent in PEQ. During pathogenicity tests, *C. tumefaciens* was observed to cause symptoms between about 2 – 24 weeks after inoculations on different host plants including avocado (Marlatt and Ridings 1974; Rodriguez et al. 1985a; Rodriguez et al. 1985b; Fucikovsky 1992)
- swollen galls produced by *C. tumefaciens* infections can eventually crack, revealing pycnidia (fungal fruiting structures) which can be spread by water splash (Brown et al. 2012; Dale et al. 2021). However, this is unlikely to occur because regular inspections at Level 3A PEQ are likely to detect galls. Additionally, there are measures in place to manage the risks associated with wastewater, waste, and accidental transfer (see section 2.2), therefore viable *C. tumefaciens* propagules are highly unlikely to escape level 3A PEQ to infect a suitable host.
- Searches of Google Scholar, CPC and Google using the search terms ‘Sphaeropsis tumefaciens’, ‘Cophinforma tumefaciens endophytic infection’ found no evidence for asymptomatic *S. tumefaciens* infections or longer latent periods in avocado plants and other host plants.

Therefore, if *C. tumefaciens* is imported on avocado budwood, the residual biosecurity risk is likely to be managed by the active growing period in 3A PEQ.

Given the arguments and evidence above, that is:

- established *C. tumefaciens* infections are highly likely to be detected, and infected material removed or destroyed, during preparation for export, or in phytosanitary inspections pre-export and on arrival in New Zealand
- residual biosecurity risks from recent infections or contamination of the surface of avocado budwood are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ

the likelihood of *C. tumefaciens* entering New Zealand associated with *Persea americana* budwood is considered to be **negligible**, with **low** uncertainty.

5.5.5 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of *C. tumefaciens* entering New Zealand associated with *Persea americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ) is considered to be negligible with low uncertainty;
- therefore, continuing the assessment was considered unnecessary; and

the overall level of assessed risk to New Zealand from *C. tumefaciens* on imported *Persea americana* budwood (as in the commodity description) is considered to be **negligible**, with **low** uncertainty.

5.5.6 Management considerations

Cophinforma tumefaciens causes galls and tumours that occurs on stems, branches, and trunks of infected plants (Naylor 1963; Fucikovsky 1992; Fucikovsky 1994; Brown et al. 2012). Galls and tumours vary in size reaching up to 8cm in diameter and can occur in large numbers (Fucikovsky 1992). They can lead to death of affected twigs (Fucikovsky 1994; Baker et al. 2008) and in severe cases result in the death of the entire plant (Fucikovsky 1994).

C. tumefaciens is unlikely to persist as an asymptomatic infection on host plant or plant material. Symptoms are likely to become visible between 2 - 18 weeks of infection. This suggests that symptoms of pre-existing infections are likely to be detected within the period of level 3A PEQ.

Infections caused by *C. tumefaciens* appears to be related to water stress. Dieback symptoms and plant death have been observed to be more severe in warm and wet conditions followed by a period of drought (Dale et al. 2021).

Disease caused by *C. tumefaciens* can be controlled by ensuring that uninfected nursery plants are not be kept in close proximity to or placed under tress infected with *C. tumefaciensi* (Fucikovsky 1994).

5.5.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Cophinforma tumefaciens causes galls on stems, branches, and trunks of infected plants (Naylor 1963; Fucikovsky 1992; Fucikovsky 1994; Brown et al. 2012). Multiple galls can occur on the bark of twigs and can girdle affected part leading to death (Fucikovsky 1994; Baker et al. 2008). In severe cases, it can result in the death of the entire plant (Fucikovsky 1994; Baker et al. 2008). It caused galls of different sizes (up to 8cm in diameter) and shapes on branches and young stems of avocado plants in Mexico (Fucikovsky 1992).

- ***What are the known environmental conditions conducive to symptom expression?***

Infections caused by *C. tumefaciens* appear to be related to water stress. Dieback symptoms and plant death have been observed to be more severe in warm and wet conditions followed by a period of drought (Dale et al. 2021).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Cophinforma tumefaciens is unlikely to persist as an asymptomatic infection on host plant or plant material because infections have been observed to occur mainly through wounds (Marlatt and Ridings 1974; Rodriguez et al. 1985a; Rodriguez et al. 1985b). Recently infected material (cut budwood) as a result of contamination during preparation is likely to exhibit symptoms within the PEQ period.

- ***What is the optimum season for conducting detection testing?***

Optimum season for conducting testing has not been specified.

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5.6 *Diaporthe perseae*

Diaporthe perseae is a fungal plant pathogen that causes stem-end rot (postharvest) disease in avocado.

5.6.1 Taxonomic description

Scientific name: *Diaporthe perseae* (Zerova) Gomes et al. (2013)

Order/Family: Diaporthales/Diaporthaceae

Other names include: *Phomopsis perseae* Zerova (1940)

Taxonomic notes: *Diaporthe perseae* was also referred to as *Phomopsis perseae* (known as the asexual state). Historically it was commonly referred to as *Phomopsis perseae*, but for the purpose of this PRA, it will be referred to as *Diaporthe perseae*.

5.6.2 Hazard identification

Diaporthe perseae is not known to be present in New Zealand. However, two *Phomosis* spp. (asexual stage of *Diaporthe*) were reported by Hartill and Everett (2002) to be associated with avocado in New Zealand. Neither of these were confirmed as *Diaporthe perseae.*

- *Diaporthe perseae* is not listed in PPIN (2021)
- *Diaporthe perseae* is recorded as absent from New Zealand in Biota NZ (2022)
- *Diaporthe perseae* is a regulated pest for New Zealand, has unwanted status but is not a notifiable organism (ONZPR 2021)

Diaporthe perseae has the potential to establish (and spread) in New Zealand because:

- The host plant, avocado is grown commercially, and in home gardens in some parts of New Zealand.
- It is an endophytic pathogenic fungus, that has been isolated from twigs, branches and trunks not showing diseased symptoms. It can be present on all parts of an infected tree (Zerova 1940; Darvas and Kotze 1987).
- Climate is unlikely to be a barrier to its establishment and spread in New Zealand because it is reported to occur in places such as Australia and South Africa, which have a similar climate (CMI) ≥ 0.7 to New Zealand (Phillips et al. 2018).
- *Diaporthe perseae* can spread naturally by wind and rain or by human movement of infected nursery stock and movement of infected fruits to avocado growing areas.

Diaporthe perseae has the potential to cause harm to New Zealand because:

- *Diaporthe perseae* has been associated with and shown to cause postharvest (stem-end) rot disease in avocado (Darvas and Kotzé 1981; Darvas and Kotze 1987; Korsten et al. 1994).
- *Diaporthe perseae* can cause cankerous lesions on trunks of avocado trees in South Africa (Van der Merwe and Kotzé 1991).
- Postharvest rot diseases caused by *D. perseae* can affect the marketability of avocado crops in New Zealand. It is an important commercial crop in New Zealand worth up to $110 million in international sales and up to 56.6 million in domestic sales (Plant & Food Research 2020).

Diaporthe perseae is associated with *P. americana* budwood because:

- *Persea americana* is a known host of *Diaporthe perseae* (Darvas and Kotzé 1981; Darvas and Kotze 1987; Darvas and Wehner 1987);
- *Diaporthe perseae* can occur endophytically on all parts of a tree (Darvas and Kotze 1987). It can occur on twigs, branches and pedicels of avocado plants. From the pedicel, it can

eventually colonise the inflorescence of the stem-end tissue and cause stem-end rot (Hartill and Everett 2002).

Given the arguments and evidence above, *D. perseae* meets the criteria to be a hazard on *Persea americana* budwood (as per the commodity description) imported to New Zealand.

5.6.3 Risk assessment

Biology and epidemiology of Diaporthe perseae

Biology and host range

Diaporthe perseae is an endophytic (can inhabit plant tissues without causing disease) and pathogenic fungus (Wilson 1995) that is known to cause stem-end rot disease (Darvas and Kotze 1987). Depending on the host and the health of the host plants, some species of *Diaporthe* can exist as harmless endophytes or as pathogens (Udayanga et al. 2011). Some are host specific as is the case of *Diaporthe perseae* (Farr et al. 2002), which has mostly been reported as a pathogen of *Persea americana* (avocado).

Diaporthe perseae was originally described when conidiospores (asexual spores) from the branches of dying trees and leaves of avocado *Persea gratissima* (*americana*) were observed and isolated in Russia (Zerova 1940; Uecker 1988). It has since been reported to cause postharvest disease of avocado in Australia (Peterson 1978; Muirhead et al. 1982) and South Africa (Darvas and Kotze 1987) and has been isolated from young avocado fruits in the Netherlands (Gomes et al. 2013). It is, however, unclear if *D. perseae* is still present in Australia. It had been reported to be associated with avocado by Peterson (1978) in Australia in the late 1970’s and there is no information on its present status in Australia. Lim et al. (2019) recently reported *D. perseae* as one of the fungi observed in stem-end rot disease of mangoes in Malaysia. It was also recently reported to be isolated along with other *Diaporthe* on healthy leaves of *Citrus grandis* in China (Dong et al. 2020) and from *Camellia sinensis* (tea leaves) in Taiwan (Ariyawansa et al. 2021) (Table 5-9).

Diaporthe perseae can occur endophytically on all parts of a tree (Darvas and Kotze 1987). It can occur in twigs, branches and pedicels of avocado plants. From the pedicel, it can eventually colonise the inflorescence of the stem-end tissue and cause stem-end rot (Hartill and Everett 2002). However, infection can remain latent (not show symptoms) prior to harvest with symptoms becoming apparent after harvest (Peterson 1978). Immature fruits are more vulnerable to stem-end rot infections than mature fruits (Kotzé and Darvas 1985).

Stem-end rot is often reported to be caused by several fungal pathogens (Darvas and Kotze 1987; Korsten et al. 1994; Guarnaccia et al. 2016). Pathogenicity tests using several pathogens that cause stem-end rot disease found that *D. perseae* appeared to be a weak pathogen of stem-end rot disease in avocado (Peterson 1978; Darvas and Kotzé 1981; Menge and Ploetz 2003). Korsten et al. (1994) also found that *D. perseae* was less frequently isolated from diseased avocado fruits than other postharvest rot causing fungi. It was observed to cause less severe disease symptoms than other pathogens (Darvas and Wehner 1987). However, it is important to note that some pathogenic fungi such as *Lasiodiplodia theobromae* might be isolated at low frequency but are destructive wound pathogens of avocado.

Table 5-9: Plant host range and disease cause by *Diaporthe perseae*

Continent	Country	Host plant	Symptoms /disease	CMI*	Reference
Africa	South Africa	*Persea americana* (Avocado)	Stem-end rot	0.5–0.9	(Darvas and Kotze 1987; Darvas and Wehner 1987)
Asia	China	*Citrus grandis*	Asymptomatic	0.4-0.8	Dong et al. (2020)
	Malaysia	*Mangifera indica* (Mango)	Stem-end rot	0.4-0.5	Lim et al. (2019)
	Taiwan	*Camellia sinensis* (tea leaves)	Asymptomatic	0.4–0.5	Ariyawansa et al. (2021)

Continent	Country	Host plant	Symptoms /disease	CMI*	Reference
Europe	Netherlands	*Persea americana* (Avocado)	Fruit rot	0.9	Gomes et al. (2013)
	Russia	*Persea americana* (Avocado)	Dieback disease	0.3-0.8	(Zerova 1940; Uecker 1988)
Oceania	Australia	*Persea americana* (Avocado)	Post-harvest disease	0.4-0.9	(Peterson 1978; Muirhead et al. 1982)

*(Phillips et al. 2018)

Reproduction and spread

Under field conditions, spores are generally produced when tissues (on twigs, branches and stems) are dead (Darvas and Kotze 1987; Uecker 1988; Menge and Ploetz 2003; Gomes et al. 2013), then grey, white or creamy colonies containing conidiomata appear (Gomes et al. 2013). This can usually serve as a source of primary infection (Dann et al. 2013), can be transferred to open wounds on the plant, the pedicel, and fruits, and can potentially be spread by rain splashes or wind (Menge and Ploetz 2003).

Most stem-end rot infections occur during harvest through the pedicel, pedicel scars or wounds (Darvas and Wehner 1987). Infections can occur through wounds on the bark of the tree (which can also be a source of infection) or wounds created through picking of fruit in the field (Kotzé and Darvas 1985; Hartill and Everett 2002). It can also be spread by the use of the same cutting implement from an infected plant to a non-infected plant (Hartill and Everett 2002).

Diaporthe perseae, in its endophytic phase, can be latent or dormant without showing any symptoms (Dann et al. 2013; Dong et al. 2020). This means that symptomless cuttings of avocado infected with *D. perseae* can be transmitted and spread through grafting especially in commercial avocado plant nurseries. Under unfavourable environmental conditions such as drought, frost or hail, plants colonised by *D. perseae* can become stressed and begin to exhibit symptoms (Udayanga et al. 2011).

Infection and spread of *D. perseae* (as well as other pathogenic fungi associated with avocado) occurs mostly during prolonged wet periods of rain and high humidity. Peterson (1978) found that natural infection of fruits did not occur in dry conditions but occurred after four or more conservative days of rainfall.

Symptoms

Pathogenic fungi such as *D. perseae* that are endophytic usually have a latent or dormant phase within the plant tissues (Wilson 1995). They can be present throughout the life cycle of infected plants without showing visible symptoms. Symptoms of *D. perseae* such as lesions and rot, tend to only occur after harvest (Peterson 1978). This means that in its latent phase, it can occur in healthy looking avocado twigs and stems, with no visible symptoms.

In its pathogenic form, *D. perseae* colonises the xylem and the phloem at the stem-end of the fruit. It eventually becomes necrotrophic (secretes enzymes that kill the host tissues while living on the nutrients from the dead tissues), causing stem-end rot by colonising the fruit pulp (Karunanayake and Adikaram 2020).

Symptoms develop after harvest, at the onset of fruit ripening (Karunanayake and Adikaram 2020). Visible symptoms begin as brownish or black rot at the fruit pedicel which progresses internally causing discolouration and decay. Internal decay may extend on the external part of the fruit leading to brown or black discolouration (Darvas and Wehner 1987; Karunanayake and Adikaram 2020). Mycelium and abundant spores might be observed on the abscission scar of infected fruit and can eventually cover the fruit as ripening progresses, leading to softening and shrivelling of the entire fruit (Menge and Ploetz 2003).

Conditions for symptom development

Symptom development of stem-end rot disease varies with pathogens and can be slower or faster depending on the pathogen (Karunanayake and Adikaram 2020) as well as the environmental conditions. For example, cool temperature is thought to promote *D. perseae* infection. Additionally water stress appears to generally stimulate latent infections by several fungi (Menge and Ploetz 2003), suggesting that colonised plants are likely to exhibit pathogenic effects of *D. perseae* in dry conditions.

By inoculating avocado (Martin Grande) seedlings under artificial condition, (Van der Merwe and Kotzé 1991) found that *D. perseae* caused lesions on cuttings and within cambium tissues (phloem and xylem tissues) ten days after inoculation at 26°C- 28°C. Within 20 days, inoculated plants completely dried out and died (Van der Merwe and Kotzé 1991)**.** The same inoculation method resulted in no symptom development on “Duke 7” avocado variety, suggesting *D. perseae* might affect some avocado varieties and not others (Van der Merwe and Kotzé 1991).

Diaporthe perseae was observed to cause fruit rot after eight to eleven days in artificial post-harvest inoculations in Fuerte, Edranol, and Hass varieties but no rot was observed in the Ryan variety (Darvas and Kotzé 1981).

5.6.4 Likelihood of entry

Diaporthe perseae has been reported to be present in seven countries. It has been reported to be associated with avocado in South Africa, Russia, Australia and the Netherlands (Peterson 1978; Muirhead et al. 1982; Darvas and Kotze 1987; Uecker 1988; Gomes et al. 2013). It is, however, unclear if *D. perseae* is still present in Australia. It was reported to be associated with avocado by Peterson (1978) in Australia in the late 1970’s and there is no information on its present status in Australia.

In its endophytic phase, it can be present on leafless and rootless avocado budwood without showing visible symptoms. Therefore, it is likely to that it will not be detected during export preparation and pre-export phytosanitary inspection to New Zealand or on arrival to New Zealand. Given that it has been reported to be present in at least seven countries, *D. perseae* can be associated with the commodity when it arrives in Post Entry Quarantine (PEQ).

All imported avocado germplasm will undergo PEQ at level 3A, during which the budwood will undergo a minimum of three distinct phenological periods of active growth. The period in Level 3A PEQ is not likely to impact the likelihood of entry of *D. perseae*. This is because infected budwood that is grafted in PEQ level 3A is not likely to show any visible symptoms within the time in PEQ if the fungus is in a latent phase. However, symptoms such as dead twigs and sporulation of the grafted plants can develop if the plants become subjected to stress (such as drought). Spores are generally produced when tissues (on twigs, branches and stems) are dead (Darvas and Kotze 1987; Uecker 1988). It therefore highly unlikely that spores will be produced on imported budwood and escape from PEQ without being detected.

Disease symptoms (stem-end rot) associated with *D. perseae* occur mostly on the fruit after harvest. This cannot be detected in PEQ level 3A because grafted budwood will not be in PEQ long enough to produce suitably mature fruit. As such, infected plants are likely to remain undetected unless molecular diagnostic testing for *D. perseae* is carried out on infected tissue.

Given the arguments and evidence above, that is:

- *Diaporthe perseae* has been reported to be present in seven countries
- it can be present on leafless and rootless avocado budwood without showing visible symptom
- Infected budwood that is grafted in PEQ level 3A is not likely to show any visible symptoms within the time in PEQ if the fungus is in a latent phase.
- Infected plants are likely to remain undetected unless molecular diagnostic testing for *D. perseae* is carried out on infected tissue.

the likelihood of *D. perseae* entering New Zealand associated with *Persea americana* budwood is considered to be high, with low uncertainty.

5.6.5 Likelihood of exposure

This assessment is made on the basis that *D. perseae* has entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *D. perseae*.

If avocado plants are infected with *D. perseae* at the time of their release from post entry quarantine, they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *D. perseae* in New Zealand from *Persea americana* budwood is considered high, with low uncertainty.

5.6.6 Likelihood of establishment

This assessment is made on the basis that *D. perseae* has been successfully exposed to a suitable host plant in the New Zealand environment.

Diaporthe perseae is recorded to be present in parts of Australia, South Africa, the Netherlands and Russia which have a CMI range of between 0.4 - 0.9. CMI of ≥ 0.7 indicates a similar climate to the whole of New Zealand (Phillips et al. 2018). As such, the conditions in some parts of New Zealand are likely to be suitable for D. *perseae* to occur and spread. Therefore, climate would not be a barrier to the establishment of *D. perseae* in New Zealand.

Avocado plants are widely grown in some parts of New Zealand commercially in orchards and in-home gardens mostly in the North Island and some parts of the South Island (Chapter 2), therefore suitable hosts are available in those areas. *D. perseae* can spread by the movement of infected nursery stock for planting. However, its spread might be limited because it is generally believed to be specific to avocado (Darvas and Kotze 1987; Darvas and Wehner 1987; Uecker 1988). However, it was recently reported to be associated with stem-end in mangoes in Malaysia (Lim et al. 2019), isolated from healthy citrus leaves in China (Dong et al. 2020) and from tea plants in Taiwan (Ariyawansa et al. 2021). These recent molecular studies are indicating that many *Diaporthe* species have wider host ranges that historically reported (Lim et al. 2019; Dong et al. 2020; Ariyawansa et al. 2021). Therefore, although it is likely that *D. perseae* will spread in avocado growing areas of New Zealand, there is moderate uncertainty because it may be able to infect other hosts.

Spores of *D. perseae* can be present on infected fruit (Menge and Ploetz 2003) and can be a source of infection if infected fruit is discarded near avocado plants. Therefore *D. perseae* can be spread to new areas by human movement of infected fruit.

Given the arguments and evidence above, that is:

- The climatic conditions in some parts of New Zealand are likely to be suitable for D. *perseae* to occur and spread.
- Suitable avocado host plants are available in orchards and in-home gardens mostly in the North Island and some parts of the South Island. *D. perseae* can spread by the movement of infected nursery stock for planting.
- Spores of *D. perseae* can be present on infected fruit and can be spread to new areas by human movement of infected fruit.

the likelihood of *D. perseae* establishing in New Zealand is considered high, with low-moderate uncertainty.

5.6.7 Impacts in New Zealand

Diaporthe perseae is reported as present in seven countries (Table 5-9). It causes stem-end rot (postharvest) disease in mostly avocado (Darvas and Wehner 1987) and have been reported to cause stem-end rot in mangos (Lim et al. 2019). It can be spread by the use of the same cutting implement from an infected plant to a non-infected plant (Hartill and Everett 2002), and spores produced on infected plants can be spread by water splash and infect plants through wounds created through picking of fruit in the field (Kotzé and Darvas 1985; Hartill and Everett 2002).

Economic impacts

Diaporthe perseae causes stem-end rot disease symptoms that can go unnoticed and mainly become apparent after harvest. Diseased symptoms of stem-end rot become apparent at the onset of fruit ripening (Karunanayake and Adikaram 2020). Infected fruits can mask symptoms in certain storage conditions such as low temperatures (Darvas 1982) with rot symptoms becoming apparent later outside those conditions. Muirhead et al. (1982) reports *D. perseae* as a less important pathogen of stem-end rot disease than other stem-end rot causing fungi such as *Dothiorella aromatica* in Queensland Australia. Stem-end rot caused by *Colletotrichum gloeosporioides* and *Phomopsis* spp. are also common in New Zealand (Hartill and Everett 2002).This suggests that while *D. perseae* can result in some postharvest losses of avocado fruits, overall losses may not differ from current levels of stem-end rot diseases.

Infections by *D. perseae* could affect market access of exports of avocado to other parts of the world where *D. perseae* is not reported to be present. Because *D. perseae* is reported to cause diseases in avocado in few countries including Australia (Table 5-9). The fact that it is recorded as present in Australia, which is New Zealand's major export market, suggests it is not likely to have a significant impact on export. But its presence in New Zealand can potentially result in trade barriers to countries other countries where it is has not been reported to be present.

Pathogenicity testing using different stem-end rot pathogens showed that some avocado cultivars were more susceptible to stem-end rot infections than others. The Hass variety was observed to be less susceptible to stem-end rot disease caused by *D. perseae* than Fuerte, Edranol and the Ryan varieties (Darvas 1982). *D. perseae* is likely to infect and cause diseases in New Zealand. This might however be limited in New Zealand because the Hass variety is the main avocado presently grown (Chapter 2). However, its impact will likely be higher if more susceptible varieties of avocado are grown in the future.

Given the arguments and evidence above, that is:

- *D. perseae* can result in some postharvest losses of avocado, overall losses may not differ from current levels of stem-end rot disease in New Zealand.
- Current methods of control of other stem-end rot causing pathogens in New Zealand may also be effective for controlling diseases caused by *D. persea*.
- Infection by *D. perseae* could affect market access of exports of avocado to other parts of the world where *D. perseae* is not reported to be present.
- Infection caused by *D. perseae* might be limited in New Zealand because the Hass variety which is the main avocado is recorded to be less susceptible to stem-end rot disease.

the economic impact of *D. perseae* to New Zealand is considered to be low, with moderate uncertainty.

Environmental impacts

Diaporthe perseae has been reported to infect mainly avocado with only one recent association with mangoes by Lim et al. (2019). *Diaporthe perseae* is also asymptomatic in *Camellia* and *Citrus* (Table

5-9). Five indigenous New Zealand plant species belong to the family Lauraceae, the same family as avocado (*Perseae americana*) (NZPCN 2021). Searches from Google, Google scholar, **U.S. National Fungus Collections Fungus-Host Distributions Database,** CABI and EPPO using the search term '*Diaporthe perseae* and the environment' and '*Phomopsis perseae* and the environment' found no record of *D. perseae* infecting other plants species outside the *Persea* genus except for recent report of its association to mangoes and asymptomatic infections in *Camellia* and *Citrus*. This suggests that *D. perseae* is relatively host specific and is not likely to affect native plants and the environment.

Given the arguments and evidence above, the impact on the environment from the establishment of *D. perseae* in New Zealand is considered to be low, with low uncertainty.

Health impacts

Searches using the search term '*Diaporthe perseae* impacts on health' and '*Phomopsis perseae* impact on health' from Google, Google scholar, CABI and Pubmed found no record of *D. perseae* causing any health impact on humans and animals.

Given the arguments and evidence above, the health impact of *D. perseae* to New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

Avocado has become popular and is widely consumed in New Zealand. It is commonly grown in home gardens in some parts of the country. Stem-end rot disease causes rot in avocado and makes the fruit inedible and can cause scarcity in local markets.

Given the arguments and evidence above, the sociocultural impact of *D. perseae* to New Zealand is considered to be low, with low uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, health and society is considered to be low, with low uncertainty.

5.6.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is high with low uncertainty,
- assuming entry, the likelihood of exposure is high with low uncertainty,
- assuming successful exposure, the likelihood of establishment is high with low-moderate uncertainty,
- the overall impact on the New Zealand economy, environment, health and society is considered to be low with low uncertainty.

the overall level of assessed risk to New Zealand from *D. perseae* on imported *Persea americana* budwood (as in the commodity description) is considered to be **low**, with **moderate** uncertainty.

5.6.9 Management considerations

Symptoms caused by *D. perseae* are not likely to be expressed during the growing period in PEQ. *Diaporthe perseae* can occur in healthy looking avocado twigs and stems and be present throughout the life cycle of infected plants without showing visible symptoms; stem-end rot symptoms only develop after fruit harvest (Peterson 1978).

In general pathogenic endophytes such as *D. perseae* can exhibit symptoms under environmental stresses such as drought, hail or frost (Udayanga et al. 2011), but the specific conditions for symptom expressions for *D. perseae* are not well described.

Samples can be taken for testing from any part of the avocado leafless budwood and testing different parts of the budwood might be required. This is because *D. perseae* can occur in healthy looking avocado twigs and stems and be present throughout the life cycle of infected plants without showing visible symptoms (Peterson 1978). However, it is not clear if the pathogen will be evenly distributed or not throughout an infected avocado budwood and that the right bit will be tested. Optimum season or conditions for conducting detection testing has not been specified.

5.6.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Symptoms caused by *D. perseae* is not likely to be expressed during the growing period in PEQ. It can occur in healthy looking avocado twigs and stems and be present throughout the life cycle of infected plants without showing visible symptoms with stem-end rot symptoms developing after fruit harvest (Peterson 1978).

- ***What are the known environmental conditions conducive to symptom expression?***

In general, pathogenic endophytes such as *D. perseae* can exhibit symptoms under environmental stresses such as drought, hail or frost (Udayanga et al. 2011), but the specific conditions for symptom expressions for *D. perseae* are not well described.

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Samples can be taken for testing from any part of the avocado leafless budwood and testing different parts of the budwood might be required. This is because *D. perseae* can occur in healthy looking avocado twigs and stems and be present throughout the life cycle of infected plants without showing visible symptoms (Peterson 1978). It is not clear if it will be evenly distributed or throughout an infected avocado budwood; therefore, there is a high chance of false negatives.

- ***What is the optimum season for conducting detection testing?***

An optimum season or conditions for conducting detection testing has not been identified from the literature.

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5.7 *Diaporthe sterilis*

Diaporthe sterilis is a fungus that has relatively recently been isolated from branch cankers on avocado orchard plants in Italy.

5.7.1 Taxonomic description

Scientific name: *Diaporthe sterilis* L. Lombard, G. Polizzi & Crous (2014)

Order/Family: Diaporthales/Diaporthaceae

5.7.2 Hazard identification

Diaporthe sterilis is not known to be present in New Zealand:

- *Diaporthe sterilis* is not listed in NZOR (2021), Biota NZ (2022) or PPIN (2021)
- *Diaporthe sterilis* is not listed in ONZPR (2021)

Diaporthe sterilis has the potential to establish (and spread) in New Zealand because:

- The host plant, avocado, is grown commercially, and in home gardens in some parts of New Zealand.
- *Vaccinium corymbosum* (blueberry) is also a known host (Lombard et al. 2014) of *D. sterilis* and it is grown commercially in both the North and South Island of New Zealand.
- It is an endophytic pathogenic fungus that can be present on twigs, branches and trunks not showing diseased symptoms (Udayanga et al. 2011).
- Climate is unlikely to be a barrier to its establishment and spread in New Zealand, having been reported to occur in Italy which has a similar climate (CMI ≥ 0.7) to New Zealand (Phillips et al. 2018).
- *Diaporthe sterilis* can spread by human movement of infected nursery stock and movement of infected fruits to avocado growing areas.

Diaporthe sterilis has the potential to cause harm to New Zealand because:

- *Persea americana* is a known host of *D. sterilis* (Guarnaccia et al. 2016) as well as *V. corymbosum* (blueberry)
- *Diaporthe sterilis* can occur endophytically on plant parts (Lombard et al. 2014). *Diaporthe* spp. can occur on twigs, branches and pedicels of avocado plants. From the pedicel, they can eventually colonise the inflorescence of the stem-end tissue and cause stem-end rot (Hartill and Everett 2002).
- Avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$50.6 million (Plant & Food Research 2020).

Diaporthe sterilis is associated with *P. americana* budwood because:

- *Diaporthe sterilis* was isolated from diseased avocado tissues that were showing symptoms of shoot and branch cankers from orchards in Italy and caused stem-end rot disease in fruits during pathogenicity tests (Guarnaccia et al. 2016).

Given the arguments and evidence above, *D. sterilis* meets the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.7.3 Risk assessment

Biology and epidemiology of Diaporthe sterilis

Biology and host range

Diaporthe sterilis was first described when it was isolated from *V. corymbosum* (blueberry) in Italy where it was observed in association with trunk cankers (Lombard et al. 2014). It was associated with avocado in Italy after being isolated from diseased avocado tissues showing symptoms such as shoot and branch cankers as well as stem-end rot disease in fruits from orchards (Guarnaccia et al. 2016). From 94 fungal isolates sampled from avocado orchards in Italy, six pathogenic fungi were identified, and *D. sterilis* was isolated the least frequently (13%). The fungus has also been isolated from dead aerial branches of the flowering plant *Cytisus sp.* in Italy (Dissanayake et al. 2017). Having been recently described, there is limited information about *D. sterilis* and at present it is only known to be associated with three plant species (in three plant families) and reported only in three provinces in Italy (Table 5-10)

Table 5-10: Known distribution and hosts of Diaporthe sterilis.

Continent	Country	State/Region	Host plant	CMI*	Reference
Europe	Italy	Catania Province, Sicily	*Persea americana* (avocado)	0.8	Guarnaccia et al. (2016)
			Vaccinium corymbosum (blueberry) (Ericaceae)	0.8	Lombard et al. (2014)
		Forli-Cesena Province	*Cytisus sp.* (Fabaceae)	0.8–0.9	Dissanayake et al. (2017)

*(Phillips et al. 2018)

Depending on the host and the health of the host plants, most species of *Diaporthe* can exist as harmless endophytes or as pathogens (Udayanga et al. 2011). *Diaporthe sterilis* has been isolated (along with other *Diaporthe* spp.) from both symptomatic and asymptomatic *Vaccinium* spp. plant materials in Italy (Lombard et al. 2014).

Symptoms

Diaporthe sterilis was observed to cause cankers in branches of avocado plants and has been isolated from cankered shoots and branches (Guarnaccia et al. 2016). While *D. sterilis* was not isolated from diseased fruits collected, it was observed to cause stem-end rot disease of fruits when a pathogenicity test was carried out to satisfy Koch's postulates (Guarnaccia et al. 2016). During pathogenicity tests, internal tissues of avocado stems infected with *D. sterilis* were examined after three months and tissue discolouration was observed (Guarnaccia et al. 2016). However, isolates of *Diaporthe* spp. including *D. sterilis* caused less severe symptoms (shoot cankers and stem-end rot) than other fungal species in avocado. This indicates that it is not as virulent as some other pathogenic fungi that cause cankers and stem-end rot diseases (Guarnaccia et al. 2016).

Infected fruits developed brown to dark brown margins at the stem end after 10 days incubation at 25°C. The flesh of the fruit showed discoloured internal vascular bundles and as the rot progressed during fruit ripening, the fruits , became brown and shrivelled (Guarnaccia et al. 2016).

Diaporthe sterilis is also known to cause disease symptoms on *V. corymbosum* (Lombard et al. 2014). It caused cankers at the base of a trunk, green stems and twigs resulting in the development of brown lesions and twig blight. Cankers present at the crown of the plant resulted in mortality (Lombard et al. 2014). Cankerous stems were also observed to have internal discolouration of the vascular tissues. Red to brown spots were also observed on the leaves (Lombard et al. 2014).

Transmission and spread

Members of the genus *Diaporthe* in their endophytic phase can have a latent or dormant phase without showing any symptoms (Dann et al. 2013; Dong et al. 2020). This means that, potentially, symptomless cuttings of avocado can be infected with the fungus. They may be transmitted and spread through grafting especially in commercial avocado plant nurseries. Under unfavourable environmental conditions such as drought, frost or hail, colonised plants can become stressed and begin to exhibit symptoms (Udayanga et al. 2011).

Avocado fruits can be systemically infected with stem-end rot disease caused by pathogenic fungi from the genus *Diaporthe.* However, Guarnaccia et al. (2016) found that *D. sterilis* did not infect avocado fruits with stem-end rot systemically. During the study, *D. sterilis* was not isolated from infected fruits (with stem-end rot symptoms) from the orchard but it caused stem-end rot disease when infected artificially during pathogenicity tests (Guarnaccia et al. 2016). Under field conditions, fruits can get infected during harvest through the pedicel, pedicel scars or wounds (Darvas and Wehner 1987) and can be spread by the use of infected cutting implements during fruit harvest (Hartill and Everett 2002).

Conditions for symptom development

Specific conditions for symptom development of *D. sterilis* has not been specified.

5.7.4 Likelihood of entry

Diaporthe sterilis has so far been reported in association with avocado only in the island of Sicily (Guarnaccia et al. 2016), and a very limited distribution on other hosts on mainland Italy (Lombard et al. 2014; Guarnaccia et al. 2016).

Members of the genus *Diaporthe* are known to be present on leafless and rootless avocado budwood without showing visible symptoms (Udayanga et al. 2011). Given many *Diaporthe* species share similar characteristics such as asymptomatic infection, it is possible that *D. sterilis* may not be detected during export preparation and pre-export phytosanitary inspection to New Zealand or on arrival to New Zealand. Therefore, *D. sterilis* has the potential to be associated with the commodity when it arrives in Post Entry Quarantine (PEQ).

All imported avocado germplasm will undergo PEQ at level 3A, during which the budwood will undergo a minimum of three distinct phenological periods of active growth. However, it is unclear if *D. sterilis* is likely to show symptoms and be detected in Level 3A PEQ. This is because conditions for symptom development of infection by *D. sterilis* have not been specifically described. However, from pathogenicity test, internal tissues of avocado stems were examined after three months and tissue discolouration was observed. Infected fruits developed brown to dark brown margin at the stem end after 10 days incubation at 25℃ (Guarnaccia et al. 2016). The fact that *D. sterilis* has not been observed to produce conidia in culture (Guarnaccia et al. 2016) suggests it is possible that spores might not be produced on seedlings derived from infected budwood in PEQ, but can develop symptoms during the PEQ period. However, molecular diagnostic test might be required to detect asymptomatic infections.

Given the arguments and evidence above, that is:

- *Diaporthe sterilis* has the potential to be present on imported leafless/rootless avocado budwood and may not show visible symptoms.
- Infected budwood may exhibit symptoms during level 3A PEQ. However, this is uncertain because only a single study has been conducted, which found that inoculated avocado plants showed discolouration of internal stem after three months.
- Many *Diaporthe* species share similar characteristics such as asymptomatic infection, so *D. sterilis* may be present on leafless and rootless avocado budwood without showing visible symptoms. Molecular diagnostic testing may be required to detect asymptomatic infections.

the likelihood of *D. sterilis* entering New Zealand associated with *Persea americana* budwood is considered to be high, with **moderate** uncertainty.

5.7.5 Likelihood of exposure

This assessment is made on the basis that *D. sterilis* has entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *D. sterilis*.

If avocado plants are infected with *D. sterilis* at the time of their release from Post Entry Quarantine they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *D. sterilis* in New Zealand from *Persea americana* budwood is considered **high**, with **moderate** uncertainty. The uncertainty is due to the little information available about this species.

5.7.6 Likelihood of establishment

This assessment is made on the basis that *D. sterilis* has been successfully exposed to a suitable host plant in the New Zealand environment.

Diaporthe sterilis is recorded to be present in regions of Italy that have a CMI range of between 0.8-0.9. CMI, suggesting that the conditions in most parts of New Zealand are likely to be suitable for D. *sterilis* to occur and spread. Therefore, climate is not expected to be a barrier to the establishment of *D. sterilis* in New Zealand.

Avocado plants are widely grown in some parts of New Zealand commercially in orchards and in-home gardens mostly in the North Island and some parts of the South Island (Chapter 2). Blueberries are grown in parts of the North and South Island of New Zealand (Coriolis 2020), Therefore, suitable hosts are available in both the North and South Island of New Zealand for *D. sterilis* to establish.

Given the arguments and evidence above, that is:

- The climatic conditions in most parts of New Zealand are likely to be suitable for D*. sterilis* to establish.
- Suitable host plants (avocado and blueberry) are available in the North and South Island of New Zealand.
- Uncertain why D*. sterilis* hasn't yet spread to different parts of Europe where hosts plants are available and there are limited controls on plant movement.

the likelihood of *D. sterilis* establishing in New Zealand is considered **moderate**, with **moderate** uncertainty. The uncertainty is due to the little information available about this species.

5.7.7 Impacts in New Zealand

Diaporthe sterilis has so far been reported to be present in Italy in association with avocado, blueberry and *Cytisus* sp. (Lombard et al. 2014; Guarnaccia et al. 2016). It was isolated from cankers on avocado branches on one occasion (Guarnaccia et al. 2016). It was also reported to cause cankers on trunks, twigs and stems of blueberry plants in Italy (Lombard et al. 2014).

Diaporthe sterilis is likely to be spread by the movement of infected nursery stock for planting. Avocado plants are widely grown in some parts of New Zealand commercially in orchards and in-home gardens mostly in the North Island and some parts of the South Island (New Zealand Avocado 2021). Blueberries are grown in parts of the North and South Island of New Zealand (Coriolis 2020), Therefore, suitable hosts are available in both the North and South Island of New Zealand for *D. sterilis* to spread.

Economic impacts

Diaporthe species including *D. sterilis* was observed to cause stem/shoot cankers of avocado in Italy (Guarnaccia et al. 2016), however, It was one of the least frequent and virulent fungus obtained from diseased trunks and shoots (Guarnaccia et al. 2016). Reports of this fungus affecting avocado fruits by causing stem-end-rot, have come from artificial infection i.e., pathogenicity tests (Guarnaccia et al. 2016), but there are no reports of this *D. sterilis* naturally infecting avocado fruits. *D. sterilis* is likely to cause less severe disease symptoms in avocado compared to other fungal pathogens such as *Colletotrichum gloeosporioides,* and *Phomopsis* spp. that are already present and common in New Zealand (Hartill and Everett 2002). This suggests that while *D. sterilis* can result in some postharvest losses of avocado fruits, overall losses may not differ from current levels of stem-end rot diseases in New Zealand.

Infections by *D. sterilis* may affect market access of exports of avocado to other parts of the world where *D. sterilis* is not reported to be present. Because *D. sterilis* is reported to cause diseases in avocado in only one country, its presence in New Zealand can potentially result in trade barriers to countries where it is has not been reported to be present.

Diaporthe sterilis causes disease symptoms on blueberries; it caused cankers at the base and brown lesions on stems resulting in twig blight (Lombard et al. 2014). The blueberry industry is a growing industry in New Zealand with an estimate of NZ$70-80 million in both domestic and international trade in 2020 (Coriolis 2020). While *D. sterilis* has the potential to affect blueberry production in New Zealand and can lead to reduced yield, its overall impact on blueberry production has not been quantified.

Given the arguments and evidence above, that is:

- *D. sterilis* can cause trunk cankers and potentially result in some postharvest losses of avocado. Overall losses may not differ from current levels of losses caused by other pathogenic fungi on avocado already present in New Zealand.
- *Vaccinium corymbosum* is a known host of *D. sterilis.* The potential impacts on blueberry production have not been assessed and quantified but could potentially affect blueberry production in New Zealand and lead to reduced yield. In 2020, blueberry industry in New Zealand was estimated to be worth between NZ$70-80 million in both domestic and international trade.

the economic impact of *D. sterilis* to New Zealand is considered to be **very low** with **moderate** uncertainty.

Environmental impacts

Infection by *D. sterilis* has only been demonstrated to be associated with avocados, blueberries *and Cytisus* sp. so far (Lombard et al. 2014; Guarnaccia et al. 2016). Although it has only been recorded in three host plants, it is likely it can infect other plants. Having been associated with *Cytisus* sp. in Italy (Dissanayake et al. 2017), it can potentially infect and be an inoculum source (that can infect other plant species) in the two similar weed species (*Cytisus scoparius* and *Genista monspessulana*) which have naturalised and are widely spread in New Zealand (Weedbusters 2022).

Searches from Google, Google scholar, **U.S. National Fungus Collections Fungus-Host Distributions Database,** CABI and EPPO using the search term '*Diaporthe sterilis* and the environment' found no record of *D. sterilis* causing environmental impacts.

Given the arguments and evidence above, the impact on the environment from the establishment of *D. sterilis* in New Zealand is considered to be ***very low***, with ***moderate*** uncertainty.

Human health impacts

Searches using the search term '*Diaporthe sterilis* impacts on health' from Google, Google scholar, CABI and Pubmed found no record of *D. sterilis* causing any health impact on humans and animals.

Given the arguments and evidence above, the human health impact of *Diaporthe sterilis* to New Zealand is considered to be ***negligible*** with ***low*** uncertainty.

Sociocultural impacts

Avocado has become popular and is widely consumed in New Zealand. It is commonly grown in home gardens in some parts of the country. *Diaporthe sterilis* can potentially cause diseases such as stem-end rot disease. This can result in rot of avocado fruits making them inedible and can lead to reduced yield for home gardeners and scarcity in local markets. Blueberry is also grown in commercial quantities and widely consumed in New Zealand. The impact of *D. sterilis* on avocado has not been assessed and quantified.

Given the arguments and evidence above, the sociocultural impact of *D. sterilis* to New Zealand is considered to be **very *low***, with ***low*** uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, human health and society is considered to ***very low*** with ***moderate*** uncertainty.

5.7.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is **high**, with **moderate** uncertainty
- assuming entry, the likelihood of exposure is **high**, with **moderate** uncertainty
- assuming successful exposure, the likelihood of establishment is **moderate**, with **moderate** uncertainty.
- the overall impact on the New Zealand economy, environment, human health and society is considered to be **very low**, with **moderate** uncertainty.

the overall level of assessed risk to New Zealand from *D. sterilis* on imported *P. americana* budwood (as in the commodity description) is considered to be **low**, with **moderate** uncertainty.

5.7.9 Management considerations

Diaporthe sterilis was observed in association with cankers in branches of avocado plants when it was isolated along with other fungi on cankered shoots and branches. It was not isolated from diseased fruits collected during this study, but it caused stem-end rot disease of fruits when a pathogenicity test was carried out to satisfy Koch's postulates (Guarnaccia et al. 2016).

Infected fruits developed brown to dark brown margins at the stem end after 10 days incubation at 25℃. The flesh of the fruit showed discoloured vascular bundles and the rot progressed as the fruit ripened, it generally became brown and shrivelled (Guarnaccia et al. 2016).

Environmental conditions conducive for development of trunk cankers caused by *D. sterilis* have not been specified. During pathogenicity test, fruits infected with *D. sterilis* developed dark to brown margins at the stem end after 10 days incubation at 25℃ (Guarnaccia et al. 2016).

Samples can be taken for testing from any part of the avocado leafless budwood and testing different parts of the budwood might be required. Because, like other *Diaporthe* species, it can be present on twigs, branches and trunks not showing diseased symptoms (Udayanga et al. 2011).

Optimum season for conducting detection has not been specified.

5.7.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

D. sterilis was isolated along with other fungi from cankered shoots and branches of avocado plants. It was not isolated from diseased fruits collected during this study, but it caused stem-end rot disease of fruits when a pathogenicity test was carried out to satisfy Koch's postulates (Guarnaccia et al. 2016).

- ***What are the known environmental conditions conducive to symptom expression?***

Environmental conditions conducive for development of trunk cankers caused by *D. sterilis* have not been specified. During pathogenicity test, fruits infected with *D. sterilis* developed brown to dark brown margin at the stem end after 10 days incubation at 25℃ (Guarnaccia et al. 2016).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Samples can be taken for testing from any part of the avocado leafless budwood and testing different parts of the budwood might be required. Because like other *Diaporthe's* it can be present on twigs, branches and trunks not showing diseased symptoms (Udayanga et al. 2011).

- ***What is the optimum season for conducting detection testing?***

Optimum season for conducting detection has not been specified.

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5.8 *Dothiorella aromatica*

Dothiorella aromatica is a fungal plant pathogen that causes stem-end rot (postharvest) disease in avocado. It has also been associated with stem-end rot disease in mangos.

5.8.1 Taxonomic description

Scientific name: *Dothiorella aromatica* (Sacc.) Petr. & Syd. (1927)

Order/Family: Botryosphaeriales/ Botryosphaeriaceae

Other names include: *Macrophoma aromatica* Sacc. (1915)

5.8.2 Hazard identification

Dothiorella aromatica is not known to be present in New Zealand.

- *Dothiorella aromatica* is not listed in PPIN (2021)
- *Dothiorella aromatica* is recorded as absent from New Zealand in Biota NZ (2022)
- *Dothiorella aromatica* is a regulated pest for New Zealand, has unwanted status but is not a notifiable organism in ONZPR (2021).

Dothiorella aromatica has the potential to establish (and spread) in New Zealand because:

- the host plant, avocado is grown commercially, and in home gardens in some parts of New Zealand.
- it is an endophytic pathogenic fungus, which has been isolated from twigs, stems, and inflorescence of asymptomatic avocado plants. It can be present on all parts of an infected tree (Johnson et al. 1992).
- climate is unlikely to be a barrier to its establishment and spread in New Zealand because it occurs in Australia and South Africa, which have a similar climate (CMI) ≥ 0.7 to New Zealand (Phillips et al. 2018).
- *Dothiorella aromatica* can spread naturally by wind and rain or by human movement of infected nursery stock and movement of infected fruits to avocado growing areas.

Dothiorella aromatica has the potential to cause harm to New Zealand because:

- *Dothiorella aromatica* has been associated with and shown to cause postharvest (stem-end) rot disease in avocado (Darvas and Kotzé 1981; Darvas and Kotze 1987b; Korsten et al. 1994).
- postharvest rot diseases caused by *D. aromatica* can affect the marketability of avocado crops in New Zealand. It is an important commercial crop in New Zealand worth up to $110 million in domestic sales and up to 56.6 million in domestic sales (Plant & Food Research 2019).

Dothiorella aromatica is associated with *P. americana* budwood because:

- *Persea americana* is a known host of *D. aromatica* (Peterson 1978; Darvas and Kotzé 1981; Darvas and Kotze 1987b; Darvas and Wehner 1987);
- *Dothiorella aromatica* can occur endophytically on twigs, branches and pedicels of and infected avocado plant (Darvas and Kotze 1987b). It can eventually colonise the inflorescence from the pedicel of the stem-end tissue and cause stem-end rot (Hartill and Everett 2002).

Given the arguments and evidence above, *D. aromatica* meets the criteria to be a hazard on *Persea americana* budwood (as per the commodity description) imported to New Zealand.

5.8.3 Risk assessment

Biology and epidemiology of Dothiorella aromatica

Biology and host range

Depending on the host and the health of the host plants, species from the family Botryosphaeriaceae can exist as harmless endophytes or as pathogens (Udayanga et al. 2011). *Dothiorella aromatica* can be an endophytic (can inhabit plant tissues without showing visible symptoms) and pathogenic fungus (Johnson et al. 1992; Wilson 1995) that is known to cause stem-end rot disease and post-harvest rot (Kotzé and Darvas 1985; Darvas and Kotze 1987b). *Dothiorella aromatica* can occur on twigs, branches and pedicels of avocado plants (Darvas and Kotze 1987b; Johnson et al. 1992). From the pedicel, it can eventually colonise the inflorescence (flowers) of the of the stem-end tissue and cause stem-end rot (Hartill and Everett 2002).

Stem-end rot is often reported to be caused by several fungal pathogens (Darvas and Kotze 1987b; Korsten et al. 1994; Guarnaccia et al. 2016). *Dothiorella aromatica* was one of the most frequently isolated pathogens from avocado exhibiting stem-end rot disease symptoms collected from avocado producing areas in South Africa (Korsten et al. 1994) (Table 5-11).

Table 5-11: *Dothiorella aromatica* host and geographic range, and disease symptoms

Continent	Country	State/Region	Host plants	Symptoms /disease	CMI*	Reference
Oceania	Australia	Queensland	*Persea americana* (avocado)	Stem-end rot	0.7	(Peterson 1978)
			Magnifera indica (Mango)	Stem-end rot	0.6 - 0.9	(Johnson et al. 1992)
Africa	South Africa		*Persea americana* (avocado)	Stem-end rot	0.6 - 0.8	(Darvas and Kotzé 1978; Darvas and Kotze 1987a)

*(Phillips et al. 2018)

Reproduction and spread

Fruiting bodies of *D. aromatica* are rarely seen on twigs and branches of avocado plants, but have been observed in abundance on fallen fruits on the ground and can also be observed on dead branches or barks (Darvas and Kotze 1987b). These fallen fruiting bodies usually can serve as a source of primary infection (Dann et al. 2013) and can be transferred to open wounds on plants, the pedicel, and fruits, and can potentially be spread by rain splashes or wind (Menge and Ploetz 2003).

Most stem-end rot infections occur during harvest through the pedicel, pedicel scars or wounds (Darvas and Wehner 1987). Infections can occur through wounds on the bark of the tree or wounds created through picking of fruit in the field (Kotzé and Darvas 1985; Hartill and Everett 2002). It can also be spread by the use of the same cutting implement from an infected plant to a non-infected plant (Hartill and Everett 2002).

Dothiorella aromatica, in its endophytic phase, can be latent or dormant without showing any symptoms (Johnson et al. 1992). This means that symptomless cuttings of avocado plants infected with *D. aromatica* can be transmitted and spread through grafting especially in commercial avocado plant nurseries.

Infection and spread of *D. aromatica* (as well as other pathogenic fungi associated with avocado) occurs mostly during prolonged wet periods of rain and high humidity (Darvas and Kotzé 1978; Peterson 1978; Darvas et al. 1987). Peterson (1978) found that natural infection of fruits did not occur in dry conditions but occurred after four or more conservative days of rainfall.

Symptoms

Dothiorella aromatica can have a latent or dormant phase of infection within the plant tissues (Wilson 1995). Hence, it can be present throughout the life cycle of infected plants without showing visible symptoms. Symptoms of *D. aromatica* such as lesions and rot, which produces spores, tend to only occur after harvest (Darvas and Kotzé 1978; Peterson 1978). This means that in its latent phase, *D. aromatica* can occur in healthy looking avocado twigs and stems, with no visible symptoms.

In its pathogenic phase, *D. aromatica* colonises the xylem and the phloem at the stem-end of the fruit (Karunanayake and Adikaram 2020). *Dothiorella aromatica* eventually becomes necrotrophic (secretes enzymes that kill the host tissues while living on the nutrients from the dead tissues), causing stem-end rot by colonising the fruit pulp (Karunanayake and Adikaram 2020). Infection can remain latent (not show symptoms) prior to harvest with symptoms only becoming apparent after harvest (Peterson 1978). Immature fruits are more vulnerable to stem-end rot infections than mature fruits (Kotzé and Darvas 1985).

Stem-end rot symptoms develop after harvest, at the onset of fruit ripening (Darvas and Kotzé 1978; Darvas et al. 1987; Karunanayake and Adikaram 2020). Visible symptoms begin as brownish or black rot at the fruit pedicel which progresses internally causing discolouration and decay. Internal decay may extend on the external part of the fruit leading to brown or black discolouration (Darvas and Wehner 1987; Karunanayake and Adikaram 2020). Mycelium and abundant spores might be observed on the abscission scar of infected fruit and can eventually cover the fruit as ripening progresses, leading to softening and shrivelling of the entire fruit (Menge and Ploetz 2003).

Pathogenicity tests using avocado fruits showed that all artificially infected fruits developed symptoms (lesions) 10 days post inoculation (Peterson 1978; Kotzé and Darvas 1985).

Conditions for symptom development

Infection and spread of *D. aromatica* (as well as other pathogenic fungi associated with avocado) occurs mostly during prolonged wet periods of rain and high humidity (Darvas and Kotzé 1978; Peterson 1978; Darvas et al. 1987). Peterson (1978) found that natural infection of fruits did not occur in dry conditions but occurred after four or more conservative days of rainfall. Under laboratory conditions, temperature was observed to affect spore germination of *D. aromatica*, with the minimum and maximum temperature limits of 10-35℃ (Denner et al. 1986) and an optimum growth temperature of 25℃ (Schoeman 2005).

5.8.4 Likelihood of entry

Dothiorella aromatica has been reported to be associated with avocado in South Africa and Australia and is associated with mangos in Australia (Darvas and Kotzé 1978; Peterson 1978; Johnson et al. 1992).

In its endophytic phase, it can be present inside leafless and rootless avocado budwood without showing visible symptoms (Johnson et al. 1992). Therefore, it is likely it will not be detected during export preparation and pre-export phytosanitary inspection to New Zealand or on arrival to New Zealand.

All imported avocado germplasm will undergo PEQ at level 3A, during which the budwood will undergo a minimum of three distinct phenological periods of active growth. The period in Level 3A PEQ is not likely to impact the likelihood of entry of *D. aromatica*. Seedlings derived from infected budwood grafted in PEQ level 3A are not likely to show visible symptoms within the time in PEQ. This is because *D. aromatica* has only been reported to cause symptoms (stem-end rot disease) on avocado fruits (Peterson 1978; Darvas et al. 1987), therefore unlikely to produce visible symptoms. Fruiting bodies of *D. aromatica* are rarely seen on twigs and branches of avocado plants, but have been observed in abundance on fallen fruits on the ground and can also be observed on dead branches or barks (Darvas et al. 1987). It is, highly unlikely that spores will be produced on seedlings derived from infected imported budwood and escape from PEQ without being detected. As such, infected seedlings are likely to remain undetected unless molecular diagnostic testing for *D. aromatica* is carried out.

Given the arguments and evidence above, that is:

- it can be present on leafless and rootless avocado budwood without showing visible symptom
- infected budwood that is grafted in PEQ level 3A is not likely to show any visible symptoms within the time in PEQ if the fungus is in a latent phase.
- infected plants are likely to remain undetected unless molecular diagnostic testing for *D. aromatica* is carried out.

the likelihood of *D. aromatica* entering New Zealand associated with *Persea americana* budwood is considered to be high, with low uncertainty.

5.8.5 Likelihood of exposure

This assessment is made on the basis that *D. aromatica* has entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *D. aromatica*.

If avocado plants are infected with *D. aromatica* at the time of their release from post entry quarantine, they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *D. aromatica* in New Zealand from *Persea americana* budwood is considered high, with low uncertainty.

5.8.6 Likelihood of establishment

This assessment is made on the basis that *D. aromatica* has been successfully exposed to a suitable host plant in the New Zealand environment.

Dothiorella aromatica is recorded to be present in Australia (Queensland) and South Africa which have a Composite match index (CMI) range of between 0.6-0.8. A CMI of ≥ 0.7 indicates a similar climate to the whole of New Zealand (Phillips et al. 2018). As such, the conditions in some parts of New Zealand are likely to be suitable for *D. aromatica* to occur and spread. Therefore, climate is not considered to be a barrier to the establishment of *D. aromatica* in New Zealand.

Avocado plants are widely grown in some parts of New Zealand commercially in orchards and in-home gardens mostly in the North Island and some parts of the South Island (New Zealand Avocado 2021), therefore suitable hosts are available in those areas. *Dothiorella aromatica* can spread by the movement of infected nursery stock for planting. However, its spread might be limited by host availability because it has only been recorded to be associated with avocado (Darvas and Kotzé 1978) and mangoes (Peterson 1978; Johnson et al. 1992). However, mangos are not grown in New Zealand; therefore, it is likely that *D. aromatica* will be restricted to avocado growing areas of New Zealand.

Spores of *D. aromatica* are usually produced on rotting fruits and have been observed in abundance on fallen fruits on the ground (Darvas et al. 1987). This suggests that infected fruit that is discarded near avocado plants could be a source of inoculum. Therefore, *D. aromatica* could be spread to new areas by human movement of infected fruit.

Given the arguments and evidence above, that is:

- the climatic conditions in some parts of New Zealand are likely to be suitable for D*. aromatica* to occur and spread.
- suitable avocado host plants are available in orchards and in-home gardens mostly in the North Island and some parts of the South Island. *D. aromatica* can spread by the movement of infected nursery stock for planting.
- spores of *D. aromatica* can be present on infected fruit and can be spread to new areas by human movement of infected fruit.

the likelihood of *D. aromatica* establishing in New Zealand is considered high, with low-moderate uncertainty.

5.8.7 Impacts in New Zealand

Dothiorella aromatica have been reported to be associated with avocado and mangoes, causing stem-end rot (postharvest) disease (Darvas and Wehner 1987; Johnson et al. 1992). *Dothiorella aromatica* can occur in plant tissue without showing symptoms (Johnson et al. 1992). As such, symptomless cuttings of avocado plants infected with *D. aromatica* can be transmitted and spread through grafting especially in commercial avocado plant nurseries. Fruiting bodies of *D. aromatica* have been observed in abundance on fallen fruits on the ground and on dead branches or barks (Darvas and Kotze 1987b). These fallen fruiting bodies usually can serve as a source of primary infection (Dann et al. 2013) and can be transferred to open wounds on plants, the pedicel, and fruits, and can potentially be spread by rain splashes or wind (Menge and Ploetz 2003).

Economic impacts

Dothiorella aromatica causes stem-end rot disease symptoms that can go unnoticed and mainly become apparent after harvest. Stem-end rot symptoms become apparent at the onset of fruit ripening (Karunanayake and Adikaram 2020). Low temperatures can mask symptoms on infected fruit (Darvas 1982), rot symptoms can become apparent outside those stem-end rot caused by *Colletotrichum gloeosporioides* and *Phomopsis* spp. are also common in New Zealand (Hartill and Everett 2002). This suggests that while *D. aromatica* can result in some postharvest losses of avocado fruits, it is likely to be managed by present practices in place for managing stem-end rot disease in New Zealand. Post-harvest disease caused by *D. aromatica* can be controlled to an extent by pre-harvest fungicide sprays such as benomyl, captafol, Cu-oxychloride and Cu-hydroxide (Darvas and Kotze 1987a). Post-harvest disease caused by *D. aromatica* can be controlled to an extent by pre-harvest fungicide sprays such as benomyl, captafol, Cu-oxychloride and Cu-hydroxide (Darvas and Kotze 1987a). Overall losses may not differ from current levels of stem-end rot diseases.

Infections by *D. aromatica* can affect market access of exports of avocado to other parts of the world where *D. aromatica* is not reported to be present (Table 5-11). The fact that it is recorded as present in Australia, which is New Zealand's major export market, suggests it is not likely to have a significant impact on export. But its presence in New Zealand can potentially result in trade barriers to other countries where it is has not been reported to be present.

Given the arguments and evidence above, that is:

- *Dothiorella aromatica* can result in some postharvest losses of avocado, but overall losses may not differ from current levels of stem-end rot disease in New Zealand.
- current methods of control of other stem-end rot causing pathogens in New Zealand may also be effective for controlling disease caused by *D. aromatica*.
- the establishment of *D. aromatica* is not likely to affect market access of export of avocado to Australia (which is one of New Zealand's major export market) but could potentially affect market access of export to other parts of the world where it is not reported to be present.

the economic impact of *D. aromatica* to New Zealand is considered to be **low**, with **low** uncertainty.

Environmental impacts

Dothiorella aromatica has been reported to infect avocado and mangos (Darvas and Kotzé 1978; Peterson 1978; Johnson et al. 1992). Searches from Google, Google scholar, U.S. National Fungus Collections Fungus-Host Distributions Database, CABI and EPPO using the search term '*Dothiorella aromatica* and the environment' found no record of *D. aromatica* infecting other plants. This suggests that *D. aromatica* is not likely to affect native plants and the environment.

Given the arguments and evidence above, the impact on the environment from the establishment of *D. aromatica* in New Zealand is considered to be very low, with low uncertainty.

Human health impacts

Searches using the term '*Dothiorella aromatica* impacts on health' from Google, Google scholar, CABI and Pubmed found no record of *D. aromatica* causing any health impact on humans and animals.

Given the arguments and evidence above, the health impact of *D. aromatica* to New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

Avocado has become popular and is widely consumed in New Zealand. It is commonly grown in home gardens in some parts of the country. Stem-end rot disease causes rot in avocado and makes the fruit inedible and can cause scarcity in local markets.

Given the arguments and evidence above, the sociocultural impact of *D. aromatica* to New Zealand is considered to be low, with low uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, health and society is considered to be low, with low uncertainty.

5.8.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is high with low uncertainty,
- assuming entry, the likelihood of exposure is high with low uncertainty,
- assuming successful exposure, the likelihood of establishment is high with low-moderate uncertainty,
- the overall impact on the New Zealand economy, environment, health and society is considered to be low with low uncertainty.

the overall level of assessed risk to New Zealand from *D. aromatica* on imported *Persea americana* budwood (as in the commodity description) is considered to be **low**, with **moderate** uncertainty.

5.8.9 Management considerations

Symptoms caused by *D. aromatica* are not likely to be expressed during the growing period in PEQ. It can occur in healthy looking avocado twigs and stems and be present throughout the life cycle of infected plants without showing visible symptoms; stem-end rot symptoms only develop after fruit harvest (Peterson 1978).

Infection and spread of *D. aromatica* (as well as other pathogenic fungi associated with avocado) occurs mostly during prolonged wet periods of rain and high humidity (Darvas and Kotzé 1978; Peterson 1978; Darvas et al. 1987). Peterson (1978) found that natural infection of fruits did not occur in dry conditions but occurred after four or more conservative days of rainfall. Under laboratory conditions, germination of *D. aromatica* spore were affected by temperature, with a suitable growing range of 10-35°C (Denner et al. 1986) and an optimum growth temperature of 25°C (Schoeman 2005).

Infection and spread of *D. aromatica* (as well as other pathogenic fungi associated with avocado) occurs mostly during prolonged wet periods of rain and high humidity (Darvas and Kotzé 1978; Peterson 1978; Darvas et al. 1987). Peterson (1978) found that natural infection of fruits did not occur in dry conditions but occurred after four or more conservative days of rainfall.

Samples can be taken for testing from any part of the avocado leafless budwood and testing different parts of the budwood might be required. This is because *D. aromatica* can occur in healthy looking avocado twigs and stems and be present throughout the life cycle of infected plants without showing visible symptoms (Peterson 1978). However, it is not clear if the pathogen will be evenly distributed or not throughout an infected avocado budwood and that the right bit will be tested.

Post-harvest disease caused by *D. aromatica* can be controlled to an extent by pre-harvest fungicide sprays such as benomyl, captafol, Cu-oxychloride and Cu-hydroxide (Darvas and Kotze 1987a). Optimum season or conditions for conducting detection testing has not been specified.

5.8.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Symptoms caused by *D. aromatica* are unlikely to be expressed during the growing period in PEQ. *Dothiorella aromatica* can occur in healthy looking avocado twigs and stems and can be present throughout the life cycle of infected plants without showing visible symptoms, with stem-end rot symptoms (which is the only symptom it has been reported to cause) developing in immature fruit on the tree and ripening fruit post-harvest (Peterson 1978; Darvas and Wehner 1987).

- ***What are the known environmental conditions conducive to symptom expression?***

In general, pathogenic endophytic fungi can exhibit symptoms under environmental stresses such as drought, hail or frost (Udayanga et al. 2011). However, because *D. aromatica* causes postharvest rot which only develops during fruit ripening (Darvas and Kotzé 1978; Darvas and Kotze 1987a), conditions conducive for symptom expression in avocado seedlings derived from budwood infected with *D. aromatica* have not been specified. This is because they are not likely to express symptoms within the level 3A PEQ period. However, under laboratory conditions, germination of *D. aromatica* spores occur within a range of 10-35°C (Denner et al. 1986) and optimum growth occurs at 25°C (Schoeman 2005).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Samples can be taken for testing from any part of the avocado leafless budwood and testing different parts of the budwood might be required. This is because *D. aromatica* can occur in healthy looking avocado twigs and stems and be present throughout the life cycle of infected plants without showing visible symptoms (Peterson 1978). It is not clear if it will be evenly distributed throughout an infected avocado budwood; therefore, there is a high chance of false negatives.

- ***What is the optimum season for conducting detection testing?***

An optimum season or conditions for conducting detection testing has not been identified from the literature.

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5.9 *Elsinoe perseae* (avocado scab)

Elsinoe perseae (avocado scab) is a fungal pathogen that causes brown scabby lesions on fruit, leaves and stems of avocado plants. It can reduce avocado fruit yield, quality and marketability.

5.9.1 Taxonomic description

Scientific name: *Elsinoe perseae* (Jenkins) Rossman & W.C. Allen (2016)

Order/Family: Myriangiales/ Elsinoaceae

Other names include: *Sphaceloma perseae* Jenkins (1934), avocado scab

Taxonomic notes: *Elsinoe perseae* was originally confused with *E. fawcettii*, the causal agent of citrus scab. However, Jenkins (1934) showed it to be a distinct species and demonstrated in pathogenicity tests that *E. perseae* does not infect citrus.

5.9.2 Hazard identification

Elsinoe perseae is not known to be present in New Zealand:

- however in the past *Sphaceloma perseae* (a synonym of *E. perseae*) was recorded in error (NZFungi2 2021). For example, Hartill (1991) misidentified *E. perseae* as a postharvest disease on avocado, and that work was cited recently by Fan et al. (2017). It is not listed in PPIN (2021) under either name.
- *Elsinoe perseae* is a regulated pest for New Zealand (ONZPR 2021), however it is listed in ONZPR under the synonym *Sphaceloma perseae*.

Elsinoe perseae has the potential to establish (and spread) in New Zealand because:

- Hosts are likely to be available, at least in some areas of New Zealand. The reported host of *E. perseae* is avocado (*Persea americana*) and other *Persea* spp., and avocado plants are commonly grown in New Zealand, in home gardens and commercial production, mainly in the north of the North Island.
- The climate is unlikely to limit its development and spread: *Elsinoe perseae* is found in some areas such as Rio Grande do Sul, Brazil and parts of Argentina and South Africa (CABI 2021; EPPO 2021) which have a Climate Match Index (CMI) ≥ 0.7 indicating a similar climate to the whole of New Zealand (Phillips et al. 2018).

Elsinoe perseae has the potential to cause harm to New Zealand because:

- It causes scabby lesions on avocado fruit, reducing their marketability and value, and sometimes causing loss of yield due to premature fruit drop (Stevens and Piper 1941; Ploetz et al. 2011; Everett and Siebert 2018).
- Avocados are an important commercial crop in New Zealand. In the 2018–2019 financial year, export sales of fresh avocados earned NZ$110 million and domestic sales earned NZ$ 56.5 million (Plant & Food Research 2019).

Elsinoe perseae is associated with *Persea americana* budwood because:

- avocado (*P. americana*) is commonly reported as the host of *E. perseae* (CABI 2021; EPPO 2021);
- *Elsinoe perseae* causes lesions on avocado shoots and twigs appearance (Jenkins 1934; Stevens and Piper 1941).

Given the arguments and evidence above, *E. perseae* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.9.3 Risk assessment

***Biology and epidemiology of* Elsinoe perseae**

Table 5-12. The geographical distribution of *Elsinoe perseae* (CPC 2021; EPPO 2021)

Region	Country	**State/Province** (if recorded)	CMI range*
Africa	Guinea		0.4–0.5
	Morocco		0.5–**0.8**
	South Africa		0.5–**0.9**
	Zambia		0.5–0.6
	Zimbabwe		0.5–**0.7**
Asia	Philippines		0.3–0.4
	Taiwan		0.4–**0.8**
North America and the Caribbean	Barbados		0.3–0.6
	Bermuda		Not determined
	Costa Rica		0.4–0.5
	Cuba		0.5–0.6
	Dominica		Not determined
	Dominican Republic		0.5–**0.7**
	El Salvador		0.3–0.5
	Guadeloupe		Not determined
	Guatemala		0.3–0.6
	Haiti		0.5
	Honduras		0.4–0.6
	Jamaica		0.5–0.6
	Mexico		0.4–**0.8**
	Nicaragua		0.4–0.6
	Panama		0.5–0.6
	Puerto Rico		0.5–**0.8**
	USA (restricted distribution)	Florida	0.5–**0.8**
		Texas	0.6–**0.8**
South America	Argentina (restricted distribution)	Jujuy (Aguirre 2003)	0.6–**0.8**
		Tucumán	0.6–**0.9**
	Brazil (restricted distribution)	Espirito Santo	0.5–0.6
		Rio Grande do Sul	**0.7–0.9**
	Guyana		0.4–0.5
	Peru		0.4–**0.8**
	Venezuela		0.4–**0.8**

* Climate Match Index (CMI) with all of New Zealand (Phillips et al. 2018).

Hosts

Elsinoe perseae is widely reported as a pathogen of *Persea* species, mainly *Persea americana* (e.g. CPC 2021; EPPO 2021 and other references cited in this PRA). Specimens recorded as '*Elsinoe perseae*' or '*Sphaceloma perseae*' in the Kew Royal Botanic Gardens collection are mainly from *P. americana*, with some records from *Persea drymifolia* or *Persea gratissima* (HerbIMI 2021), which are synonyms of *P. americana*. Farr and Rossman (2021) record *P. americana*, *P. gratissima* and a *Persea* sp. as hosts. The USDA fungal collection records numerous specimens collected from *P. americana*, two specimens from *P. drymifolia* and several specimens collected from *Persea* spp. (Farr and Rossman 2021). All the *E. perseae* or *S. perseae* specimens in the Kew and USDA collections were collected from *Persea* species.

Elsinoe perseae causes severe scab disease in some avocado cultivars, while others are less affected (Stevens and Piper 1941; Ploetz et al. 2011). For example, the Lula cultivar is now seldom planted in Florida because it is extremely sensitive to *E. perseae*, and the Hass and Fuerte cultivars are moderately susceptible (Ploetz et al. 2011).

Searches found a single record of *E. perseae* on pineapple (*Ananas comosus*) in the USDA fungal database (Farr and Rossman 2021). However, this is highly likely to be a mistaken identification, given that there are no other records of *E. perseae* or *S. perseae* from pineapple or other plant hosts apart from *Persea* species.

Geographical distribution

The recorded distribution of *E. perseae* is shown in Table 5-12.

Symptoms

Elsinoe perseae infects young leaves, stems and fruit of avocado, causing scab like lesions up to 3 mm in diameter (Jenkins 1934). Scab lesions may be isolated, or many spots may coalesce (Stevens and Piper 1941; Ploetz et al. 2011). Leaves may become stunted, crinkled and distorted (Ploetz et al. 2011).

Lesions on young leaves begin as dark red spots that become brown to almost black as the leaf ages (Jenkins 1934). The tissue at the centre of older leaf spots is dry and dead and may fall out, leaving holes in the leaves (Jenkins 1934).

On the skin of fruit, *E. perseae* causes dark brown, raised oval spots or lesions (Jenkins 1934). Severe infections cause leaf-curling and distort twigs and fruit (Stevens and Piper 1941). In severe infections, spots may merge to form irregular areas of hard tissue over most of the fruit surface (Stevens and Piper 1941) (**Error! Reference source not found.**). As the size of the fruit increases, cracks and fissures may develop in the affected areas (Stevens and Piper 1941).

On green bark of twigs and petioles, the spots are raised, darker purple and have a smoother surface (Stevens and Piper 1941).

Figure 5-2 Scab lesions on avocado fruit (Fructibus, CC0, via Wikimedia Commons; https://commons.wikimedia.org/wiki/File:3_x_Avocado_scab_2017_A.jpg; accessed 22/6/2021)

Reproduction and transmission

Conidiophores (asexual spore-bearing structures) of *E. perseae* are produced in acervuli (open saucer-shaped fruiting structures that rupture the epidermis of host organs) in lesions on leaves, fruit or stems (Jenkins 1934; Stevens and Piper 1941). Jenkins (1934) observed that the abundant conidiophores and conidia formed a dense velvety olive to brown covering over the lesions, which gradually weathered away, until only occasional conidiophores remained in lesions on four-month-old fruit.

Under cool moist weather conditions, abundant conidia are produced in scab lesions (Stevens and Piper 1941). The optimum temperature for release of conidia is from 8 to 28 ℃, and fewer conidia were found in the environment at temperatures below 8 ℃ (Marroquín-Pimentel 1999). The optimum conditions for lesion development are a relative humidity higher than 80% and a temperature between 10 – 26 ℃ (Marroquín-Pimentel 1999). The number of new lesions decreases at temperatures below 8 ℃ or above 30 ℃ (Marroquín-Pimentel 1999). Spores may be produced repeatedly in old lesions when environmental conditions are suitable (Stevens and Piper 1941). Scab lesions on old leaves, twigs or fruit remaining on the tree provide inoculum for infection in the following year (Stevens and Piper 1941; Ploetz et al. 2011).

Mature leaves and fruit are not susceptible to new infections with *E. perseae* (Stevens and Piper 1941). Leaves become resistant to infection when the leaf hardens (Stevens and Piper 1941), approximately one month after they emerge (Ploetz et al. 2011). The upper surface of the avocado leaf is more highly susceptible to infection by *E. perseae* than the lower surface (Jenkins 1934). Fruit become resistant at between a third and half their final size (Stevens and Piper 1941; Ploetz et al. 2011).

Injuries caused by thrips provide entry wounds for *E. perseae* (Ploetz et al. 2011). In Mexico, damage by thrips (most commonly *Scirtothrips* and *Frankliniella* spp.) and the appearance of *E. perseae* scab symptoms on young avocado fruit are closely correlated, if environmental conditions are also suitable, because wounds caused by thrips provide infection sites for the fungus (Marroquín-Pimentel 1999; Avila-Quezada et al. 2002; Avila-Quezada et al. 2003)

Several other *Elsinoe* species have a sexual reproductive stage (teleomorph) in which airborne ascospores are produced (Hyde et al. 2013; Fan et al. 2017). However, searches of CAB abstracts and Google using the search terms '*Elsinoe perseae*' and 'asci' or 'ascospores' found no reports of the sexual stage of *E. perseae*. Jenkins (1934) reported that the sexual (perfect) ascospore-producing stage of *Sphaceloma perseae* had not been found, although she had once observed what appeared to be immature *Elsinoe* asci in avocado leaf lesions.

Time to symptom development

In field trials, the time to symptom development is 7–10 days once spores are applied to susceptible avocado tissues in suitable environmental conditions (Stevens and Piper 1941). This has been replicated in many inoculation experiments on susceptible avocado tissues, including leaves, stems and fruit (Stevens and Piper 1941). For example, young unfolding avocado leaves (0.7 to 2 cm in length) were inoculated by securing filter paper or cotton wool soaked with conidia to the leaf with wax paper and a rubber band (Jenkins 1934). The inoculum was removed after 1–2 days. The progression of symptoms was:

- Initial symptoms (minute reddish spots) took approximately 7 days to develop (Jenkins 1934);
- By the tenth day the lesions had increased to a diameter of 0.7 mm and were 'dragon's blood red' on the upper surface and ‘light lobelia violet’ on the lower surface of the leaf (Jenkins 1934);
- Over the following week, the lesions developed a yellowish border and the epidermis blistered and ruptured (Jenkins 1934);
- The circular perforation at the centre of the blistered area later expanded to expose the necrotic leaf tissue underneath (Jenkins 1934).

5.9.4 Likelihood of entry

Elsinoe perseae causes scablike lesions on avocado stems, therefore it can be associated with leafless, rootless avocado budwood cuttings. Ploetz et al. (2011) speculated that *E. perseae* could be moved to new areas in infected scion material (i.e. shoot cuttings such as budwood) but is more easily moved on infected fruit. In addition, ungerminated conidia of *E. perseae* could be present on the surface of the budwood and would be unlikely to be detected by visual inspection without magnification.

The geographic distribution is shown in Table X. *Elsinoe perseae* has been reported in many parts of North America and the Caribbean and the Caribbean, as well as some parts of South America, Africa and Asia.

Given that *E. perseae* can be present on leafless, rootless avocado budwood cuttings and is widely reported in avocado growing countries, it can be associated with the commodity at the time of export. However, if avocado budwood cuttings have an established *E. perseae* infection, it is highly likely that lesions will be detected, and infected material removed or destroyed, during preparation for export, or in phytosanitary inspections pre-export or on arrival in New Zealand.

As part of the commodity description, all imported avocado germplasm, will undergo a growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ. This period in 3A PEQ will impact on likelihood of entry in the following manner:

- Ungerminated *E. perseae* conidia are highly unlikely to remain viable (alive and still capable of infecting the plant) on the surface of avocado budwood during preparation of the material for export, transit to New Zealand, surface disinfection (cuttings dipped in 1% sodium hypochlorite for 2 minutes) upon arrival in PEQ and subsequent grafting of the buds onto new rootstock.
- Furthermore, if viable conidia remain on a bud once it is grafted, infection is likely to occur once the bud breaks dormancy to form young unfolding leaves. In this case, visible lesions are likely to develop within 10 days (see above), and to be detected during routine inspections.
- Searches of Google Scholar, CAB abstracts, CPC and Google using the search terms ‘Elsinoe perseae’, ‘Sphaceloma perseae’ or ‘avocado scab’ found no evidence for asymptomatic *E. perseae* infections or longer latent periods in avocado plants46. Budwood or grafted plants infected with *E. perseae* are therefore almost certain to develop symptoms and be detected and destroyed before they are released from PEQ.

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46 Spanish and Portuguese references were also checked when they were found in searches on the species names.

- Conidia are spread by watersplash, windborne rain and insects (thrips) (Stevens and Piper 1941; Ploetz 2003). Level 3A PEQ has measures in place to manage the risks associated with insects, wastewater, waste and accidental transfer (see section 2.2), therefore viable *E. perseae* propagules are highly unlikely to escape level 3A PEQ to infect a suitable host.

Therefore, if *E. perseae* is imported on avocado, the residual biosecurity risks are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ.

Given the arguments and evidence above, that is:

- established *E. perseae* infections are highly likely to be detected, and infected material removed or destroyed, during preparation for export, or in phytosanitary inspections pre-export or on arrival in New Zealand;
- residual biosecurity risks from recent infections or ungerminated conidia on the surface of avocado budwood are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ;

the likelihood of *E. perseae* entering New Zealand associated with *Persea americana* budwood is considered to be negligible, with low uncertainty.

Therefore, further assessment is considered unnecessary at this time; and likelihoods of exposure and establishment and impacts to New Zealand were not assessed.

5.9.5 Overall level of assessed risk to New Zealand

Based on the assessment of likelihood above, that:

- the likelihood of *E. perseae* entering New Zealand associated with *Persea americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ) is considered to be negligible with low uncertainty;
- therefore, continuing the assessment was considered unnecessary; and

the overall level of risk to New Zealand from *E. perseae* on imported *Persea americana* budwood is considered to be negligible, with low uncertainty.

5.9.6 Management considerations

Elsinoe perseae causes lesions up to 3 mm in diameter on young leaves and stems (Jenkins 1934). New lesions on leaves may initially appear as tiny dark red spots (Jenkins 1934). Lesions are likely to occur on the upper surface of leaves (Jenkins 1934). Older lesions (and therefore lesions on older stems, leaves and fruit) have a brown to black scabby appearance (Jenkins 1934; Stevens and Piper 1941). Symptoms become visible in 7 — 10 days following infection. This suggests that symptoms of pre-existing *E. perseae* infections are highly likely to be detected by visual inspection during preparation for export or on arrival in New Zealand. The number of new lesions decreases at temperatures below 8 °C or above 30 °C (Marroquín-Pimentel 1999), so symptoms may be slower to develop if the budwood is stored and transported at temperatures below 8 °C. Nevertheless, *E. perseae* is highly unlikely to persist as an asymptomatic infection on susceptible tissues.

Ungerminated conidia may remain on older unsusceptible plant parts or dormant buds. If viable conidia have remained on the surface of buds, infections are likely to develop when the plants break dormancy and produce new shoots. Infection occurs at moderate temperatures (between 10 – 26°C) and during periods of relative humidity above 80% (Marroquín-Pimentel 1999) (e.g. periods of rain, overhead irrigation, or heavy morning dew). Therefore, temperatures in PEQ are likely to favour infection, but there may not be enough water on the surface of the plant to allow conidia to germinate and infect susceptible tissues. Nevertheless, conidia are highly unlikely to remain viable on *P.*

americana budwood through pre- and post-export handling and the PEQ period. Therefore, prophylactic testing in the absence of visible lesions is unnecessary.

Everett et al. (2011) developed primers to detect and identify *E. perseae* in PCR testing of DNA extracted from scab lesions on avocado skins, and it is likely that this test can be used for diagnostic testing in budwood.

5.9.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Elsinoe perseae causes lesions up to 3 mm in diameter on young leaves and stems (Jenkins 1934). New lesions on leaves may initially appear as tiny dark red spots (Jenkins 1934). Lesions are likely to occur on the upper surface of leaves (Jenkins 1934). Older lesions (and therefore lesions on older stems, leaves and fruit) have a brown to black scabby appearance (Jenkins 1934; Stevens and Piper 1941).

- ***What are the known environmental conditions conducive to symptom expression?***

Infection occurs during periods of relative humidity above 80% (Marroquín-Pimentel 1999) (e.g. periods of rain, overhead irrigation, or heavy morning dew). Moderate temperatures (between 10 – 26℃) favour infection (Marroquín-Pimentel 1999). Symptoms become visible in 7 — 10 days following infection (Stevens and Piper 1941). The number of new lesions decreases at temperatures below 8 ℃ or above 30 ℃ (Marroquín-Pimentel 1999).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Elsinoe perseae is highly unlikely to persist as an asymptomatic infection on susceptible tissues, although ungerminated conidia may remain on older unsusceptible plant parts or dormant buds. If viable conidia have remained on the surface of buds, infections are likely to develop when the plants break dormancy and produce new shoots. Conidia are unlikely to remain viable on unsusceptible plant parts through the PEQ period so prophylactic testing in the absence of visible lesions is unnecessary

- ***What is the optimum season for conducting detection testing?***

Not applicable.

5.9.8 References

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5.10 *Gliocladiopsis forsbergii, Gliocladiopsis peggii* and *Gliocladiopsis whileyi*

Gliocladiopsis is a genus of soilborne fungi found mostly in tropical and sub-tropical regions of the world. Species of this genus are regarded as secondary plant pathogens or saprobes. *Gliocladiopsis forsbergii, G. peggii* and *G. whileyi* have been isolated frequently from necrotic roots of avocado trees, but pathogenicity studies have so far only been done for one strain of *G. peggii*. Genus level information is being used where necessary as limited information about the ecology of these fungi is available.

5.10.1 Taxonomic description

Scientific name: *Gliocladiopsis forsbergii* L.E. Parkinson, E.K. Dann & R.G. Shivas (2016)

Scientific name: *Gliocladiopsis peggii* L.E. Parkinson, E.K. Dann & R.G. Shivas (2016)

Scientific name: *Gliocladiopsis whileyi* L.E. Parkinson, E.K. Dann & R.G. Shivas (2016)

Order/Family: Hypocreales/Nectriaceae

5.10.2 Hazard identification

Gliocladiopsis forsbergii, G. peggii and *G. whileyi* are not known to be present in New Zealand:

- *Gliocladiopsis forsbergii, G. peggii* and *G. whileyi* are not listed in either Biota NZ (2022) or PPIN (2021), and
- *Gliocladiopsis forsbergii, G. peggii* and *G. whileyi* are not listed in ONZPR (2021) and their regulatory status has not been assessed (ONZPR 2021).

Gliocladiopsis forsbergii, G. peggii and *G. whileyi* have the potential to establish (and spread) in New Zealand because:

- their natural host (*P. americana*) is available for its establishment (NZPCN 2021), and
- other *Gliocladiopsis* species, such as *G. tenuis* and *G. curvata*, have established and are present in New Zealand (NZOR 2021; Biota NZ 2022), and
- suitable climate is available for their establishment (Phillips et al. 2018).

Gliocladiopsis forsbergii, G. peggii and *G. whileyi* have the potential to cause harm to New Zealand because:

- *Gliocladiopsis forsbergii, G. peggii* and *G. whileyi* are likely secondary plant pathogens that could contribute to avocado tree decline, root rot, reduced tree vigour and predisposed trees to infection by other pathogens (Parkinson et al. 2017a).

Gliocladiopsis forsbergii, G. peggii and *G. whileyi* are associated with *P. americana* budwood because:

- *Persea americana* is a known host of *G. forsbergii, G. peggii* and *G. whileyi* (Parkinson et al. 2017a);
- *Gliocladiopsis* species are mostly soilborne but have also been recovered as endophytes from flowers, stems, leaves and seeds (Refaei et al. 2011; Liu and Cai 2013; Gordillo and Decock 2019), roots (as *Glionectria* in Hakizimana et al. (2011)), and from stem lesions (Parkinson et al. 2017a).

Given the arguments and evidence above, *G. forsbergii, G. peggii* and *G. whileyi* fulfil the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.10.3 Risk assessment

Biology and epidemiology of Gliocladiopsis forsbergii*,* Gliocladiopsis peggii *and* Gliocladiopsis whileyi

Host range

Gliocladiopsis species are considered soilborne (Parkinson et al. 2017b) and many isolates have been collected from soil (Lombard and Crous 2012). However, some *Gliocladiopsis* isolates have been collected from diverse symptomatic terrestrial plant material including *P. americana* (in Ecuador, Australia), *Syzygium aromaticum* (Indonesia), *Archontophoenix purpurea* (New Zealand), *Chamaedorea elegans* (USA), *Araucaria* sp. (Malaysia), *Vanilla* sp. (Indonesia), *Indigofera* sp. (Indonesia) (Lombard and Crous 2012), from leaf litter (Lombard and Crous 2012), and decaying wood submerged in forest freshwater in China (Liu and Cai 2013). Although their host range is not well studied, *G. forsbergii, G. peggii* and *G. whileyi* have all been isolated from diseased *P. americana* stems or roots (Lombard and Crous 2012; Parkinson et al. 2017b).

Symptoms and biology

There are limited pathogenicity studies on *Gliocladiopsis* species (Dann et al. 2012; Parkinson et al. 2017b). Parkinson et al. (2017b) fulfilled Koch's postulates, i.e. re-isolation of an isolate of *G. peggii* that had been isolated from necrotic avocado roots. Parkinson et al. (2017b) could not demonstrate pathogenicity at least for the tested isolate of *G. peggii* after 5 weeks. However, *G. forsbergii, G. peggii* and *G. whileyi* have been frequently isolated from necrotic roots of avocado (black root rot) and stem lesions in Australia and Ecuador (Parkinson et al. 2017b) either together with other fungi or alone. This suggests a weak or secondary pathogenic life mode (Dann, pers. comm). So far, only one strain of *G. peggii* has been tested for pathogenicity in a glasshouse study (Dann et al. 2012; Parkinson et al. 2017b), i.e., only for seedlings, in limited environmental conditions and for a limited period of time. However, considering the frequent isolation from symptomatic plant material (Crous and Wingfield 1993; Lombard and Crous 2012; Liu and Cai 2013), this PRA considers the species *G. forsbergii, G. peggii* and *G. whileyi* to be secondary or opportunistic pathogens (Ramírez-Gil and Morales 2019; Solís-García et al. 2021).

Transmission

There is limited information about the transmission and spread of *Gliocladiopsis* species. Crous and Wingfield (1993) consider this group soilborne fungi. Spread is exacerbated by frequent irrigation (and over-irrigation), crowded seedling arrangements and poor nursery hygiene practices (Crous and Wingfield 1993). Desiccation and unfavourable environmental conditions for fungal growth have little effect on the primary survival of spores of nectriaceous fungi, as these highly resistant resting structures can survive for several years, infesting soil and host debris, and will germinate or sporulate when conditions become favourable (Parkinson 2017). Uncertainty of the transmission mode arises from *Gliocladiopsis* species recovered as endophytes from flowers, roots, stems, leaves and seeds (Liu and Cai 2013). This suggests a possible endophytic life mode for at least some species within the genus.

Symptom development and symptom expression

Symptom development of *Gliocladiopsis* species is not well understood. Likely factors influencing symptom expression depend on a variety of biotic and abiotic conditions. Co-infection with other pathogens, such as with Botryosphaeriaceae fungi, is thought to accelerate symptom expression (Parkinson 2017). For example, the co-infection disease pathway tends to begin with initial infection of rootstock cuttings at the graft union with Botryosphaeriaceae fungi. Infection is spread during propagation, followed by root infection with nectriaceous fungi in soil. As a result, the reported nectriaceous pathogens disrupt root function, while botryosphaeriaceeous pathogens attack the xylem (Parkinson 2017). Some studies suggest synergistic effects of *Gliocladiopsis* species with root pathogens such as *Phytophthora* spp (Parkinson 2017; Ramírez-Gil and Morales 2019; Solís-García et al. 2021). Other studies indicate no pathogenicity of *Gliocladiopsis* species but an endophytic life mode such as growth promotion (Parkinson 2017) and antagonistic effects to other fungi (Pecundo et al. 2021).

5.10.4 Likelihood of entry

This assessment is made on the basis that *G. forsbergii, G. peggii* and *G. whileyi* are present in the countries where avocado budwood is being imported from. *Gliocladiopsis forsbergii, G. peggii* and *G. whileyi* have been reported from diseased avocado trees in Australia (Parkinson 2017) and from Ecuador (as *Gliocladiopsis* sp., Liu and Cai (2013)).

The mode of infection is unknown but likely occurs over roots and wounds. The limited amount of information available suggests that *Gliocladiopsis* species are likely soilborne (Lombard and Crous 2012; Parkinson et al. 2017b). However, *Gliocladiopsis* spp have been isolated from stem lesions of avocado (Parkinson et al. 2017a; Parkinson et al. 2017b). *Gliocladiopsis* species have also been isolated as endophytes (Liu and Cai 2013). As a result, it cannot be ruled out that *Gliocladiopsis* species are associated with *P. americana* budwood internally and surface sterilisation (as in the commodity description) would not affect the fungus.

Symptom expression and time to symptom development is not well understood for *Gliocladiopsis* species. In case of symptom expression during PEQ, the most likely observed symptoms would be black root rot or less likely stem lesions (Parkinson 2017). However, as with most secondary pathogens, it seems more likely that *Gliocladiopsis* species would remain asymptomatic or endophytic during PEQ until favourable conditions or co-infection with other pathogens. Therefore, *Gliocladiopsis* species could pass undetected during PEQ without molecular testing.

Given the arguments and evidence above, that is:

- limited information suggests that *Gliocladiopsis* species are likely soilborne (Crous and Wingfield 1993; Lombard and Crous 2012), and
- some *Gliocladiopsis* species have also been isolated as endophytes (Liu and Cai 2013), and
- *Gliocladiopsis* species could remain asymptomatic or endophytic during PEQ (Parkinson 2017)

the likelihood of *G. forsbergii, G. peggii* and *G. whileyi* entering New Zealand associated with *P. americana* budwood is considered to be low, with high uncertainty.

5.10.5 Likelihood of exposure

This assessment is made on the basis that *G. forsbergii, G. peggii* and *G. whileyi* have entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is known to be a suitable host for *G. forsbergii, G. peggii* and *G. whileyi* (Parkinson et al. 2017a).

If avocado plants are infected with *G. forsbergii, G. peggii* and *G. whileyi* at the time of their release from PEQ they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *G. forsbergii, G. peggii* and *G. whileyi* in New Zealand from *P. americana* budwood is considered high, with low uncertainty.

5.10.6 Likelihood of establishment

This assessment is made on the basis that *G. forsbergii, G. peggii* and *G. whileyi* has been successfully exposed to a suitable host plant in the New Zealand environment.

The natural host range of *G. forsbergii, G. peggii* and *G. whileyi* is unknown but they have all been isolated from *P. americana* (Parkinson et al. 2017a). This host in the form of cultivated avocados is available for its establishment in the North Island (see section 2.3). A saprobic life mode of these fungi seems also likely (Lombard and Crous 2012; Liu and Cai 2013).

Some avocado growing regions, such as East Australia (from where the species have been described), have a similar climate to the whole of New Zealand, as well as to the Auckland and Northland regions, as indicated by a climate match index of ≥0.7 (Phillips et al. 2018). Therefore, climate suitable for the establishment of *G. forsbergii, G. peggii* and *G. whileyi* is readily available. In addition, climate suitability is supported by the fact that other *Gliocladiopsis* species have already established in New Zealand (Biota NZ 2022).

Given the arguments and evidence above, that is:

- their natural host (*P. americana*) is available for its establishment (NZPCN 2021), and
- a saprophytic life mode is supported, and
- other *Gliocladiopsis* species, such as *G. tenuis* and *G. curvata*, have established and are present in New Zealand (NZOR 2021; Biota NZ 2022), and
- suitable climate is available for their establishment (Phillips et al. 2018)

the likelihood of *G. forsbergii, G. peggii* and *G. whileyi* establishing in New Zealand is considered high, with low uncertainty.

5.10.7 Impacts in New Zealand

Gliocladiopsis species are considered soilborne and spread is exacerbated by frequent irrigation (and over-irrigation), crowded seedling arrangements and poor nursery hygiene practices (Crous and Wingfield 1993). Unfavourable environmental conditions for fungal growth have little effect on the primary survival of spores of nectriaceous fungi, as these highly resistant resting structures can survive for several years, infesting soil and host debris, and will germinate or sporulate when conditions become favourable (Parkinson 2017). Other *Gliocladiopsis* species have been recovered as endophytes from flowers, roots, stems, leaves and seeds (Liu and Cai 2013). This suggests a possible endophytic life mode for at least some species within the genus.

Economic impacts

There is very little evidence for economic impacts of *G. forsbergii, G. peggii* and *G. whileyi*. Although frequently isolated from necrotic avocado roots in Australia, limited pathogenicity tests are available. It seems likely that *G. forsbergii, G. peggii* and *G. whileyi* are secondary plant pathogens that could contribute to avocado tree decline, root rot, reduced tree vigour and predisposed trees to infection by other pathogens (Parkinson et al. 2017a). However, their economic impact has not been demonstrated as multiple biotic and abiotic factors are likely to be involved in disease expression. The limited evidence suggest that this group of fungi is not a severe pathogen (Dann et al. 2012; Parkinson et al. 2017b), and that existing orchard management practises would manage the impact of these fungi (Dann, pers. comm).

Given the arguments and evidence above, that is:

- *Gliocladiopsis forsbergii, G. peggii* and *G. whileyi* are likely secondary pathogens that should be managed by existing orchard management practises

the economic impact of *G. forsbergii, G. peggii* and *G. whileyi* on New Zealand is considered to be very low, with moderate uncertainty.

Environmental impacts

There are no reports of environmental impacts of *Gliocladiopsis* species (google scholar search terms ["wild plants" AND " *Gliocladiopsis*"] and ["reservoir plants" AND " *Gliocladiopsis*"] and ["*Gliocladiopsis*" AND "natural environments"]). There is one report of *G. forsbergii* isolated from a

Grevillea sp (Parkinson et al. 2017a). Other than this report, *G. forsbergii, G. peggii* and *G. whileyi* have so far only been isolated from avocado (Farr and Rossman 2021).

Given the arguments and evidence above, the impact on the environment from the establishment of *G. forsbergii, G. peggii* and *G. whileyi* in New Zealand is considered to be negligible, with low uncertainty.

Human health impacts

There are no known human health impacts associated with *Gliocladiopsis* species (google scholar search terms ["human health" AND " *Gliocladiopsis*"] and ["toxin" AND " *Gliocladiopsis*"]).

Given the arguments and evidence above, the human health impact of *G. forsbergii, G. peggii* and *G. whileyi* on New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

A response to an outbreak of *Gliocladiopsis* could involve the removal of infected or exposed avocado trees growing on residential properties. This is likely to cause distress to affected residents, however, this impact is likely to be localised and short-lived. Moreover, a response seems unlikely given that severe pathogenicity has never been reported for *Gliocladiopsis*.

Given the arguments and evidence above, the sociocultural impact of *G. forsbergii, G. peggii* and *G. whileyi* on New Zealand is considered to be negligible, with low uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, human health and society is considered to be very low, with moderate uncertainty.

5.10.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is low
- assuming entry, the likelihood of exposure is high
- assuming successful exposure, the likelihood of establishment is high
- the overall impact on the New Zealand economy, environment, human health and society is considered to be very low

the overall level of assessed risk to New Zealand from *G. forsbergii, G. peggii* and *G. whileyi* on imported *P. americana* budwood (as in the commodity description) is considered to be very low, with moderate uncertainty.

5.10.9 Management considerations

The economic impact of *Gliocladiopsis forsbergii, G. peggii* and *G. whileyi* has not been demonstrated as multiple biotic and abiotic factors are likely to be involved in disease expression. The limited evidence suggest that this group of fungi on avocado is not a severe pathogen (Dann et al. 2012; Parkinson et al. 2017b), and that existing orchard management practises would manage the impact of these fungi (Dann, pers. comm).

5.10.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Gliocladiopsis species can remain asymptomatic or endophytic during PEQ (Lombard and Crous 2012). In case of symptom expression black root rot is the most likely symptom to develop (Parkinson 2017). Stem lesions are less likely to develop (Parkinson 2017).

- ***What are the known environmental conditions conducive to symptom expression?***

There is no information available about what environmental conditions favour symptom expression or a pathogenic life stage of *Gliocladiopsis* species (Liu and Cai 2013). However, it has been suggested that co-infection with other pathogens could favour symptom expression (Refaei et al. 2011; Solís-García et al. 2021).

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

There is little information available about asymptomatic infection. However, a strain of *Gliocladiopsis peggii* has been isolated from asymptomatic roots (Dann et al. 2012). Usually, infected root samples or stem lesions are taken for sampling (Parkinson 2017; Parkinson et al. 2017b).

- ***What is the optimum season for conducting detection testing?***

There is no information available about the optimum season for conducting testing.

5.10.11 References

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5.11 *Grovesinia moricola* (zonate leaf spot)

Grovesinia moricola (zonate leaf spot) is a fungal pathogen that produces characteristic target-shaped necrotic rings leading to leaf blight and defoliation in a broad range of host species. The disease has been reported infecting *Persea americana* (avocado) in the USA and Brazil, causing round leaf lesions with dark margins. It has the potential to infect a number of economically significant hosts in New Zealand.

5.11.1 Taxonomic description

Scientific name: *Grovesinia moricola* (I. Hino) Redhead

Order/Family: Helotiales/ Schlerotiniaceae

Other names include: *Grovesinia pyramidialis, Botrytis moricola, Cristulariella moricola, Cristulariella pyramidalis, Hinomyces moricola*

Taxonomic notes:

This fungus was first described under the name *Botrytis moricola* (Narumi-Saito et al. 2006). Previous anamorphic synonyms are *C. moricola* and *H. moricola* and other taxonomic synonyms are *G. pyramidalis* and *C. pyramidalis* (Robert et al. 2005; Johnston et al. 2014).

5.11.2 Hazard identification

Grovesinia moricola is not known to be present in New Zealand:

- *Grovesinia moricola,* or any of the synonyms listed above, are not listed in NZOR (2021), Biota NZ (2022) or PPIN (2021).
- *Grovesinia moricola* is not listed in ONZPR (2021). *Grovesinia pyramidalis* is listed as regulated in ONZPR (2021), and *Cristulariella moricola* is listed as 'not assessed'.

Grovesinia moricola has the potential to establish (and spread) in New Zealand because:

- Many reported hosts of *G. moricola* are present in New Zealand, including avocado, apple, tomato and grapevine.
- The climate is unlikely to limit its development and spread: *G. moricola* has been reported in USA (southeastern states), South Korea, Taiwan, and Japan. These regions have a CMI ranging from 0.6-0.8, indicating a similar climate to many parts of New Zealand (Phillips et al. 2018). The fungus is also reported in regions of India and Brazil which have a CMI ranging from 0.4-0.6. This suggests a relatively broad range of climatic tolerance for this species. Latham (1974) reports that growth is favoured by high humity and moderate to cool temperatures, suggesting the New Zealand climate is likely to be favourable for the establishment of this fungus species

Grovesinia moricola has the potential to cause harm to New Zealand because:

- It is reported to cause leaf lesions and defoliation of a large number of host species, which may reduce the productivity of economically important species such as avocado, apple, tomato and grapevine. Infections in tomatoes were reported to result in a 50% reduction in yield (Dillard et al. 1995). Loss of productivity to this degree would have significant economic impacts.

Grovesinia moricola is associated with *P. americana* budwood because:

- The fungus has been reported infecting the leaves of avocados in Brazil (Bezerra et al. 2008). Because green budwood may include some leaf tissue within the buds, there is the potential for this fungus to be associated with the commodity. Also, while this fungus is predominantly reported as a foliar pathogen, it has been reported infecting the stems of potato (Trolinger et al. 1978) and tomato (Dillard et al. 1995), suggesting that the fungus may infect the stems or leaf buds of avocado budwood.

Given the arguments and evidence above, *Grovesinia moricola* meets the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.11.3 Risk assessment

***Biology and epidemiology of* G. moricola**

Host range

Grovesinia moricola has been reported infecting a wide range of host plants in many different plant families. A survey of literature found that at least 100 species, belonging to 42 plant families, are naturally infected by this fungus (Appendix 3). There were two reports found of infection of avocado with *G. moricola*; one in USA (Trolinger et al. 1978) and one in Brazil (Bezerra et al. 2008). There are three other hosts of *G. moricola* reported in the Lauraceae family; *Cinnamomum kanehirae* (camphor), *Lindera benzoin* (spicebush) and *Sassafras albidum* (sassafras) (Trolinger et al. 1978; Fu and Lin 2012).

Distribution

Grovesinia moricola has been reported from the regions and countries listed in Table 5-13.

Table 5-13: The geographical distribution of Grovesinia moricola.

Region	Country (Location)	Reference	CMI
Asia	South Korea (Gimcheon, Goseong, Chuncheon and Yeongdong)	Cho et al. (2012); Cho et al. (2013); Cho et al. (2017); Shin et al. (2019); Park et al. (2020)	0.6-0.7
	India (Jabalpur and Khargone)	Vyas et al. (1982)	0.4-0.5
	Taiwan (Taipei and Wulai)	Su and Leu (1983); Hu et al. (2002); Fu and Lin (2012)	0.6-0.7
	Japan (Akita Province)	Trolinger et al. (1978); Takahashi and Tanba (1980)	0.7-0.8
North America	USA (Georgia, Louisiana, North Carolina, South Carolina, Alabama, Maryland, Illinois, West Virginia, Arkansas, New York)	Cline et al. (1983); Colyer et al. (1992); Brenneman et al. (1993); Holcomb (1994); Dillard et al. (1995); Grand and Vernia (2004); Blake et al. (2015); Aubrey et al. (2017)	0.7-0.8
South America	Brazil (Bahia)	Bezerra et al. (2008)	0.5-0.6

Symptoms

Symptoms of *G. moricola* are often reported as small round lesions with dark margins that grow larger and eventually coalesce, leading to leaf blight and defoliation. Lesions are frequently described as zonated or target-shaped rings. In avocado the symptoms are described as greyish-brown, round to irregular leaf lesions with dark margins, less than 1cm broad (Bezerra et al. 2008). Symptoms on *Cinnamomum kanehirae* (also in the family Lauraceae) are reported as brown lesions on the leaves that fuse as they become larger, causing leaf blight and eventual defoliation (Fu and Lin 2012). No evidence was found in the literature of asymptomatic infection occurring in any host species.

On many hosts the development of symptoms of *G. moricola* infection develop rapidly (Leahy 1995). Inoculation experiments have frequently reported the development of symptoms within 2 to 4 days (Hu et al. 2002; Cho et al. 2012; Fu and Lin 2012; Cho et al. 2013; Cho et al. 2017; Shin et al. 2019). Holcomb (1994) reported that symptoms developed on *Halesia diptera* (silverbell) within 24 hours of inoculation.

Optimal conditions for growth and symptom development

It is frequently reported that outbreaks of *G. moricola* occur during periods of heavy or extended rainfall (Vyas et al. 1982; Aubrey et al. 2017). Numerous inoculation experiment have successfully demonstrated pathogenicity in a range of host species at incubation temperatures ranging from 15 - 30℃ (Brenneman et al. 1993; Holcomb 1994; Hu et al. 2002; Shin et al. 2019). Latham (1974) investigated the impacts of temperature and moisture on this fungus and found that lessions developed at all humidities tested, but fruiting bodies did not develop below 96% relative humidity. Maximum growth occurred when inoculated leaves are wet and is favoured by moderate to cool temperatures (9-24℃).

Reproduction and transmission

Transmission of spores is thought to occur by splashing of water (Bezerra et al. 2008). This fungus also readily produces sclerotia on artificial medium within two weeks, and on artificially inoculated leaves within four weeks (Shin et al. 2019). These sclerotia are believed to be important for overwintering of the fungus (Trolinger et al. 1978; Cline et al. 1983).

5.11.4 Likelihood of entry

Grovesinia moricola has been reported to be present in six countries (Table 5-13), affecting a wide variety of host plants. It has been reported infecting the leaves of avocados in Brazil (Bezerra et al. 2008), while the symptoms of an infection on avocado in USA are not stated (Trolinger et al. 1978). While this fungus is predominantly reported as a foliar pathogen, it has been reported infecting the stems of potato (Trolinger et al. 1978) and tomato (Dillard et al. 1995), suggesting that the fungus has the potential to be present on plant stems. Also, green budwood may include some leaf tissue within the buds. Therefore, there is potential for *G. moricola* to be imported into PEQ on *P. americana* budwood.

Grovesinia moricola infections appear on the surface of leaves and stems, so it is expected that disinfection of budwood entering PEQ (1% sodium hypochlorite dip for 2 minutes) would be effective in elimating any infection on the surface. Although, it is uncertain if the fungus may persist in small crevices within the leaf buds.

If *G. moricola* is imported into New Zeland, and disinfection were not suffient to remove the pathogen, it is very likely that it would produce visible symptoms in PEQ. There are no results of inoculation experiments available for avocado to determine the period to develop symptoms or the potential to be asymptomatic on this host. However, there are numerous inoculation studies of other host species that demonstrate rapid symptom development and asymptomatic infection has not been reported in any host species. Most inoculation studies show symptom development on leaf material within 2 to 4 days of inoculation (Hu et al. 2002; Cho et al. 2012; Cho et al. 2013; Cho et al. 2017; Shin et al. 2019), including another Lauraceae species, *Cinnamomum kanehirae* (Fu and Lin 2012). This suggests that if *G. moricola* was present on avocado budwood imported to New Zealand, it is highly likely that symptoms would develop within the period of growth in PEQ.

Based on the mode of reproduction and transmission, there is no evidence available to suggest that *G. moricola* would not be successfully contained within Level 3A PEQ conditions.

Given the arguments and evidence above, that is:

- there is limited potential for the association of *G. moricola* with avocado budwood cuttings, and

- infection by *G. moricola* is highly likely to be detected during the period of growth in Level 3A PEQ;

the likelihood of *G. moricola* entering New Zealand associated with *P. americana* budwood is considered to be negligible, with low uncertainty. Therefore, further assessment is considered unnecessary at this time; likelihoods of exposure and establishment and impacts to New Zealand are not further assessed.

5.11.5 Overall level of associated risk to New Zealand

Based on the assessment of likelihood above, that:

- the likelihood of *G. moricola* entering New Zealand associated with *P. americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ) is considered negligible, with low uncertainty;

the overall level of risk to New Zealand from *G. moricola* on imported *P. americana* bedwood is considered to be negligible, with low uncertainty.

5.11.6 Management considerations

Grovesinia moricola is frequently reported producing rapidly developing and highly visible symptoms on the foliage of a wide range of host species. There is only a single source reporting the symptoms of *G. moricola* in avocado. Bezerra et al. (2008) reported the disease causing greyish-brown, round to irregular leaf lesions with dark margins, less than 1cm broad. This appears to be consistent with the symptoms reported in many other host species. There was no evidence found of asymptomatic infections on any host species.

It is frequently reported that outbreaks of *G. moricola* on a variety of hosts occur during periods of heavy or extended rainfall (Vyas et al. 1982; Aubrey et al. 2017). Relative humity greater than 96% is required for the development of fruiting bodies (Latham 1974). Maximum growth occurs when inoculated leaves are wet and is favoured by moderate to cool temperatures (9-24°C). Leahy (1995) reports that optimum conditions for disease development are cool and wet weather during mid to late summer.

Grovesinia moricola appears to be readily isolated in many hosts from leaf samples exhibiting symptoms. The fungus may occur on either side of the leaves (Leahy 1995), and may also be present on plant stems (Trolinger et al. 1978; Dillard et al. 1995). This fungus can be identified based on the distinctive conidial morphology, or sequencing of the internal transcribed spacer (ITS) region of the ribosomal DNA using PCR primers ITS1 and ITS4 (Aubrey et al. 2017; Shin et al. 2019).

5.11.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

In avocado, *G. moricola* has been reported causing greyish-brown, round to irregular leaf lesions with dark margins, less than 1cm broad (Bezerra et al. 2008). These symptoms are consistent with those that have been observed in many other host species.

- ***What are the known environmental conditions conducive to symptom expression?***

It is frequently reported that outbreaks of *G. moricola* on many hosts occur during periods of heavy or extended rainfall (Vyas et al. 1982; Aubrey et al. 2017). Relative humity greater than 96% is required for the development of fruiting bodies, but lessions still develop at lower humidity (Latham 1974). Maximum growth occurs when inoculated leaves are wet and is favoured by moderate to cool temperatures (9-24℃).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Grovesinia moricola appears to be readily isolated from leaf samples exhibiting symptoms. There was no evidence found of asymptomatic infection occurring in any host species. The fungus may occur on either side of the leaves (Leahy 1995).

- ***What is the optimum season for conducting detection testing?***

Leahy (1995) reports that optimum conditions for disease development are cool and wet weather during mid to late summer. Maximum growth occurs when inoculated leaves are wet and is favoured by moderate to cool temperatures (9-24 degrees Celsius) (Latham 1974). Relative humity greater than 96% is required for the development of fruiting bodies.

5.11.8 References

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5.12 *Lasiodiplodia pseudotheobromae*

Lasiodiplodia pseudotheobromae is a fungal plant pathogen that has been reported to cause dieback, cankers and postharvest diseases (stem-end rot and fruit rot disease) in many woody plant species which includes avocado.

5.12.1 Taxonomic description

Scientific name: *Lasiodiplodia pseudotheobromae* A.J.L. Phillips, A. Alves & Crous 2008

Order/Family: Botryosphaeriales/Botryosphaeriaceae

Other names include: None found

Taxonomic notes: *Lasiodiplodia pseudotheobromae* was initially identified as a cryptic species of *L. theobromae* (Alves et al. 2008). This is because they have similar colony features. However, *L. pseudotheobromae* was discovered to be a different species with a larger and a different conidia shape (Munirah et al. 2017).

5.12.2 Hazard identification

Lasiodiplodia pseudotheobromae is not known to be present in New Zealand:

- *L. pseudotheobromae* is not listed in PPIN (2021)
- *L. pseudotheobromae* is recorded as absent from New Zealand in Biota NZ (2022)
- *Lasiodiplodia pseudotheobromae* is not listed in ONZPR (2021).

Lasiodiplodia pseudotheobromae has the potential to establish (and spread) in New Zealand because:

- host plants are grown commercially and in home gardens in most parts of New Zealand (Table 5-14).
- climate is unlikely to be a barrier to its establishment and spread in New Zealand as it reported to occur in different parts of the world (Table 5-14) which have similar climates (CMI ≥ 7) to New Zealand (Phillips et al. 2018).
- It has been associated with a wide range of host plants commonly grown commercially and in home gardens such as, several *Citrus* spp., and *Vitis vinifera* (grapevine) (Table 5-14).
- *Lasiodiplodia pseudotheobromae* can spread by human movement of infected nursery stock and movement of infected fruits.

Lasiodiplodia pseudotheobromae has the potential to cause harm to New Zealand because:

- *Lasiodiplodia pseudotheobromae* causes pre and postharvest diseases such as branch cankers, dieback and fruit rot. Such diseases can affect the marketability of host crops in New Zealand. It is an important commercial crop in New Zealand worth up to $110 million in domestic sales and up to 56.6 million in domestic sales (Plant & Food Research 2020).

Lasiodiplodia pseudotheobromae can be associated with *Persea americana* budwood because:

- *Persea americana* is a known host of *Lasiodiplodia pseudotheobromae* (Bragard et al. 2021; Liang et al. 2021; Rodríguez-Gálvez et al. 2021)
- *Lasiodiplodia pseudotheobromae* infects avocado stems (Onaebi et al. 2020; Liang et al. 2021; Rodríguez-Gálvez et al. 2021).

Given the arguments and evidence above, *L. pseudotheobromae* meets the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.12.3 Risk assessment

Biology and epidemiology of Lasiodiplodia pseudotheobromae

Biology and host range

Lasiodiplodia pseudotheobromae is a pathogenic fungus from the Botryosphaeriaceae family (Alves et al. 2008; Trakunyingcharoen et al. 2015; Serrato-Diaz et al. 2020). Like most fungi from the Botryosphaeriaceae family, it can occur as an endophyte or a latent opportunistic pathogen that causes symptoms when the host plant becomes stressed (Slippers and Wingfield 2007; Burgess et al. 2019; Liang et al. 2021). However, there are few published studies that have tested asymptomatic tissues to determine if members of the genus *Lasiodiplodia* have an asymptomatic phase on plants they infect (Johnson et al. 1992). So far, the few reported studies of asymptomatic infection by *L. pseudotheobromae* have been from asymptomatic native non-crop plant species in Australia, Thailand, China and India (Kapoor and Saxena 2014; Aramsirirujiwet et al. 2016; Burgess et al. 2019; de Silva et al. 2019). It was also recently isolated from dead leaves of *Magnolia* spp. in China, indicating that it can be saprobic (de Silva et al. 2019).

Lasiodiplodia pseudotheobromae infects a broad range of host crop plants over a broad geographical distribution (Table 1). It is a common pathogen of many woody plant species including *Citrus, Coffee, Gmelina, Rosa* and *Acacia* species(Alves et al. 2008). It is associated with diseases such as cankers, dieback, fruit rot and stem-end rot many host plant species in different parts of the world (Table 5-14).

Most *Lasiodiplodia* spp. are pathogens that are common in tropical and subtropical areas. They generally have an optimal growth temperature of about 30℃ (Burgess et al. 2019). However, *L. pseudotheobromae*, unlike other members of the genus, can grow at temperatures as low as 10℃ (Slippers et al. 2004; Liang et al. 2021).

Lasiodiplodia pseudotheobromae has been reported to be one of the most virulent of *Lasiodiplodia* that can cause stem cankers, dieback of branches, stem-end rot and fruit rot in different host plants in different parts of the world (Table 5-14) (Alves et al. 2008; Ismail et al. 2012; Rodríguez-Gálvez et al. 2017; Liang et al. 2021). It is one of the main pathogens that can cause diseases such as blight, dieback disease, stem-end rot and fruit rot in avocado (Trakunyingcharoen et al. 2015; Onaebi et al. 2020; Bragard et al. 2021; Liang et al. 2021; Rodríguez-Gálvez et al. 2021). In severe cases these disease symptoms that lead to death of infected trees (Slippers and Wingfield 2007; Ismail et al. 2012).

Table 5-14: The distribution, hosts and diseases cause by *Lasiodiplodia pseudotheobromae* (Countries in **bold** are where *Lasiodiplodia pseudotheobromae* has been reported on *P. americana* (avocado). (–) in blank cells indicates information was not available)

Continent	Country	State/Region	Host plant	Symptoms /Diseases	CMI*	Reference
Africa	Cameroon	Mbalmayo and Nkoemvone	*Terminala* spp. and *Theobroma cacao* (Cacao)	Dieback (in Cocoa)	0.5	(Boyogueno et al. 2012)
	Nigeria	Enugu	*Persea americana* (Avocado)	Fruit rot	0.4	(Onaebi et al. 2020)
	Zaire (DRC)	unknown	*Coffea* sp.	-	0.5-0.6	(Alves et al. 2008)
	Egypt	-	*Mangifera indica* (Mango)	Cankers and dieback	0.4-0.6	(Ismail et al. 2012)
Asia	Bangladesh	Rajshahi	*Citrus* sp.	Stem-end rot	0.4	(Sultana et al. 2018)
	China	Guangxi, Hubei, Zhejiang, Hunan, Jiangxi, Fujiang, Guangdong	*Citrus* spp.	Fruit rot	0.6-0.7	(Chen et al. 2021)

Continent	Country	State/Region	Host plant	Symptoms /Diseases	CMI*	Reference
	China	Shandong Province	*Malus pumila* (Apple)	Canker and Shoot dieback	0.7	(Xue et al. 2019)
	China	Hainan	*Plukenetia volubilis* (sacha peanut)	Branch cankers	0.5	(Wang and Song 2021)
	China	Hubei, Jiangsu	*Vitis vinifera* (grapevine)	Pedicel and peduncle discolouration	0.7	(Dissanayake et al. 2015)
	China	Shuyang, Jiangsu Province	*Celtis sinensis* (Chinese hackberry)	Stem cankers	0.7	(Liang et al. 2020)
	China	Guangzhou, Guangdong	*Ormosia pinnata*	Dieback	0.5-0.6	(Li et al. 2020)
	China	Unknown	*Pteridium aquilinum* (Eagle fern)	-	0.4-0.8	(Alves et al. 2008; Rodríguez-Gálvez et al. 2021)
	China	Yunnan	*Magnolia* spp.	Asymptomatic/ saprobic	0.6-0.8	(de Silva et al. 2019)
	India	Manipur	*Mangifera indica* (Mango)	Fruit rot	0.5-0.6	(Bui et al. 2018)
	India	Yelandur Karnataka	*Aegle marmelos*	Asymptomatic	0.5	(Kapoor and Saxena 2014)
	Indonesia	Sulawesi	Cocoa	Dieback and stem cankers	0.4-0.6	(Asman et al. 2020)
	Malaysia	Throughout the peninsular	*Mangifera indica* (Mango)	Fruit rot	0.4-0.5	(Munirah et al. 2017)
	Malaysia	Kelantan, Pahang, Perak, Sabah, Sarawak, Selangor	*Sansevieria trifasciata* (snake plant)	Leaf blight	0.4–0.5	(Kee et al. 2019)
	Thailand	Lamphun Province	*Dimocarpus longan* (Logan fruit)	Fruit rot	0.5	(Pipattanapuckdee et al. 2019)
	Thailand	Chiang Mai Province	*Persea americana* (Avocado)	Fruit rot (stem-end rot, mid-section rot), stem cankers	0.5	(Trakunyingcharoen et al. 2015)
	Thailand	Chiang Mai Province	*Hevea brasiliensis* (Para rubber)	Stem cankers	0.5	(Trakunyingcharoen et al. 2015)
	Thailand	Bangkok	*Hottuynia cordata* (Plookao)	Asymptomatic	0.4	(Aramsirirujiwet et al. 2016)
	Thailand	Chiang Rai	*Cynometra malaccensis*	Leaf spot	0.4	(Gomdola et al. 2020)
	Taiwan	Southern cities, Tainan, Chaiyi	*Persea americana* (Avocado)	Branch canker and fruit rot	0.4-0.8	(Liang et al. 2021)
Europe	Netherlands	Unknown	*Rosa* sp.	-	0.9	(Alves et al. 2008)
Europe-Asia	Turkey	Mersin province Cukurova	*Citrus limon* (Lemon)	Postharvest fruit rot	0.5-0.9	(Awan et al. 2016)
Central America	Costa Rica	Unknown	*Gmelina arborea* *Acacia mangium*	-	0.4-0.6	(Alves et al. 2008)

Continent	Country	State/Region	Host plant	Symptoms /Diseases	CMI*	Reference
Middle East (Asia)	Iran	Northern and Southern Province	*Citrus* spp.	Dieback, cankers and fruit rot	0.4-0.8	(Abdollahzadeh et al. 2009)
	Israel	-	*Persea americana* (Avocado)	Branch dieback, branch wilting, mortality of young plants, stem-end rot.	0.5-0.7	(Bragard et al. 2021)
North America	Mexico	Puebla and Veracruz	*Citrus latifolia* (Persian lime)	Gummosis, stem canker, branch dieback	0.5-0.8	(Bautista-Cruz et al. 2019)
	USA	Puerto Rico	*Perseae americana* (avocado)	Branch cankers	0.5-0.6	
	USA	Puerto Rico	*Citrus* sp. (tangerine)	Branch dieback	0.5-0.6	(Serrato-Diaz et al. 2020)
Oceania	Australia	Northern Western Australia	*Mangifera indica* (Mango), Macadamia	Cankers, dieback, and stem-end rot	0.4	(Sakalidis et al. 2011; Burgess et al. 2019; Jeff-Ego and Akinsanmi 2019)
	Australia	Western Australia	*Vitis vinifera*	Cankers and dieback	0.4	(Burgess et al. 2019)
South America	Brazil	Rio Grande do Norte, Paraíba	*Carica papaya* (papaya)	Stem-end rot	0.5-0.6	(Netto et al. 2014)
	Brazil	Minas Gerais, Bahia	*Annona crassiflora* (marolo)	Dieback	0.5-0.7	(Machado et al. 2019)
	Brazil	Minas Gerais, Bahia	*Annona muricata* (soursop)	Dieback	0.5-0.7	(Machado et al. 2019)
	Brazil	Minas Gerais, Bahia	*Annona squamosa* (sugar apple)	Dieback	0.5-0.7	(Machado et al. 2019)
	Brazil	Minas Gerais, Bahia	*Annona atemoya* (atemoya)	Dieback	0.5-0.7	(Machado et al. 2019)
	Brazil	São Francisco, Assú and Siriji Valleys (North-eastern Brazil)	*Vitis vinifera* (Table grapes)	Decline symptoms	0.4-0.6	(Correia et al. 2013)
	Brazil	Minas Gerais and the Espirito Santo	*Jatropha curcas* (Purging nut)	Wilt, leaf fall, root/collar rot	0.5-0.6	(Machado et al. 2014; Bautista-Cruz et al. 2019)
	Brazil	Sao Paulo State	*Diospyros kaki* (Persimmon)	Fruit rot	0.6-0.7	(Júnior et al. 2017)
	Peru	Lambayeque, Lima, Piura, La Libertad regions.	*Persea americana* (Avocado)	Dieback	0.4-0.8	(Rodríguez-Gálvez et al. 2021)
	Peru	Piura	*Mangifera indica* (Mango)	Dieback	0.4-0.7	(Rodríguez-Gálvez et al. 2017)
	Suriname	-	*Citrus auranitacum*	-	0.4-0.5	(Alves et al. 2008)

Continent	Country	State/Region	Host plant	Symptoms /Diseases	CMI*	Reference
	Uruguay	Entire country	*Myrcianthes pungens*	-	0.7-0.9	(Pérez et al. 2010)
	Venezuela	Monagas State	*Acacia mangium*	Trunk cankers	0.5	(Castro-Medina et al. 2014)

*(Phillips et al. 2018)

Reproduction and spread

Healthy plants can be infected through natural openings and wounds such as pruning wounds (Slippers and Wingfield 2007; Rodríguez-Gálvez et al. 2021). The fungi can be transmitted by contaminated tools during grafting and pruning. Infections have been observed to begin at pruning wounds suggesting that it is an important path through which the fungus enters the plant (Liang et al. 2021). Propagation of infected bud wood and seedlings can also be a source of transmission.

Spores on infected symptomatic avocado stems appear as white powder from black lesions (Rodríguez-Gálvez et al. 2021). Spores can be present all year round on infected twigs and in the soil but are mostly spread during the summer months (Liang et al. 2020). They can be spread by wind, rain and insects (Xiaoqin et al. 2001).

Symptoms

Lasiodiplodia pseudotheobromae causes dieback, branch cankers and fruit rot. The species has been identified and isolated from symptomatic avocado plants and fruits (Onaebi et al. 2020; Liang et al. 2021; Rodríguez-Gálvez et al. 2021).

Under field conditions, necrosis was observed on the stems of infected young avocado plants (Liang et al. 2021). Small necrotic spots can be observed around infected openings which can extend into the internal tissues (Rodríguez-Gálvez et al. 2021). This can proceed into dieback symptoms that begins on the branch of the tree (Liang et al. 2021; Rodríguez-Gálvez et al. 2021), and can lead to defoliation of the infected branch. Infection can progress to other branches and the entire tree (Rodríguez-Gálvez et al. 2021).

In pathogenicity tests, avocado stems inoculated with *L. pseudotheobromae* developed black lesions that excreted white powder (spores) (Liang et al. 2021; Rodríguez-Gálvez et al. 2021). Lesions appeared 10 days after inoculation (Rodríguez-Gálvez et al. 2021), and expanded by 1.1 cm over two to four weeks. Tissues underneath the lesion turned brown along the vascular bundle of the stems (Liang et al. 2021). After 10 days of incubation of inoculated avocado stems, necrotic lesion followed by the development of dark spores with white aerial mycelium were observed (Rodríguez-Gálvez et al. 2021).

Lasiodiplodia pseudotheobromae caused black lesions on both wounded and unwounded avocado fruits. External lesions became visible after two days of inoculation and lesions grew larger as time progressed (2-4 weeks). Fruit pulp of infected avocado turned soft, water-soaked, and black underneath the lesions (Liang et al. 2021).

Optimal growth conditions

Optimal temperature for mycelial growth of *L. pseudotheobromae* isolates under laboratory conditions was 30℃ with a range of 10-35℃ (Liang et al. 2021).

5.12.4 Likelihood of entry

Lasiodiplodia pseudotheobromae has been reported to be present in 23 countries affecting a wide variety of host plants, including multiple records of it being isolated from avocado (Table 5-14). It has been isolated multiple times from aerial parts (stems, branches and fruits) of avocado plants indicating that it can be associated with leafless avocado budwood (Trakunyingcharoen et al. 2015; Onaebi et al. 2020; Bragard et al. 2021; Liang et al. 2021; Rodríguez-Gálvez et al. 2021).

Under field conditions, necrosis was observed on stems of young avocado plants infected by *L. pseudotheobromae* (Liang et al. 2021). Avocado budwoods with established *L. pseudotheobromae* infection are likely to exhibit symptoms such as necrotic lesions and discolouration of internal tissues. As such, infected budwoods could be detected and destroyed during preparation for export, on phytosanitary inspection pre-export (if this occurs), or on arrival in New Zealand.

Species from the family Botryosphaeriaceae can occur as latent opportunistic pathogens that do not cause disease symptoms (asymptomatic) on host plants. They can eventually become pathogenic and express symptoms when the host plant is stressed. Very few published studies have tested asymptomatic tissues to determine if *Lasiodiplodia* spp. might have an asymptomatic phase. One study found *L. theobromae* to occur endophytically in the stem tissue of mango trees (Johnson et al. 1992). There is no evidence that *L. pseudotheobromae* occurs as a latent and asymptomatic phase on cultivated crop plants such as avocado. However, like other endophytic latent pathogens in its family, it is possible that it can occur on asymptomatic leafless, rootless avocado budwood that might not be the detected during preparation for export or on arrival in New Zealand.

Studies that have carried out pathogenicity tests found that symptoms were expressed within 10 days (Liang et al. 2021). Avocado stems infected with *L. pseudotheobromae* were observed to excrete white powder and form black lesions at the inoculation site during pathogenicity tests (Liang et al. 2021; Rodríguez-Gálvez et al. 2021). The active growing period at level 3A PEQ (a minimum of three distinct phenological growing events) will likely result in expression of symptoms.

Given the arguments and evidence above, that is:

- *Lasiodiplodia pseudotheobromae* has been reported to be present in 23 countries.
- *Lasiodiplodia pseudotheobromae* has been isolated from avocado plants (stems, branches and fruits) and can be associated with leafless avocado budwood.
- avocado budwood with established *L. pseudotheobromae* infection can exhibit symptoms such as necrotic lesions and discolouration of internal tissues that would be detected during preparation for export or on arrival in New Zealand.
- *Lasiodiplodia pseudotheobromae* can potentially be present on asymptomatic leafless, rootless avocado budwood and might not be the detected during preparation for export or on arrival in New Zealand, with high uncertainty.
- if *L. pseudotheobromae* is imported on leafless avocado budwood, the active growing period at level 3A PEQ (a minimum of three distinct phenological growing events) will likely result in expression of symptoms.

the likelihood of *L. pseudotheobromae* entering New Zealand associated with *Persea americana* budwood is considered to be ***low***, with ***moderate*** uncertainty.

5.12.5 Likelihood of exposure

This assessment is made on the basis that *L. pseudotheobromae* has entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *L. pseudotheobromae*.

If avocado plants are infected with *L. pseudotheobromae* at the time of their release from post entry quarantine, they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *Lasiodiplodia pseudotheobromae* in New Zealand from *Persea americana* budwood is considered **high**, with **moderate** uncertainty.

5.12.6 Likelihood of establishment

This assessment is made on the basis that *Lasiodiplodia pseudotheobromae* has been successfully exposed to a suitable host plant in the New Zealand environment.

Lasiodiplodia pseudotheobromae is recorded to be present in 23 countries covering five continents (Table 5-14). Countries where it has been recorded have CMI ranges of between 0.4 - 0.9. A CMI of ≥ 0.7, indicates a similar climate to the whole of New Zealand (Phillips et al. 2018). Although it is common in tropical areas with warmer temperatures (Burgess et al. 2019), *L. pseudotheobromae* is the only species in the genus that is observed to grow at temperatures as low as 10°C, with an optimum growth temperature of 30°C, in laboratory conditions (Slippers et al. 2004; Liang et al. 2021). This suggests that the climatic conditions in some parts of New Zealand are likely to be suitable for it to establish, especially the North Island where climatic conditions are warmer. Therefore, climate is not likely to be a barrier for the establishment of *L. pseudotheobromae* in New Zealand, especially the north of the North Island.

Lasiodiplodia pseudotheobromae is known to infect a wide range of host plants (Table 5-14) including some economic important crop plants such as *Vitis vinifera* (grapevine), *Citrus* spp. and *P. americana* (avocado) (Dissanayake et al. 2015; Bragard et al. 2021; Chen et al. 2021; Liang et al. 2021). These plants are grown commercially in different parts of New Zealand. For example, avocado plants are widely grown commercially in orchards and in-home gardens mostly in the North Island and some parts of the South Island (Chapter 2). *Vitis vinifera* (grapes) is grown commercially in parts of the North and South Island and *Citrus* spp. are grown both commercially and in home gardens all around New Zealand. Therefore, suitable hosts are available for *L. pseudotheobromae* to establish and spread in all parts of New Zealand.

Lasiodiplodia pseudotheobromae can spread by the movement of infected nursery stock for planting and the use of contaminated tools used for grafting and pruning, and soil. Spores can be present all year round on infected twigs and in the soil, but are mostly spread during the summer months (Liang et al. 2020). These can be a source of infection and can be spread by wind, rain (Liang et al. 2020). Spread of the pathogen can also be enhanced by insects. For example, the insect pest *Sahlbergella singularis* (Mirid bug) was observed to feed on cocoa pods resulting in wounds which served as access points for infection by *Lasiodiplodia* spp. (Anikwe and Otuonye 2015). There are about six introduced bugs from the same family (Miridae) in New Zealand, three of which are pests feeding on buds, flowers, developing fruits and young shoots of economic significant crops in New Zealand (Eyles 1999). It is possible that mirid bugs present in New Zealand can play are role in enhancing the spread of opportunistic pathogenic fungus such as *L. pseudotheobromae*.

Given the arguments and evidence above, that is:

- *Lasiodiplodia pseudotheobromae* has been reported to be present in countries with a similar climate to New Zealand (parts of Australia, China, Netherland, and Uruguay). The climatic conditions in some parts of New Zealand are likely to be suitable for it to occur and spread, especially the North Island where climatic conditions are warmer
- it is known to affect a wide range of host plants (Table 5-14), some of which are widely grown commercially in orchards and in home gardens in parts of both the North and South Island of New Zealand.
- spores of *L. pseudotheobromae* can be present all year round on infected twigs and in the soil and can be spread through the human movement of infected nursery stock, fruits, contaminated soil and pruning material.

the likelihood of *Lasiodiplodia pseudotheobromae* establishing in New Zealand is considered to be **high**, with **moderate** uncertainty.

5.12.7 Impacts in New Zealand

Lasiodiplodia pseudotheobromae have been reported to be associated with avocado and a wide range of host plants, including economic important crops such as grapevine, citrus and persimmons (Table 1). It can cause branch cankers, dieback symptoms and postharvest rot diseases (Onaebi et al. 2020; Liang et al. 2021; Rodríguez-Gálvez et al. 2021). The fungus can be transmitted by the use of contaminated tools during grafting and pruning (Liang et al. 2021). Propagation of infected bud wood and seedlings can also be a source of transmission. Spores of *L. pseudotheobromae* on infected symptomatic avocado plants can be spread by wind, rain and insects (Xiaoqin et al. 2001).

Economic impacts

Species from the genus *Lasiodiplodia* are aggressive pathogens, affecting a wide range of woody plant species, that lead to the decrease in orchard productivity of economically important crops such as avocado (Valencia et al. 2019). In severe cases they can cause disease symptoms that lead to death of infected trees (Slippers and Wingfield 2007; Ismail et al. 2012), resulting in significant economic loss.

Lasiodiplodia pseudotheobromea is an aggressive pathogen of avocado and thus, it can potentially cause severe diseases and lead to economic loss in the avocado industry (Rodríguez-Gálvez et al. 2021). Avocado is increasingly becoming a significant horticultural crop in New Zealand. At the end of the 2020 financial year, 39,078 tonnes of avocado produced in New Zealand and earning up to NZ$56 million and NZ$110 million in domestic and international sales respectively (Plant & Food Research 2020). Therefore, infections by *L. pseudotheobromea* can potentially result in yield loss which will have an impact on both the domestic and international market.

Infections by *L. pseudotheobromea* could affect market access of exports of avocado to other parts of the world where *L. pseudotheobromea* is not reported to be present. Its presence in New Zealand could potentially result in trade barriers to countries where it is has not been reported to be present.

Lasiodiplodia pseudotheobromea has been reported to cause disease in other economically important host plants causing symptoms such as post-harvest rot, dieback, and tree/branch cankers. As such economic impact caused by *L. pseudotheobromea* is not limited to avocado. It can cause up to 10% yield loss in some crop plants (Awan et al. 2016; Júnior et al. 2017), as well as lead to reduced productivity and longevity of the plants. For example:

- it has been reported as one of the most virulent pathogens responsible for causing necrotic lesions and gum exudes of Persian limes in Mexico, post-harvest rot of Lemon in Turkey, tree cankers in orchards of *Citrus reticulata* and stem/trunk cankers, stem-end rot and fruit rot of *Citrus* spp. in Pakistan (Awan et al. 2016; Bautista-Cruz et al. 2019; Ahmed et al. 2020). These disease symptoms can result in reduced yields and affect the marketability of infected crops. *Citrus* spp. are crops of economic value in New Zealand both domestically and internationally. From pathogenicity tests, *L. pseudotheobromea* was observed to cause damage in 40-50% of fruit surface of lemons after five days (Awan et al. 2016). In 2019, limes, lemons and mandarins had a domestic value of NZ$41 million, with lemons and mandarins having export values of NZ$8 million (Plant & Food Research 2020).
- it has been associated with dieback symptoms and pedicel/peduncle discolouration in grapes in Brazil and China respectively (Dissanayake et al. 2015; Correia et al. 2016). Such symptoms are increasingly significant in reducing longevity of grapevines leading to reduced productivity. Reduction in crop yield and losses can potentially have a significant economic consequence for New Zealand because wine is the second most valuable horticultural exports of New Zealand account to for up to NZ $1.9 billion (Plant & Food Research 2020).
- it has been found to cause shoot and branch dieback and cankers of apple trees in China (Xue et al. 2019). This can potentially lead to reduced productivity and reduced yield of infected trees. Apples are one of the most significant horticulture export crops for New Zealand worth up to NZ$876 million in export in 2020 (Plant & Food Research 2020).
- it was found to cause post-harvest rot of persimmons in Brazil resulting in an estimated 10% loss in crop yield. Persimmons are grown in New Zealand with a value of NZ$1.3 million and

NZ$10.7 million in domestic and export market respectively in 2020 (Plant & Food Research 2020).

While it is known that severe outbreaks of *L. pseudotheobromea* can result in significant yield loss of infected orchards, it might also require the complete removal of severely infected productive trees (avocado, citrus, persimmons, and grapevine) known to require years six to eight years to reach full production. Outbreaks can be triggered by environmental stresses such as drought, hail or frost and trigger outbreaks (Udayanga et al. 2011; Liang et al. 2021). This is likely to occur on a localized scale and causing significant economic on a localized scale.

Given the arguments and evidence above, that is:

- *L. pseudotheobromea* has been reported to be an aggressive pathogen of avocado that can potentially result in reduced productivity and yield loss. This can have an impact on both the domestic and international market.
- infections by *L. pseudotheobromea* can affect market access of exports of avocado to other parts of the world where *L. pseudotheobromea* is not reported to be present
- because *L.* pseudotheobromea affects other economic important hosts plants, it can lead to reduced productivity and longevity of the plants such as grapevine, apples and persimmons resulting in additional economic loss.
- There is moderate uncertainty that infection outbreaks are likely to be sporadic and on a localized scale.

the economic impact of *Lasiodiplodia pseudotheobromea* to New Zealand is considered to be moderate-high, with moderate uncertainty.

Environmental impacts

There is little evidence of *L. pseudotheobromea* impacting on non-crop plant species. It has mostly been reported to cause diseases in cultivated crop plant species (Table 5-14). Although, in a recent study in Australia, a closely related species *L. theobromea* was reported to cause disease in some Australian native plant species (*Adansonia* spp.) (Burgess et al. 2019). *Lasiodiplodia theobromea* is a related species that is already in New Zealand and it has so far not been reported to infect native species in New Zealand. In the same study, *L. pseudotheobromea* was isolated from asymptomatic tissues of *Adansonia gregorii* (Burgess et al. 2019). It has also been isolated from an asymptomatic native perennial herb *Houttuynia cordata* in Thailand (Aramsirirujiwet et al. 2016) and from a deciduous shrub *Aegle marmelos* in India (Kapoor and Saxena 2014). This indicates that it might occur as an endophyte on non-crop species and probably be a source of infection for crop plants that might be grown in close proximity (Burgess et al. 2019). For example Boyogueno et al. (2012) isolated *L. pseudotheobromea* from asymptomatic bark and branches of native species of *Terminalia* spp. growing in close proximity to symptomatic *Theobroma cacao* plants in Cameroon. Therefore, *L. pseudotheobromea* is likely to have a low impact on New Zealand's environment.

Given the arguments and evidence above, the impact on the environment from the establishment of *Lasiodiplodia pseudotheobromea* in New Zealand is considered to be low, with low uncertainty.

Human health impacts

Google and google scholar searches using the search term '*Lasiodiplodia pseudotheobromea* on health' found only one study where *L. pseudotheobromea* was isolated from an eye infection (keratitis) of a mango farmer after a traumatic eye injury that is thought to be the result of inoculation of the fungus from a mango tree branch when the injury occurred (Vanam et al. 2019). *Lasiodiplodia pseudotheobromea* is reported to cause rot in mangoes and can be present on mango tree branches in a latent endophytic infection (Ismail et al. 2012; Rodríguez-Gálvez et al. 2017). Reports of human infection by *Lasiodiplodia* spp. has involved closely related species *L. theobromea* (Maslen et al. 1996; Summerbell et al. 2004) which is known to be present in New Zealand. There has been no report so far that it has affected human health in New Zealand.

Given the arguments and evidence above, the health impact of *L. pseudotheobromea* to New Zealand is considered to be very low, with low-moderate uncertainty.

Sociocultural impacts

Diseases caused by *L. pseudotheobromea* can potentially lead to reduced productivity and render fruits of avocado and other host plants inedible. This can cause scarcity in local markets. Avocado has become popular and is widely consumed in New Zealand. It is commonly grown in home gardens in some parts of the country. Other host plants such as *Citrus* spp. are commonly grown in home gardens around New Zealand.

Given the arguments and evidence above, the sociocultural impact of *L. pseudotheobromea* to New Zealand is considered to be moderate, with low uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, health and society is considered to be moderate-high, with moderate uncertainty.

5.12.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is low with moderate uncertainty
- assuming entry, the likelihood of exposure high with moderate uncertainty
- assuming successful exposure, the likelihood of establishment is high with moderate uncertainty
- the overall impact on the New Zealand economy, environment, health and society is considered to be moderate-high with moderate uncertainty

the overall level of assessed risk to New Zealand from *L. pseudotheobromea* on imported *P. americana* budwood (as in the commodity description) is considered to be moderate-high, with moderate uncertainty.

5.12.9 Management considerations

Under field conditions, *L. pseudotheobromea* causes necrosis on the stems of infected young plants (Liang et al. 2021). Small necrotic spots were observed around infected openings extending into the internal tissues (Rodríguez-Gálvez et al. 2021). Over time, this can proceed into dieback symptoms that begins on the branch of the tree (Liang et al. 2021; Rodríguez-Gálvez et al. 2021), and can lead to defoliation of the infected branch. Infection can progress to other branches and the entire tree (Rodríguez-Gálvez et al. 2021).

Chemical fungicides and biocontrol agents (other microorganisms) have been used in controlling *Lasiodiplodia* species (Khanzada et al. 2005; Kotze et al. 2011). Fungicides were demonstrated to be effective for managing disease symptoms caused by *L. pseudotheobromea* in cocoa growing areas in Indonesia (Asman and Rosmana 2021).

In general, pathogenic endophytes such as *L. pseudotheobromae* can exhibit symptoms under environmental stresses such as drought, hail or frost (Udayanga et al. 2011; Liang et al. 2021), but the specific conditions for symptom expressions for *L. pseudotheobromae* are not well understood and have not been described.

PCR test can be used to detect the presence of *L. pseudotheobromae* (Alves et al. 2008). Samples can be taken from any part of the avocado leafless budwood and testing different parts of the budwood might be required. However, it is not clear if the pathogen will be evenly distributed throughout an infected avocado budwood and that the right bit will be tested.

There is no specified optimum season for conducting testing for *L. pseudotheobromae*

5.12.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Under field conditions, necrosis was observed on the stems of infected young avocado plants (Liang et al. 2021). When plants get infected, small necrotic spots can be observed around infected openings which can extend into the internal tissues (Rodríguez-Gálvez et al. 2021). This can proceed into dieback symptoms that begins on the branch of the tree (Liang et al. 2021; Rodríguez-Gálvez et al. 2021)., and can lead to defoliation of the infected branch. Infection can progress to other branches and the entire tree (Rodríguez-Gálvez et al. 2021).

- ***What are the known environmental conditions conducive to symptom expression?***

In general, pathogenic endophytes such as *L. pseudotheobromae* can exhibit symptoms under environmental stresses such as drought, hail or frost (Udayanga et al. 2011; Liang et al. 2021), but the specific conditions for symptom expression for *L. pseudotheobromae* are not well understood and have not been described.

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Samples can be taken from any part of the budwood and testing different parts of the budwood might be required. However, it is not clear if it will be evenly distributed throughout an infected avocado budwood and that the right part will be tested.

- ***What is the optimum season for conducting detection testing?***

There is no specified optimum season for conducting testing for *L. pseudotheobromae*.

5.12.11 References

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5.13 *Mycosphaerella perseae*

Mycosphaerella perseae is a fungal plant pathogen that causes silver spot disease in *Persea americana* (avocado). Silver spot disease causes lesions on the leaves of the plant.

5.13.1 Taxonomic description

Scientific name: *Mycosphaerella perseae* Miles 1917

Order/Family: Mycosphaerellales / Mycosphaerellaceae (Robert et al. 2005)

Other names include: *Sphaerella perseae* (Miles) Trotter 1928

Taxonomic notes: A *Septoria* sp. is considered the anamorph and *M. perseae* is the teleomorph. *Mycosphaerella perseae* is the preferred name (Robert et al. 2005).

5.13.2 Hazard identification

Mycosphaerella perseae is not known to be present in New Zealand:

- *Mycosphaerella perseae* is not listed in NZOR (2021), Biota NZ (2022) or PPIN (2021).
- *Mycosphaerella perseae* is not listed in ONZPR (2021).

Mycosphaerella perseae has the potential to establish (and spread) in New Zealand because:

- *Persea americana* (avocado) is a known host (Zentmyer 1961; Ploetz 2003) that is widely grown in New Zealand, in home gardens and commercial production, mainly in the north of the North Island.
- *Mycosphaerella perseae* causes leaf spot disease in tropical and subtropical countries including Mexico and Central America (Ploetz 2003; Ploetz 2007). Some regions in these areas have a climate match index (CMI) ≥ 0.7 indicating a similar climate to the whole of New Zealand (Phillips et al. 2018).

Mycosphaerella perseae has the potential to cause harm to New Zealand because:

- it causes leaf spot on avocado (Ploetz 2003; Sermeno et al. 2005). When leaves are heavily infected, necrotic lesions may encompass the entire leaf (Ploetz 2003). Defoliation may occur, but this tends to be restricted to older leaves (Ploetz 2003).
- avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$50.6 million (Plant & Food Research 2020).

Mycosphaerella perseae is associated with *Persea americana* budwood because:

- *Persea americana* is a known host of *M. perseae* (Seaver and Chardon 1926; Ploetz 2003; Sermeno et al. 2005).
- *Mycosphaerella perseae* causes leaf spots and seems to strictly parasitize leaves; however, in favourable climates and severe cases, the fungus has been recorded to cause defoliation and invade the stem (Sermeno et al. 2005).

Given the arguments and evidence above, *Mycosphaerella perseae* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.13.3 Risk assessment

Biology and epidemiology of Mycosphaerella perseae

Hosts and geographical distribution

Mycosphaerella perseae is the causative agent of silver spot disease in *Persea americana* (avocado) (Ploetz 2003). Silver spot is considered a minor problem in avocado orchards and there has been no record of this disease reducing yields (Ploetz 2003; Sermeno et al. 2005).

Mycosphaerella perseae has a distinct host range and has only been recorded on avocado (Seaver and Chardon 1926; Spaulding 1961; Ploetz 2003). It has been recorded on avocado from Puerto Rico (Seaver and Chardon 1926; Spaulding 1961), Mexico (Spaulding 1961; Ploetz 2003) and Florida, USA (Spaulding 1961).

Symptoms

Mycosphaerella perseae causes light brown to silver spots or lesions on the upper surface of leaves and darker brown spots on the lower surface of leaves (Ploetz 2003; Sermeno et al. 2005). A dark brown to black border often surrounds the spots and leaf spots are slightly depressed on the surface of leaves (Ploetz 2003). Leaf spots can vary in size but can grow to a diameter of 1-2 cm (Sermeno et al. 2005). The disease is usually first observed in the lower foliage and progresses up the tree (Sermeno et al. 2005).

Necrotic lesions spread across the surface of the leaves in heavily infected cases. Defoliation may occur, but tends to be restricted to older leaves (Ploetz 2003). In severe cases, *M. perseae* may invade the stem (Sermeno et al. 2005).

Reproduction and transmission

Mycosphaerella perseae produces ascospores (i.e., sexual spores of Ascomycota). Ascospores produced in infected and dead leaves can infect young leaves through wind, rain and insects (Ploetz 2003). The anamorph (i.e. asexual stage) of *M. perseae*, which was previously known as a *Septoria* sp., produces pycnidia (i.e. asexual fruiting bodies). Conidia (i.e. asexual spores) are released from pycnidia and secondary spread can occur via conidia from older leaves or dead twigs (Sermeno et al. 2005). In both sexual and asexual transmission, rain, fog and humid conditions favour disease development, especially during the spring season (Ploetz 2003; Sermeno et al. 2005). *Mycosphaerella perseae* requires sufficient moisture to produce infection and grows optimally at temperatures between 10-27°C (Sermeno et al. 2005).

In-field management

Pruning of trees to reduce humidity is one type of management that controls the disease in avocado production. In addition, removing infected fallen leaves (i.e. source of fungal inoculum) from the orchard is another form of disease management (Ploetz 2003). Three applications of maneb, captan or copper fungicides at 30-day intervals have been successful in treating symptoms in orchards (Ploetz 2003).

5.13.4 Likelihood of entry

Mycosphaerella perseae causes leaf spots on avocado leaves but in severe cases it has been reported to infect stems (Sermeno et al. 2005); therefore, the fungus could be associated with leafless, rootless avocado budwood cuttings. However, there is only limited evidence to support this association, based on a single technical document from El Salvador, with no primary records to verify this. However, *M. perseae* is more likely to produce ascospores and conidia on older leaves or dying and dead material (Ploetz 2003; Sermeno et al. 2005) and thus removing leaves and importing clean rootless budwood would considerably reduce the likelihood of entry on this pathway. Conidia and ascospores of other *Mycosphaerella* species (e.g. *M. ligulicola* and *M. musicola*) do not seem to survive for more than a few weeks (Blakeman and Hornby 1966; Thammaiah et al. 2007).

If avocado budwood cuttings have an established *M. perseae* infection, it is highly likely that lesions will be detected, and infected material removed or destroyed, during phytosanitary inspections on arrival in New Zealand.

As part of the commodity description, all imported avocado germplasm will undergo a growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ. This period in 3A PEQ will impact on likelihood of entry in the following manner:

- any ungerminated *M. perseae* conidia on the surface of avocado budwood are highly unlikely to remain viable (alive and still capable of infecting the plant) on the assumption of survivability of conidia of *M. ligulicola* (Blakeman and Hornby 1966) during preparation of the material for export, transit to New Zealand, surface disinfection (cuttings dipped in 1% sodium hypochlorite for 2 minutes) upon arrival in PEQ and subsequent grafting of the buds onto new rootstock.
- if viable conidia remain on a bud once the bud is grafted, infection is likely to occur once the bud breaks dormancy to form young unfolding leaves. In this case, visible lesions are likely to develop assuming the following conditions occurs, temperatures between 10–27℃ and high humidity, and thus, be detected during routine inspections.
- Searches of Google Scholar, CAB abstracts, CPC and Google using the search terms ‘*Mycosphaerella perseae*’, ‘*Sphaerella perseae*’ and ‘asymptomatic’ found no evidence for asymptomatic *M. perseae* infections or longer latent periods in avocado plants47. Budwood or grafted plants infected with *M. perseae* are therefore almost certain to develop symptoms and be detected and destroyed before they are released from PEQ.
- Conidia are spread by watersplash, windborne rain and insects (Ploetz 2003). Level 3A PEQ has measures in place to manage the risks associated with insects, wastewater, waste and accidental transfer (see section 2.2), therefore viable *M. perseae* propagules are highly unlikely to escape level 3A PEQ to infect a suitable host.

Therefore, if *M. perseae* is imported on avocado, the residual biosecurity risks are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ.

Given the arguments and evidence above, that is:

- there is very limited evidence for a potential association of *M. perseae* with leafless, rootless avocado budwood cuttings;
- established *M. perseae* infections are highly likely to be detected, and infected material removed or destroyed, during preparation for export, in phytosanitary inspections pre-export or on arrival in New Zealand;
- residual biosecurity risks from recent infections or ungerminated conidia on the surface of avocado budwood are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ;

the likelihood of *Mycosphaerella perseae* entering New Zealand associated with *Persea americana* budwood is considered to be negligible, with low uncertainty.

Therefore, further assessment is considered unnecessary at this time; and likelihoods of exposure and establishment and impacts to New Zealand were not assessed.

5.13.5 Overall level of assessed risk to New Zealand

Based on the assessment of likelihood above, that:

- the likelihood of *M. perseae* entering New Zealand associated with *Persea americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ) is considered to be negligible with low uncertainty;

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[47] Spanish and Portuguese references were also checked when they were found in searches on the species names.

- therefore, continuing the assessment was considered unnecessary; and

the overall level of risk to New Zealand from *M. perseae* on imported *Persea americana* budwood is considered to be negligible, with low uncertainty.

5.13.6 Management considerations

Mycosphaerella perseae produces visible lesions on leaves (see above) and searches on the species name found no evidence for asymptomatic infection of leaves or stem tissues. Therefore, *M. perseae* is highly unlikely to persist as an asymptomatic infection on susceptible tissues.

There is no information on time to development of symptoms, but high humidity and moisture conditions, at temperatures between 10-27°C are known to induce symptoms (Ploetz 2003; Sermeno et al. 2005). Ascospores and conidia are more likely to sporulate on older leaves or dying and dead material (Ploetz 2003; Sermeno et al. 2005) and this suggests that symptoms of pre-existing *M. perseae* infections are highly likely to be detected by visual inspection during preparation for export or on arrival in New Zealand.

If viable conidia have remained on the surface of buds, infections are likely to develop when the plants break dormancy and produce new shoots. Given the conditions described above, temperatures in PEQ are likely to favour infection, but there may not be enough water on the surface of the plant to allow conidia to germinate and infect susceptible tissues. Nevertheless, conidia are highly unlikely to remain viable on *P. americana* budwood through pre- and post-export handling and the PEQ period. Therefore, prophylactic testing in the absence of visible lesions is unnecessary.

Mycospharella spp. specific primers for nested polymerase chain reaction (PCR) have been developed, however, this was developed for *Eucalyptus* leaves (Glen et al. 2007). Other studies have demonstrated *Mycospharella* specific primers in other hosts (Guo et al. 2006) suggesting PCR techniques could be developed for diagnostic testing of *M. perseae* in avocado if symptoms are detected in PEQ.

5.13.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Mycosphaerella perseae causes lesions on leaves which can measure up to 2cm in diameter (Sermeno et al. 2005). Lesions can coalesce into larger lesions as infection spreads (Ploetz 2003). Lesions are light brown to silver on the upper surface of leaves and darker brown spots on the lower surface of leaves (Ploetz 2003; Sermeno et al. 2005). A dark brown to black border often surrounds the spots and leaf spots are slightly depressed on the surface of leaves (Ploetz 2003).

- ***What are the known environmental conditions conducive to symptom expression?***

Optimal growth occurs in high humidity and moisture conditions at temperatures between 10-27°C (Sermeno et al. 2005). The use of overhead irrigation to simulate rainfall could induce symptom expression of *M. perseae*, however, it would increase the risk of the disease spreading among plants.

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

There is no information that *M. perseae* causes asymptomatic infections and thus, there is no information on which plant part should be tested to maximise the detection of *M. perseae* in the case of asymptomatic infection.

- ***What is the optimum season for conducting detection testing?***

Rain, fog and humid conditions in the spring favour silver spot disease development (Ploetz 2003).

5.13.8 References

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5.14 *Neocosmospora perseae*

Neocosmospora perseae is a fungal pathogen reported to cause avocado trunk cankers and is probably an opportunistic pathogen infecting the plant via wounds.

5.14.1 Taxonomic description

Scientific name: *Neocosmospora perseae* Sand.-Den. & Guarnaccia (2018)

Order/Family: Hypocreales/Nectriaceae

Other names include: *Fusarium perseae* (Sand.-Den. & Guarnaccia) O'Donnell, Geiser & T. Aoki (2020)

Taxonomic notes: *Neocosmospora perseae* is a recently described member of the *Fusarium solani* species complex (FSSC). Members of this complex tend to be very similar morphologically. For this reason, members of the FSSC were historically referred to as mating strains or *formae speciales* (groups of races and biotypes of a pathogen species that can only infect plants within a certain host genus or species) of the species *Fusarium solani*. However, recent studies estimate that the *F. solani* complex includes at least 60 phylogenetically distinct species (Aoki et al. 2014). Even recently, the fungus causing a disease outbreak has often been classified as *Fusarium solani* or a member of the FSSC because of the difficulty of identifying FSSC members to species level based on morphological features. In the absence of specific studies on *N. perseae,* this assessment relies on historic information about the epidemiology of *F. solani sensu lato* (that is, used in its historic sense to encompass the whole species complex) and studies referring to *F. solani* that probably relate to different species within the complex. *Neocosmospora perseae* is in clade 3 of the FSSC, along with species such as *N. falciformis, N. keratoplastica, N. petroliphila* and *N. solani sensu stricto* which are ubiquitous in soil and decaying plants.

There is debate in the literature as to whether species in the FSSC should remain part of *Fusarium* or should be placed in the genus *Neocosmospora* (Aoki et al. 2019; Sandoval-Denis et al. 2019; Lynn et al. 2020; O'Donnell et al. 2020; Geiser et al. 2021; Lynn et al. 2021). Discussion of fungal phylogenetics (taxonomy based on molecular sequences and biological characteristics) is beyond the scope of this PRA. A name change is highly unlikely to affect the conclusions of the PRA (unless it were to synonomise an ambrosia Fusarium complex (AFC) species with a species that is present in New Zealand). Although Species Fungorum gives the current legitimate name as *Fusarium perseae*, EPPO (2021), Farr and Rossman (2021) and Mycobank (Robert et al. 2005) favour *Neocosmospora perseae*. In addition, although there is no record of *N. perseae* in Biota NZ (2022) , this database records *Neocosmospora* as a valid genus including *N. solani* replacing *F. solani sensu stricto*. To align with this, the species name *Neocosmospora perseae* has been used in this PRA.

5.14.2 Hazard identification

Neocosmospora perseae is not known to be present in New Zealand:

- Neither *N. perseae* nor its synonym *F. perseae* are listed in either Biota NZ (2022) or PPIN (2021). While there is no record of *N. perseae* in New Zealand (PPIN 2021; Biota NZ 2022), *Fusarium solani* (*sensu lato* i.e., the species complex) is present in New Zealand (Biota NZ 2022) and recently described species within the complex such as *N. perseae* could previously have been identified as *F. solani*. The Manaaki Whenua – Landcare Research collection records a single isolate of *N. solani* (ICMP 10680, identified as *F. solani*, and in clade 3, the same clade as *N. perseae*) from *P. americana* in Gisborne. However, DNA sequence of this New Zealand isolate (MG857354) is different from *N. perseae* (Ho, pers. comm.).
- *Neocosmospora perseae* is not listed in ONZPR (2021).

Neocosmospora perseae has the potential to establish (and spread) in New Zealand because:

- it is recorded from areas with a Climate Match Index (CMI) ≥0.7 indicating a similar climate to the whole of New Zealand (Phillips et al. 2018);
- avocado is a host and is commonly grown in northern parts of New Zealand, both commercially and domestically;
- transmission of *N. perseae* is likely to be similar to other species in the FSSC (that is *N. solani* in the historic broad sense). This group are soilborne fungi that opportunistically infect susceptible hosts, often through wounds, or lesions caused by other fungi (Smith 2007; Marek et al. 2013). Conidia (asexual spores) of *N. solani* are produced on plant debris on the soil surface and dispersed by water splash (Meyers and Cook 1972). Some FSSC species also produce airborne ascospores (Smith 2007). Therefore, *Neocosmospora perseae* can also be transmitted by contaminated soil and tools.

Neocosmospora perseae has the potential to cause harm to New Zealand because:

- it causes trunk cankers on mature avocado trees, reducing their productivity (Guarnaccia et al. 2018);
- avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$100 million and domestic sales earned NZ$50.5 million (Plant & Food Research 2019).

Neocosmospora perseae is associated with *Persea americana* budwood because:

- avocado (*P. americana*) is a known host of *N. perseae*;
- *Neocosmospora perseae* was isolated from trunk cankers and caused cankers on shoots of avocado seedlings that were at a similar developmental stage to the plant material used for commercial budwood.

Given the arguments and evidence above, *N. perseae* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.14.3 Risk assessment

Biology and epidemiology of Neocosmospora perseae

Host range and distribution

Neocosmospora perseae was first described as a pathogen of avocado in Italy (Guarnaccia et al. 2018) and more recently in Greece (Guarnaccia et al. 2021). It has also been reported from trees in São Paulo, Brazil (Contato et al. 2021), but in this study, the host species and symptoms were not reported (Table 5-15).

Although *formae speciales* of *F. solani* (current name *N. solani*) have historically been characterised according to host, recent phylogenetic studies have shown that some have diverse host ranges (Sandoval-Denis et al. 2019). Therefore, given that *N. perseae* is only recently described, it cannot be assumed that it is specific to avocado. Furthermore, there are many historic records of *Fusarium solani* in avocado (see below), although only one relating to trunk cankers was found in searches.

Table 5-15. Geographical distribution of *Neocosmospora perseae*

Region	Country	Locality, State/Province (if recorded)	CMI*	Host plant	Reference
Europe	Italy	San Leonardello, Catania	**0.8**	*P. americana*	Guarnaccia et al. (2018)
	Greece	Chania, Crete	**0.7**	*P. americana*	Guarnaccia et al. (2021)
South America	Brazil	Ribeirão Preto, São Paulo	0.6	Tree (species not recorded)	Contato et al. (2021)

* Climate Match Index (CMI) with all of New Zealand (Phillips et al. 2018).

Searches of Google scholar, CAB abstracts, CPC and Google using the search terms "*Neocosmospora perseae*" and "*Fusarium perseae*" (with quote marks to restrict search results to these species names) found only three primary reports (Table 5-15) and a few other records, mainly taxonomic studies, referring to Guarnaccia et al. (2018) or including *N. perseae* sequences in molecular phylogenies.

Symptoms

Neocosmospora perseae was isolated from trunk cankers on mature (14-year-old) avocado plants (Guarnaccia et al. 2018). Infected bark appeared cracked, darkly discoloured and/or slightly sunken. Reddish-brown discolouration within the canker extended into the xylem (Guarnaccia et al. 2018). Occasionally, a sugar exudate was present on the surface (Guarnaccia et al. 2018). Approximately 10% of plants showed disease symptoms (Guarnaccia et al. 2018). *Fusarium*-like fungi were the only isolates obtained from symptomatic avocado trees and all five of the characterised strains were considered to be the newly described species *N. perseae* (Guarnaccia et al. 2018).

Transmission

Guarnaccia et al. (2018) considered that transmission of *N. perseae* is likely to rely on soil contamination and plant-associated reservoirs. General information about the life cycle of members of *Fusarium solani sensu lato* (i.e. the FSSC) is therefore relevant. *Fusarium solani* is a ubiquitous soil fungus (Coleman 2016; Aoki et al. 2019; Sandoval-Denis et al. 2019; Lynn et al. 2020; Geiser et al. 2021; Lynn et al. 2021). These reviews and taxonomic treatments, and the results from Google Scholar and CABI searches on the term "*Fusarium solani*" and "*Neocosmospora solani*", indicate that the FSSC is extremely widely reported as a cause of foot and root rots, diebacks, seedling blights, branch and trunk cankers and stem and fruit rots, as well as an endophyte and saprophyte in a diverse range of hosts and environments. *Fusarium solani* opportunistically infects susceptible hosts, often through wounds, or lesions caused by other fungi (Marek et al. 2013).

Conidia (asexual spores) of *F. solani* may be produced in lesions on stems and roots or plant debris on the soil surface and are dispersed by water splash (Meyers and Cook 1972; Roy 1997). When nutrients are depleted, germinated *F. solani* conidia may form chlamydospores (resting spores) which can survive in the soil for long periods (Meyers and Cook 1972), usually in, or on the surface of plant residues (Smith 2007). Chlamydospores germinate if there is a source of organic nitrogen and sugar (Meyers and Cook 1972; Smith 2007), for example nutrients from roots or tubers of a nearby host plant. Germinating chlamydospores contain enough energy for the developing hyphae to grow a short distance but if they do not reach a suitable host, they lyse and die (Smith 2007). If the developing hyphae reach a suitable host they may colonise the cortex (the layer of cells between the epidermis and the vascular tissues of stems and roots) (Smith 2007). Some FSSC species can colonise the plant systemically following root infection. For example, *F. solani* was reisolated from stem tissues following root inoculation in cannabis plants (Punja et al. 2021), soybeans (Gai et al. 2012) and mandarin seedlings (Kurt et al. 2020).

Some members of the FSSC produce airborne ascospores (sexual spores), often in wounds or cankers on woody branches. These species can be homothallic (a single isolate can produce ascospores, without the need for an isolate of another mating type) or heterothallic (a second mating type is required for fertilisation).

It is likely that *N. perseae* can be transmitted by grafting, as this is reported as a means of transmission in other FSSC species. For example, *F. solani* disease lesions on the trunks of almond and apple nursery plants were localised around or above graft unions implying that these fungi may have gained access to trees during grafting (Marek et al. 2013).

Some species in the FSSC are reported to be spread by insects, for example the ambrosia fusarium clade (AFC) species are farmed by ambrosia beetles (see PRA on Fusarium dieback caused by AFC species (section 5.2)). However Guarnaccia et al. (2018) found no evidence that *N. perseae* was associated with any vector, as there were no beetle galleries or other sign of insect infestation in trees infected with *N. perseae*.

Growth conditions and time to symptom development

In natural infections, canker-causing pathogens such as *N. perseae* may gain entry to the plant due to frost damage or mechanical injuries such as pruning wounds (Guarnaccia et al. 2018). Cankers caused by *N. perseae* have been observed in branches of 5-year-old avocado plants and trunks of 14-year-old avocado plants. However, the only information about symptom development in *N. perseae* comes from pathogenicity testing in 6-month-old (Guarnaccia et al. 2018) or two-year-old *P. americana* seedlings (Guarnaccia et al. 2021). Stems of these plants are likely to be similar in age to stems of budwood cuttings and shoots of newly grafted plants.

Symptoms of *N. perseae* developed one month after inoculation in young avocado seedlings grown at 25 ± 1 ℃ (Guarnaccia et al. 2018; Guarnaccia et al. 2021) and 95 % relative humidity under a 12-h fluorescent light/dark regime (Guarnaccia et al. 2018). After three months, all the inoculated avocado seedlings showed disease symptoms, with serious symptoms leading to plant death (and *N. perseae* could be reisolated from these diseased seedlings) (Guarnaccia et al. 2018).

In culture (on agar plates), the temperature range for growth of *N. perseae* was 9 –36 ℃, with an optimum of 27–30 ℃ (Guarnaccia et al. 2018).

Potential for latent infections in stems

There are only two reports of *N. perseae* from avocado plants (Guarnaccia et al. 2018; Guarnaccia et al. 2021). Since those plants showed symptoms, there is no specific evidence that *N. perseae* can be latent in plant stems for long periods without showing symptoms. However, there is evidence in the literature that *Neocosmospora* species can be endophytes in plant hosts (Sandoval-Denis et al. 2019) or can switch between an endophytic and pathogenic lifecycle in response to environmental stresses. For example:

- a strain of *N. solani* isolated from the roots of a wetland plant (*Phragmites australis*) was a weak mutualist promoting root growth at medium salinity in rice, *Oryza sativa*, roots and beneficial to survival at moderate salinity but was associated with a negative effect on root length at medium high salinity (Eydoux and Farrer 2020).
- *Fusarium solani* was among the species isolated from canker lesions in dormant cold stored apple and almond (*Prunus dulcis*) seedlings (Marek et al. 2013). Symptoms included white, pink, and orange sporulation on the surface of the bark, emerging through lenticels (pores in the bark), directly over or adjacent to internal necrosis of the inner bark and cambium. These symptoms developed in the seedlings after they were left outside on pallets for approximately 7 to 14 days and became desiccated. Marek et al. (2013) considered it likely that *Fusarium* spp. were endophytically associated with the apple and almond scions (grafted budwood) as latent or asymptomatic infections.
- *Fusarium solani* was isolated as an endophyte from healthy Persian oak (*Quercus brantii*) trees (as well as from declining trees) and did not produce symptoms within three months in pathogenicity testing (Alidadi et al. 2019).

Other reports of Neocosmospora solani (i.e. FSSC species, reported as Fusarium solani) in Persea americana

Avocado has historically been reported as a host of *N. solani*. Since *N. perseae* is likely to have been reported historically as *F. solani,* these records may provide insight into its biology and epidemiology.

Neocosmospora solani (along with *Fusarium* and *Phytophthora* spp.) was isolated from trunk cankers on *P. americana* in Michoacán Mexico (Torres et al. 2000). Canker symptoms showed as cracks in the bark and as dark discoloured wood with white powder on the surface (Torres et al. 2000). Irrigated, heavily shaded groves favoured disease development. When a conidial solution was sprayed onto small wounds at the base of stems of avocado seedlings (2 cm in diameter), *N. solani* caused a faint necrosis with white granular powder on the exterior of the stem. Small red lesions under the bark did not extend deeper into the wood, suggesting that *N. solani* was a secondary parasite in this instance.

Neocosmospora solani has been reported from post-harvest stem end rot lesions of avocado fruit in Cameroon (Djeugap et al. 2015), Kenya (Wanjiku et al. 2020; Wanjiku et al. 2021) and South Africa (Darvas and Kotze 1987; Darvas et al. 1987; Korsten et al. 1995).

In Michoacán, Mexico, *N. solani* was isolated from root rots of avocado plants with stunted leaves, leaf yellowing, interrupted growth during vegetative flushes, and premature abscission of leaves (Olalde-Lira et al. 2020). In 4-month-old avocado plants, symptoms of *N. solani* took 51–56 days to develop when a conidial suspension of each isolate was added near the roots, without damaging them.

The Manaaki Whenua – Landcare Research database has many records of *N. solani* (as *F. solani*) in New Zealand, but only one is from avocado and there is no record of the plant part the fungus was collected from. This isolate was collected from *P. americana* in Gisborne in 1977 and is in Clade 3, the same clade of the FSSC as *N. perseae*) but has been confirmed as being different from *N. perseae* based on a DNA sequence analysis (https://scd.landcareresearch.co.nz/specimen/ICMP_10680; Ho, pers. comm.).

5.14.4 Likelihood of entry

Neocosmospora perseae has only been recorded from branches and trunks of avocado plants (5 or 15 years old) in naturally occurring infections with visible canker symptoms. Although *N. perseae* caused disease symptoms within one month in pathogenicity testing on young avocado seedlings (i.e. in stems of a similar developmental stage to avocado budwood), another species in the FSSC (reported as *F. solani*) has been reported in budwood of other woody hosts, possibly with a latent period before external symptoms developed (see above). Based on current evidence, there is a low likelihood that *N. perseae* can be present in or on *P. americana* budwood, and that it can escape detection during preparation for export, or in phytosanitary inspections prior to export or on arrival in New Zealand. Surface disinfection (cuttings dipped in 1% sodium hypochlorite for 2 minutes) upon arrival in PEQ is likely to kill any ungerminated spores on the surface of the cutting but is highly unlikely to eliminate an existing infection. There is moderate uncertainty about the likelihood of *N. perseae* being associated with avocado budwood, since this species is only recently described and there is no specific evidence for natural infection of very young stem tissues. At the time of writing, no published studies have investigated the possibility of asymptomatic *N. perseae* infections but other *Neocosmospora* species can be endophytes or latent pathogens in plant hosts (see section 0).

Assuming that *N. perseae* can be present in/on the commodity as described (leafless, rootless avocado budwood cuttings), and given that it is reported in at least three avocado growing countries (Italy, Greece and Brazil), it can be associated with the commodity at the time of export. If so, it is likely it can be transmitted by grafting to plants for planting generated from budwood. However, infected avocado budwood with externally visible cankers or lesions of *N. perseae* will almost certainly be removed or destroyed during preparation for export, or in phytosanitary inspections pre-export or on arrival in New Zealand. Therefore, only material with latent asymptomatic infections (or with mild internal symptoms that were not noticed at the time of grafting) is likely to enter PEQ.

As part of the commodity description all imported avocado germplasm will undergo a growing period of active growth with a minimum of three distinct phenological growing events, referred to as ‘shoot flush’ in 3A PEQ. This period in 3A PEQ will impact on likelihood of entry in the following manner:

In young (6-month-old) avocado seedlings inoculated with *N. perseae* and maintained at approximately 25 °C and 95 % relative humidity, canker and internal discolouration symptoms developed within one month (Guarnaccia et al. 2018). These environmental conditions may not be met during the PEQ period. In addition, pathogenicity tests involved inserting a mycelium plug (a colonised agar plug) into a wound formed by cutting between two nodes (Guarnaccia et al. 2018). In nature, FSSC species are often reported as infecting via wounds or disease lesions, so introduction of inoculum by wounding the plant mimics a natural infection route. However, a mycelium plug introduces much higher levels of inoculum than graft transmission or water-splash infection of wounds with *N. perseae* spores, and the agar provides an initial energy source for the pathogen (Bhunjun et al. 2021). Therefore, although symptoms of *N. perseae* developed rapidly in pathogenicity tests, natural conditions with less inoculum may not favour rapid symptom development.

Neocosmospora solani (historically reported as *F. solani*) infections are sometimes detected in nursery stock of woody species during routine inspections in Post-Entry Quarantine (MPI interceptions records, accessed 2021), suggesting that symptomatic *N. perseae* infections will usually be detected. For example, *N. solani* (not identified to *forma specialis* or variety level) was isolated from localised lesions at the nodes of cold-stored almond scions, and Marek et al. (2013) considered that these lesions developed either from spores trapped on the buds, or from endophytic inoculum present in the scion. Even when there were no external symptoms, internal necrosis caused by *N. solani* in grafted almond nursery stock could still be observed on stems with light-brown or green bark (Marek et al. 2013). However internal lesions of *N. solani* were difficult to detect in grafted almond nursery stock when the bark was dark brown, resulting in infected trees being supplied to growers. Therefore, it is likely that any symptoms caused by *N. perseae* will be detected in PEQ, but latent infections or internal symptoms may be missed.

Given the arguments and evidence above, that is:

- there is a documented association of *N. perseae* with trunks and branches of *P. americana;*
- the association with *P. americana* budwood (i.e. young stem tissues) is moderately uncertain because it is based on pathogenicity tests and information about related fungal species;
- estimates of time to symptom development are based on pathogenicity tests and suggest symptoms of *N. perseae* will usually develop and be detected prior to export or within the PEQ period. However, the method used for pathogenicity testing is likely to favour rapid infection and symptom development;
- there is evidence that FSSC species similar to *N. perseae* sometimes remain latent for long periods and may only develop symptoms when the host plant is stressed;
- internal necrosis caused by *N. perseae* may not be detected in routine inspections of plants in PEQ;

the likelihood of *N. perseae* entering New Zealand associated with *Persea americana* budwood is considered to be very low with moderate uncertainty.

5.14.5 Likelihood of exposure

This assessment is made on the basis that *N. perseae* has entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *N. perseae.*

If avocado plants are infected with *N. perseae* at the time of their release from Post Entry Quarantine they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *N. perseae* in New Zealand from *Persea americana* budwood is considered high, with low uncertainty.

5.14.6 Likelihood of establishment

This assessment is made on the basis that *N. perseae* has been successfully exposed to a suitable host plant in the New Zealand environment.

The reported distribution and host range of *N. perseae* is shown in Table 5-15, and includes San Leonardello, Catania, Italy (Guarnaccia et al. 2018) and, areas with a CMI of 0.7 indicating that they have a similar climate to the whole of New Zealand (Phillips et al. 2018). Currently the only reported host is avocado (*P. americana*) which is grown commercially and in home gardens, particularly in northern regions of New Zealand (see section 2.3). Therefore, the climate in New

Zealand is unlikely to limit the establishment of *N. perseae* and hosts are likely to be available, at least in avocado growing regions.

On culture medium, *N. perseae* produced both conidia (asexual spores) and chlamydospores (resting spores that develop from conidia and facilitate survival/overwintering in the soil) (Guarnaccia et al. 2018). Guarnaccia et al. (2018) therefore considered it likely that *N. perseae* is transmitted to new hosts via soil contamination and plant-associated reservoirs. Conidia (asexual spores) of FSSC species such as *N. perseae* are typically produced on plant debris on the soil surface and dispersed by soil water or water splash to infect roots or above ground tissues of suitable hosts. Propagules of *N. perseae* could also be carried to new areas within New Zealand by movement of infected plant material or contaminated soil, and possibly by wind or windborne rain. Therefore, it is likely that *N. perseae* would spread easily within and between areas where avocado is grown.

Given the arguments and evidence above, the likelihood of *N. perseae* establishing in New Zealand is considered high, with low uncertainty.

5.14.7 Impacts in New Zealand

Neocosmospora persea is a recently described species of the *Fusarium solani* species complex (FSSC) that causes trunk cankers in avocado (Guarnaccia et al. 2018). Members of this complex, specifically *Fusarium solani*, are ubiquitous soil fungi (Coleman 2016; Aoki et al. 2019; Sandoval-Denis et al. 2019; Lynn et al. 2020; Geiser et al. 2021; Lynn et al. 2021) and thus *N. perseae* is most likely spread through soil contamination and plant-associated reservoirs (Guarnaccia et al. 2018). There are likely multiple ways this fungus can be spread: *N. perseae* can be transmitted by grafting, as this is reported as a means of transmission in other FSSC species (e.g. Marek et al. 2013). Canker-causing pathogens such as *N. perseae* may gain entry to the plant due to frost damage or mechanical injuries such as pruning wounds (Guarnaccia et al. 2018).

Symptoms of *N. perseae* may take years before they are observed (described below) and thus *N. perseae* may have the ability to switch between an endophytic and pathogenic lifecycle in response to environmental stresses (based on evidence that *Neocosmospora* species can be endophytes in plant hosts (Sandoval-Denis et al. 2019)).

Economic impacts

Neocosmospora perseae is only recently described and its known impacts are limited to two disease outbreaks, one causing trunk cankers in 10% of 14-years-old avocado trees in an orchard in Italy and another causing branch cankers in 40% of 5-year-old avocado trees (cv. 'Hass') in a 2-ha orchard (2 ha) in Greece. Based on this information, economic impacts of *N. perseae* have been assessed only for avocado.

Although historical outbreaks of *N. perseae* were probably attributed to *N. solani* (i.e. *F. solani*), searches found only one other report of *N. solani sensu lato* from trunk cankers on avocado, which appeared to be an opportunistic secondary infection (Torres et al. 2000). This suggests that severe trunk canker symptoms from *N. perseae* in avocado commercial production are likely to be rare and localised to a few orchards or, at worst, one growing region. Alternatively, the causal agent of trunk cankers may not always be identified. Since newly planted trees take six to eight years to reach full production (https://industry.nzavocado.co.nz/grow/becoming-an-avocado-grower/, accessed 19/10/2021), if rogueing (removal of infected or damaged plants) was necessary, production losses from an outbreak could persist for several years. If there was a widespread and severe *N. perseae* outbreak in productive avocado plants in either the Bay of Plenty or Northland, it could cause moderate economic impacts, but this is an unlikely scenario. However, given that reports of *N. solani* and more recently *N. perseae* causing impacts in avocado are sporadic, it is more likely that impacts will be low, with sporadic outbreaks damaging avocado trees over a smaller geographical area, impacting a small number of growers.

Many FSSC species are pathogenic to a wide range of plant, insect and marine animal hosts (Zhang et al. 2006; O'Donnell et al. 2016; Sandoval-Denis and Crous 2018) and although *formae speciales* of *N. solani* (reported as *F. solani*) were historically characterised and named according to plant hosts, recent phylogenetic studies have shown that some have diverse host ranges (Sandoval-Denis et al. 2019). This means that there is moderate uncertainty that economic impacts of *N. perseae* will be limited to avocado. Economic impacts of *N. perseae* may need to be reassessed in future if disease outbreaks in other economically important hosts are reported.

Given the arguments and evidence above, that is:

- *Neocosmospora perseae* outbreaks are likely to be rare and sporadic but can cause unwanted impacts on avocado production in New Zealand by damaging some trees in some orchards or growing regions.
- since *N. perseae* is only recently described:
    - there is moderate uncertainty that economic impacts of *N. perseae* on the avocado industry would be low in all circumstances; and
    - there is moderate uncertainty that impacts of *N. perseae* in New Zealand would be limited to avocado, given that closely related species have diverse host ranges;

the economic impact of *N. perseae* to New Zealand is considered to be low with moderate uncertainty.

Environmental impacts

Neocosmospora perseae is only recently described and searches (see above) found no specific evidence of hosts other than avocado plants; therefore, there is no evidence to suggest that it can have impacts in the New Zealand environment. However, there is moderate uncertainty since closely related FSSC species in the same clade, such as *N. falciformis, N. keratoplastica, N. petroliphila* and *N. solani sensu stricto* are pathogenic to a range of plant, insect and marine animal hosts (Zhang et al. 2006; O'Donnell et al. 2016; Sandoval-Denis and Crous 2018). Environmental impacts of *N. perseae* may need to be reassessed in future if there are new reports that suggest that its host range is similarly broad.

Given the arguments and evidence above, the impact on the environment from the establishment of *N. perseae* in New Zealand is considered to be very low, with moderate uncertainty.

Health impacts

Neocosmospora perseae is only recently described and searches (see section 0) found no specific evidence that it causes human disease or produces mycotoxins that can harm human health. However, *N. perseae* is in the FSSC, and species in this complex cause approximately two thirds of fusarium infections in humans and other animals (Zhang et al. 2006; Chehri et al. 2015). Clade 3 FSSC species such as *N. falciformis, N. keratoplastica, N. petroliphila* and *N. solani sensu stricto* are ubiquitous in soil and decaying plants and this clade is most often reported as human pathogens, causing skin and eye infections as well as more severe internal infections in immune compromised patients (Zhang et al. 2006; Mehl and Epstein 2007; Chehri et al. 2015). *Neocosmospora perseae* is also in clade 3 (Sandoval-Denis et al. 2019) and clusters near *N. petroliphila* in a molecular phylogeny (Guarnaccia et al. 2018). Although *N. perseae* has not been recorded as a human pathogen, it is likely that it can cause opportunistic infections in humans, particularly if they are immunocompromised. However, given that such opportunistic infections are rare, and the known human FSSC pathogens *N. petroliphila* and *N. solani s.s.* are already present in New Zealand (Biota NZ 2022), the additional health impact of *N. perseae,* if any, is considered to be very low.

Members of the FSSC are not usually considered to produce significant levels of mycotoxin in crops (Munkvold et al. 2021). However, *N. solani* strains have been reported to produce several different mycotoxins in culture (Shi et al. 2016; Hoque et al. 2018; Rabaaoui et al. 2021). However, even if *N. perseae* produces mycotoxins in avocados, the resulting health impacts for avocado consumers are likely to be very low. There is moderate uncertainty in this conclusion, due to the lack of information about effects of mycotoxins produced by FSSC species.

Given the arguments and evidence above, the health impact of *N. perseae* to New Zealand is considered to be very low, with moderate uncertainty.

Sociocultural impacts

Neocosmospora perseae is only recently described and searches (see above) found no specific evidence of hosts other than avocado plants. Avocado is widely consumed in New Zealand. In some parts of the country avocados are common in home gardens, and trunk cankers caused by *N. perseae* may lead to decreased productivity or death in some domestic trees. *Neocosmospora perseae* may lead to small reductions in commercial avocado fruit production but is unlikely to cause scarcity for New Zealand consumers.

Given the arguments and evidence above, the sociocultural impact of *N. perseae* to New Zealand is considered to be very low, with moderate uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, health and society is considered to be low, with moderate uncertainty.

5.14.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is very low
- assuming entry, the likelihood of exposure is high
- assuming successful exposure, the likelihood of establishment is high
- the overall impact on the New Zealand economy, environment, health and society is considered to be low

the overall level of assessed risk to New Zealand from *N. perseae* on imported *P. americana* budwood (as in the commodity description) is considered to be low, with moderate uncertainty.

5.14.9 Management considerations

Neocosmospora perseae causes cankers (areas of dark or discoloured necrotic tissue) on stems and trunks, possibly with bleeding and a white powdery fungal growth. In pathogenicity tests, symptoms developed within one month, and it is therefore likely that visible symptoms of *N. perseae* will develop early in the PEQ period. If necrosis only affects the internal tissues, they are still likely to be detected by visual inspection on green stems, but may be missed on stems with darker bark.

There is high uncertainty about whether latent asymptomatic *N. perseae* infections occur in avocado budwood, although some *Neocosmospora* species have been reported as symptomless endophytes or can be latent for long periods in other hosts (see biology and epidemiology section 5.14.3).

Characterisation of *N. perseae* was based on EF-1α, ITS, LSU, and RPB2 sequences (Guarnaccia et al. 2018) and this may be useful for diagnostic testing (if needed).

5.14.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Neocosmospora perseae causes cankers (areas of dark or discoloured necrotic tissue) on stems and trunks, possibly with bleeding and white powdery fungal growth. If necrosis only affects the internal tissues, they are still likely to be detected on green stems but may be missed on stems with darker bark.

- ***What are the known environmental conditions conducive to symptom expression?***

Other related *Neocosmospora* species have been reported to switch from an endophytic to pathogenic lifestyle when plants are stressed, for example, by drought. *Neocosmospora perseae* is only recently described and searches (see below) found no specific information about environmental conditions favouring symptom expression in natural infection.

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Assuming that *N. perseae* can infect avocado budwood asymptomatically, it would presumably be systemic in stem tissues or, if it accessed the budwood at the time of collection, it would be concentrated at or near the cut end(s) of the budwood.

- ***What is the optimum season for conducting detection testing?***

No relevant information was found. However, since the pathogen causes stem cankers, the pathogen should be easier to detect during October and November after foliage is shed (Chapter 2).

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5.15 *Neofusicoccum nonquaesitum*

Neofusicoccum nonquaesitum is a pathogenic fungi from the family Botryoshaeriaceae. It can cause diseases such as stem/branch cankers, necrosis and dieback in several woody crop plant species (grapevine, avocado, and apples) and non-crop plant species.

5.15.1 Taxonomic description

Scientific name: *Neofusicoccum nonquaesitum* Inderb., Trouillas, R.M. Bostock & Michailides (2010)

Order/Family: Botryosphaeriales/ Botryosphaeriaceae

5.15.2 Hazard identification

Neofusicoccum nonquaesitum is not known to be present in New Zealand:

- *Neofusicoccum nonquaesitum* is not listed in either Biota NZ (2022), PPIN (2021) or NZOR (2021)
- *Neofusicoccum nonquaesitum* is not recorded in ONZPR (2021)

Neofusicoccum nonquaesitum has the potential to establish (and spread) in New Zealand because:

- host plants (avocado) are grown commercially and in home gardens in some parts of New Zealand. *Neofusicoccum nonquaesitum* is associated with hosts plants such as avocado, blueberry, grapevine and apples that are grown commercially and in home gardens in different parts of New Zealand
- Climate is unlikely to be a barrier to their establishment and spread in New Zealand because they are reported to occur in places (Table 5-16), which have a similar climate (CMI) ≥ 0.7 to New Zealand (Phillips et al. 2018)
- *Neofusicoccum nonquaesitum* can be spread by human movement of infected nursery stock to avocado growing areas.

Neofusicoccum nonquaesitum has the potential to cause harm to New Zealand because:

- *Neofusicoccum nonquaesitum* can cause cankerous lesions, necrosis and dieback symptoms on trunks of avocado trees (Carrillo et al. 2016; Guarnaccia et al. 2020).
- Diseases caused by *N. nonquaesitum* can reduce the productibility and yield of avocado trees in New Zealand as well as other economic host plants such as blueberry, grapevine and apples.

Neofusicoccum nonquaesitum is associated with *Persea americana* budwood because:

- *Persea americana* are a known host of *N. nonquaesitum* (Carrillo et al. 2016; Guarnaccia et al. 2020);
- *Neofusicoccum nonquaesitum* has been associated with avocado and have been reported to cause stem/branch cankers and dieback disease (Carrillo et al. 2016; Guarnaccia et al. 2020).

Given the arguments and evidence above, *N. nonquaesitum* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.15.3 Risk assessment

Biology and epidemiology of Neofusicoccum nonquaesitum

General Biology

Neofusicoccum nonquaesitum is a pathogenic fungi from the family Botryoshaeriaceae (Inderbitzin et al. 2010; Yang et al. 2017). Like most fungi from the family Botryoshaeriaceae, it is likely to occur as an endophyte (inhabit plant tissues without showing visible symptoms) on plant tissue or as latent opportunistic pathogen that cause disease symptoms when the host plant is subjected to physiological stress (Wilson 1995; Phillips et al. 2013). For example, incidences of disease symptoms (such as cankers and dieback) caused by Botryoshaeriaceae species were observed to coincide with severe drought in avocado production area in Chile (Valencia et al. 2019).

The family Botryoshaeriaceae is made up of morphologically diverse fungi that occur worldwide and are known to cause diseases such as cankers, necrosis, dieback and fruit rot on several woody plant species (Slippers and Wingfield 2007; Twizeyimana et al. 2013). They can infect fruit trees through wounds such as pruning wounds (Kotze et al. 2011).

Several members of the genus *Neofusicoccum* have been reported as pathogens of several perennial crop plants (McDonald and Eskalen 2011; Phillips et al. 2013). *Neofusicoccum nonquaesitum* has been reported to be associated with several perennial crop plants such as apples, avocado, almonds, grapevine, olives, and non-crop plant species (Table 5-16).

Symptoms

Neofusicoccum nonquaesitum has been reported to cause branch cankers and dieback in avocado in California, Chile, and Greece (Carrillo et al. 2016; Valencia et al. 2019; Guarnaccia et al. 2020). Branch dieback occurs when the fungus colonises the vascular tissue and affects the transport of water and nutrients causing blockage. This causes decay and weakens the infected plant and can result in reduced productivity of the plant (McDonald and Eskalen 2011).

Neofusicoccum nonquaesitum caused branch cankers, and branch dieback symptoms in avocado. Infected avocado branches exhibited branch dieback with dry brittle bark, powdery exudates with large cankers beneath the bark (Carrillo et al. 2016). It was one of the most virulent isolates among three others that was observed to cause severe damage in vascular tissues and stems of avocado resulting in dieback and branch cankers in avocados in Chile (Valencia et al. 2019).

Pathogenicity tests using avocado seedlings showed that *N. nonquaesitum* caused necrosis and lesions in the vascular system within 3-5 weeks (Carrillo et al. 2016; Valencia et al. 2019), fruiting bodies were observed on inoculated stems (Valencia et al. 2019).

Time to symptom development

Under laboratory conditions, *N. nonquaesitum* has been observed to cause disease symptoms on infected crop plants between 1 -5 weeks after inoculation (Rooney-Latham and Soriano 2016; Valencia et al. 2019; Guarnaccia et al. 2020). Pathogenicity tests using isolates of *N. nonquaesitum* inoculated on wounded one-year old avocado seedling using a mycelial plug, showed that symptoms (brown gum exudate and necrosis in the vascular system) were observed three weeks after inoculations (Valencia et al. 2019). In apples, symptoms became visible seven days post inoculation (Rooney-Latham and Soriano 2016).

Neofusicoccum nonquaesitum was used to inoculate a non-crop plant (two-three year old branches of *Umbellularia californica*) in its native environment and it exhibited symptoms (large necrotic lesions) 12 and18 months after inoculation (Lawrence et al. 2017).

Transmission and spread

Pathogens from the family Botryoshaeriaceae can infect plants through wounds caused by machines, pruning practices and through natural openings (McDonald and Eskalen 2011). Under field conditions, *N. nonquaesitum* was isolated from infected symptomatic (powdery exudates from cankered branches and stems) avocado plants (Carrillo et al. 2016; Guarnaccia et al. 2020), and they produced fruiting bodies on inoculated stems during pathogenicity tests (Valencia et al. 2019). Species from the family Botryoshaeriaceae spread by water splash, wind and the use of infected material (Whiteman 2004; Amponsah et al. 2009). As such, *N. nonquaesitum* can potentially be spread by water splash, wind, and the use of infected pruning material.

Conditions for symptom development

Infections in orchards affected by branch canker and dieback diseases caused by Botryosphaeriaceae species can remain dormant until environmental conditions become favourable for the pathogens (Menge and Ploetz 2003). This is because they are generally known to cause symptoms when an infected plant is subjected to physiological stress such as water stress (Udayanga et al. 2011).

Optimal growth Conditions

Isolate of *N. nonquaesitum* was collected (along with other pathogenic fungi) in spring from symptomatic and asymptomatic avocado orchards in Chile (Valencia et al. 2019). Under laboratory conditions, mycelial growth of isolates of *N. nonquaesitum* were observed to grow in culture within temperature range of 5° to 35℃ with optimal growth of 25℃ in culture (Valencia et al. 2019).

Table 5-16: Plant host range and diseases caused by *Neofusicoccum nonquaesitum*

Continent	Country	State/Region	Host plants	Symptoms /disease	CMI*	Reference
South America	Chile	Araucaria	*Araucaria araucana* (Molina)	Branch dieback, cankers, and decline	0.8	(Pérez et al. 2018)
		Illapel and Peumo	*Perseae americana* (avocado)	Branch dieback and Cankers	0.8	(Valencia et al. 2019)
		Panguipulli and Teodoro Schmidt	*Vaccinium corymbosum* (blueberry)	Cankers and twig dieback	0.8	(Pérez F et al. 2014)
North America	United States	California	*Persea americana* (avocado)	Branch dieback	0.5-0.8	(Carrillo et al. 2016)
		California	*Lithocarpus densiflora* (tanoak)	Bleeding bole cankers	0.5-0.8	(Rooney-Latham et al. 2012)
		California	*Malus domestica* (apples)	Branch cankers and dieback	0.5-0.8	(Rooney-Latham and Soriano 2016)
			Prunus dulcis (almonds)	Branch cankers	0.5-0.8	(Inderbitzin et al. 2010)
		California	*Sequoiadendron gigantea* (Giant sequoia)	Branch cankers	0.5-0.8	(Rooney-Latham et al. 2012)
		California	*Sequoia sempervirens* (coast redwood)	Branch cankers	0.5-0.8	(Rooney-Latham et al. 2012)
		California	*Umbelluria californica* (California bay)	Dieback and cankers	0.5-0.8	(Lawrence et al. 2017)

* (Phillips et al. 2018)

5.15.4 Likelihood of entry

Neofusicoccum nonquaesitum is known to affect a wide variety of host plants, including avocado (Table 5-16). *Neofusicoccum nonquaesitum* has been isolated from avocado branches indicating that they can be associated with leafless avocado budwood (Carrillo et al. 2016; Valencia et al. 2019; Guarnaccia et al. 2020).

Under field conditions, *N. nonquaesitum* caused branch cankers and dieback symptoms on avocado seedlings (Carrillo et al. 2016; Valencia et al. 2019; Guarnaccia et al. 2020). Avocado budwood with established *N. nonquaesitum* infections can potentially exhibit symptoms such as cankers and dieback of tissues. As such, infected budwood could be detected and destroyed during preparation for export, on phytosanitary inspection pre-export (if this occurs), or on arrival in New Zealand.

Species from the family Botryosphaeriaceae can occur as latent opportunistic pathogens that do not cause disease symptoms (asymptomatic) on host plants (Menge and Ploetz 2003). They can cause symptoms when an infected plant is subjected to physiological stress such as water stress (Wilson 1995; Udayanga et al. 2011). Like other endophytic latent pathogens in their family, it is possible that *N. nonquaesitum* can occur on asymptomatic leafless, rootless avocado budwood, and therefore might not be the detected by visual inspection during preparation for export or on arrival in New Zealand.

Pathogenicity tests using isolates (mycelial plug) of *N. nonquaesitum* inoculated on wounded seedlings and young plants (avocado and apples) found that symptoms were expressed within 1 -5 weeks (Rooney-Latham and Soriano 2016; Valencia et al. 2019; Guarnaccia et al. 2020). A non-crop plant (2-3 year old branches of *Umbellularia californica*) inoculated in its native environment exhibited symptoms 12-18 months after inoculation. As such, the active growing period at level 3A PEQ (a minimum of three distinct phenological growing events) will likely result in expression of symptoms. However, it is uncertain how long it will take for symptoms to be expressed in crop plant species in natural infection where inoculum might be at much lower concentrations.

Given the arguments and evidence above, that is:

- *Neofusicoccum nonquaesitum* has been isolated from stems and branches of avocado plants and can be associated with leafless avocado budwood.
- avocado budwood with established *N. nonquaesitum* can exhibit symptoms such as cankers on plant tissue that can be detected during preparation for export or on arrival in New Zealand.
- *Neofusicoccum nonquaesitum* can potentially be present on asymptomatic leafless avocado budwood and might not be detected by visual inspection during preparation for export and upon arrival in New Zealand, with high uncertainty.
- if *N. nonquaesitum* is imported on leafless avocado budwood, the active growing period at level 3A PEQ (a minimum of three distinct phenological growing events) will likely result in expression of symptoms.

The likelihood of *N. nonquaesitum* entering New Zealand associated with *Persea americana* budwood is considered to be ***low***, with ***moderate*** uncertainty. The uncertainty is because much of the evidence is based on characteristics of the family rather than on the species themselves.

5.15.5 Likelihood of exposure

This assessment is made on the basis that *N. nonquaesitum* has entered New Zealand undetected.

When a pest or disease arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *N. nonquaesitum*.

If avocado plants are infected with *N. nonquaesitum* at the time of their release from Post Entry Quarantine they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *N. nonquaesitum* in New Zealand from *Persea americana* budwood is considered ***high***, with ***low*** uncertainty.

5.15.6 Likelihood of establishment

This assessment is made on the basis that *N. nonquaesitum* has been successfully exposed to a suitable host plant in the New Zealand environment.

Neofusicoccum nonquaesitum is recorded to be present in a number of countries. Countries where both species have been recorded have CMI ranges of between 0.5-0.8 (Table 5-16). A CMI of ≥ 0.7, indicates a similar climate to the whole of New Zealand (Phillips et al. 2018). This suggests that the climatic conditions in some parts of New Zealand are likely to be suitable for them to establish, especially the North Island where climatic conditions are warmer. Therefore, climate is not likely to be a barrier for the establishment of *N. nonquaesitum* in New Zealand, especially the north of the North Island.

Neofusicoccum nonquaesitum is known to infect several host plants (Table 5-16) including crop plants such as apples, avocado and blueberry. These crop plant species are commercially grown in different parts of New Zealand. For example, avocado plants are widely grown commercially in orchards and in-home gardens mostly in the North Island and some parts of the South Island (Chapter 2). *Neofusicoccum nonquaesitum* has also been associated with widespread amenity plants such as oak tree species (Table 5-16), which are commonly grown in gardens and parks. Therefore, suitable hosts are available for *N. nonquaesitum* to establish and spread in all parts of New Zealand.

Species like *N. nonquaesitum* can spread by the movement of infected nursery stock for planting and the use of contaminated tools used for grafting and pruning (Whiteman 2004). Under field conditions, powdery exudates from cankered branches and stems were observed on avocado plants infected with *N. nonquaesitum* (Carrillo et al. 2016; Guarnaccia et al. 2020), and they produced fruiting bodies on inoculated stems during pathogenicity tests (Valencia et al. 2019). These fruiting bodies can be a source of infection and can be spread to uninfected plants by water splash, wind, and the use of infected pruning material.

Given the arguments and evidence above, that is:

- *Neofusicoccum nonquaesitum* is reported to be present some areas which have similar climate to New Zealand (Table 5-16). The climatic conditions in some parts of New Zealand are likely to be suitable for them to occur and spread, especially the North Island where climatic conditions are warmer
- *Neofusicoccum nonquaesitum* is associated with a range of crop and non-crop host plants (Table 5-16) that are widespread and available in New Zealand.
- *Neofusicoccum nonquaesitum* can be spread through the human movement of infected nursery stock and contaminated pruning material.

the likelihood of *Neofusicoccum nonquaesitum* establishing in New Zealand is considered *high*, with *low* uncertainty.

5.15.7 Impacts in New Zealand

Neofusicoccum nonquaesitum has been reported to be associated with several perennial crop plants such as apples, avocado, almonds and some non-crop plant species (Table 5-16). They can cause branch cankers and dieback symptoms in avocado (Carrillo et al. 2016; Valencia et al. 2019; Guarnaccia et al. 2020) and other perennial crops (Table 5-16). Branch dieback and dieback symptoms causes decay and weakens the infected plant and can affect the productivity of the plant (McDonald and Eskalen 2011). *Neofusicoccum nonquaesitum* can be spread by the movement of infected nursery stock for planting and the use of contaminated tools used for grafting and pruning and water splash (Whiteman 2004; Amponsah et al. 2009).

Economic impacts

Neofusicoccum nonquaesitum can cause branch cankers and dieback symptoms in avocado. This can affect productivity and yield of avocado, leading to economic loss in the avocado industry (Carrillo et al. 2016; Valencia et al. 2019; Guarnaccia et al. 2020). Avocado is increasingly becoming a significant horticultural crop in New Zealand. At the end of the 2020 financial year, 39,078 tonnes of avocado produced in New Zealand and earning up to NZ$56 million and NZ$110 million in domestic and international sales respectively (Plant & Food Research 2020). Therefore, infections by *N. nonquaesitum* can potentially result in yield loss which will have an impact on both the domestic and international market.

Neofusicoccum nonquaesitum has been reported to affect other economically important host plants by causing symptoms such as dieback and tree/branch cankers. As such economic impact caused by *N. nonquaesitum* is not limited to avocado. They can lead to reduced productivity of other perennial crop plants. For example:

- *Neofusicoccum nonquaesitum* can cause necrosis in apples in (Rooney-Latham and Soriano 2016).This can potentially lead to reduced productivity and reduced yield of infected trees. Apples are one of the most significant horticulture export crops for New Zealand worth up to NZ$876 million in export in 2020 (Plant & Food Research 2020).
- *Neofusicoccum nonquaesitum* has been reported to be associated with blueberry in Chile (Pérez et al. 2018). Botryosphaeriaceae species are known to be important pathogens that affect blueberries worldwide (Burgess et al. 2019; Scarlett et al. 2019). If *N. nonquaesitum* establishes in New Zealand; it can potentially lead to reduced productivity and reduced yield of infected plants. Blueberries are increasingly becoming important economic plant in New Zealand worth about NZ$44.4 million in both domestic and export in 2020 (Plant & Food Research 2020).

Given the arguments and evidence above, that is:

- *Neofusicoccum nonquaesitum* has been reported to be a pathogen of avocado that can potentially result in reduced productivity and yield loss. This can have an impact on both the domestic and international marketability of avocado
- *Neofusicoccum nonquaesitum* can cause disease and result in reduced yields of other economic important host plants such as apples and blueberries resulting in additional economic loss.

the economic impact of *N. nonquaesitum* to New Zealand is considered to be *moderate*, with *moderate* uncertainty.

Environmental impacts

Neofusicoccum nonquaesitum has been reported to cause branch dieback and decline diseases in woody plant species such as oak trees and native conifers in Chile and the United States (Table 5-16). For example, in California, *Neofusicoccum nonquaesitum* have caused cankers and dieback disease in native plants (Giant sequoia, coast redwood, and coast redwood) (Rooney-Latham et al. 2012). However, it is uncertain as to whether redwood species in New Zealand would be susceptible to infection by *N. nonquaesitum*. *Neofusicoccum parvum* which is a closely related species to *N. nonquaesitum* has been reported to infect *Araucaria* sp., *Populus nigra* and *Tibouchina lepidota* in New Zealand (Sakalidis et al. 2013). As such, if *N. nonquaesitum* establishes in New Zealand, they can possibly infect other non-crop species. Such infected plants can be a source of infection (inoculum) of crop plant that might be grown in close proximity (Inderbitzin et al. 2010; Burgess et al. 2019).

Given the arguments and evidence above, the impact on the environment from the establishment of *N. nonquaesitum* in New Zealand is considered to be low, with low uncertainty.

Human health impacts

Searches using the search term '*Neofusicoccum nonquaesitum* impacts on health' from Google, Google scholar, CABI and Pubmed found no record of *N. nonquaesitum* causing any health impact on humans and animals.

Given the arguments and evidence above, the human health impact of *N. nonquaesitum* to New Zealand is considered to be *negligible*, with *low* uncertainty.

Sociocultural impacts

Avocado has become popular and is widely consumed in New Zealand. It is commonly grown in home gardens in some parts of the country. Diseases caused by *N. nonquaesitum* can potentially lead to reduced productivity of avocado trees which can cause scarcity of avocado for consumers. Some oak species in New Zealand are amenity species, some of which are planted as landscaping plants in towns.

Given the arguments and evidence above, the sociocultural impact of *N. nonquaesitum* to New Zealand is considered to be *low,* with *low* uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, human health and society is considered to be *moderate*, with *moderate* uncertainty.

5.15.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry *low*, with *moderate* uncertainty.
- assuming entry, the likelihood of exposure *high*, with *low* uncertainty
- assuming successful exposure, the likelihood of establishment *high*, with *low* uncertainty.
- the overall impact on the New Zealand economy, environment, human health and society is considered to be *moderate*, with *moderate* uncertainty.

the overall level of assessed risk to New Zealand from *N. nonquaesitum* on imported *Persea americana* budwood (as in the commodity description) is considered to be *moderate*, with *moderate* uncertainty.

5.15.9 Management considerations

Neofusicoccum nonquaesitum causes branch cankers and dieback in avocado (Carrillo et al. 2016; Valencia et al. 2019; Guarnaccia et al. 2020). Avocado branches infected with *N. nonquaesitum* exhibited branch dieback with dry brittle bark, powdery exudates and large cankers beneath the bark (Carrillo et al. 2016).

In general, Botryoshaeriaceae species can cause disease symptoms which can develop when plants are subjected to physiological stress (Phillips et al. 2013). In avocado production areas in Chile, incidence of disease symptoms (cankers and dieback) caused by Botryoshaeriaceae species such as *N. nonquaesitum* was observed to coincide with severe drought (Valencia et al. 2019; Agustí-Brisach et al. 2020). This suggests symptoms on seedlings derived from infected budwood are likely to be expressed in dry conditions.

Samples can be taken for testing from any part of the avocado leafless budwood and testing different parts of the budwood might be required. This is because *N. nonquaesitum* can be present on plant tissues without causing symptoms (Twizeyimana et al. 2013), and can cause disease symptoms which can develop when plants are subjected to physiological stress (Phillips et al. 2013).

Molecular techniques (PCR) can be used to confirm if avocado budwood or seedlings derived from budwood are infected with *N. nonquaesitum* (Inderbitzin et al. 2010). However, it is not clear if it will be evenly distributed or throughout an infected avocado budwood; therefore, there is a high chance of false negatives.

An optimum season or conditions for conducting *N. nonquaesitum* detection testing has not been specified from the literature.

Similar species from the genus *Neofusicoccum* such as *N. parvum,* and *N. australe* are recorded as present in New Zealand and have been associated with perennial crop plants such as grapevine causing dieback disease (Amponsah et al. 2009; Tennakoon et al. 2018). Measures used to manage diseases caused by *N. parvum* are likely to manage diseases caused by *N. nonquaesitum*.

5.15.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Neofusicoccum nonquaesitum causes branch cankers and dieback in avocado (Carrillo et al. 2016; Valencia et al. 2019; Guarnaccia et al. 2020). Avocado branches infected with *N. nonquaesitum* exhibited branch dieback with dry brittle bark, powdery exudates and large cankers beneath the bark (Carrillo et al. 2016).

- ***What are the known environmental conditions conducive to symptom expression?***

In general, Botryoshaeriaceae species can cause disease symptoms which can develop when plants are subjected to physiological stress (Phillips et al. 2013). In avocado production areas in Chile, incidence of disease symptoms (cankers and dieback) caused by Botryoshaeriaceae species such as *N. nonquaesitum* was observed to coincide with severe drought (Valencia et al. 2019; Agustí-Brisach et al. 2020). This suggest that in dry conditions, seedlings derived from infected budwood are likely to be stressed and could express symptoms.

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Samples can be taken for testing from any part of the avocado leafless budwood and testing different parts of the budwood might be required. This is because species such as *N. nonquaesitum* survive on plant tissues without causing symptoms (Menge and Ploetz 2003) and cause disease symptoms which can develop when plants are subjected to physiological stress (Phillips et al. 2013). However, it is not clear if the fungi will be evenly distributed throughout an infected avocado budwood; therefore, there is a high chance of false negatives. Molecular techniques (PCR), can be used to confirm if avocado budwood or seedlings derived from budwood are infected with *N. nonquaesitum* (Inderbitzin et al. 2010).

- ***What is the optimum season for conducting detection testing?***

An optimum season or conditions for conducting *N. nonquaesitum* detection testing has not been specified from the literature.

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5.16 *Pestalotiopsis longiseta*

Pestalotiopsis longiseta has been reported to cause dieback disease in *Persea americana* (avocado) in China.

5.16.1 Taxonomic description

Scientific name: *Pestalotiopsis longiseta* (Speg.) K.Dia & Tak. Kobay (1990).

Order/Family: Xylariales/Amphisphaeriaceae

Other names include: *Pestalotia longiseta* (Dai et al. 1990).

Taxonomic notes: The genus *Pestalotiopsis* was recently segregated into three different genera (*Pestalotiopsis, Pseudopestalotiopsis* and *Neopestalotiopsis*). This was derived based on DNA sequencing and morphological characteristics (Maharachchikumbura et al. 2014).

About 16 species from the genus *Pestalotiopsis* are recorded as present in New Zealand (Biota NZ 2022). *Pestalotiopsis clavispora* and *P. versicolor* have been isolated from *Persea americana* in New Zealand (Biota NZ 2022). *Pestalotiopsis clavispora* has been reported to cause stem-end rot disease of avocado in Chile (Valencia et al. 2011) and *P. versicolor* has also been reported to cause postharvest disease in avocado in South Africa (Darvas and Wehner 1987).

5.16.2 Hazard identification

Pestalotiopsis longiseta is not known to be present in New Zealand:

- o *Pestalotiopsis longiseta* is not listed in either Biota NZ (2022) or PPIN (2021).
- o *Pestalotiopsis longiseta* is a regulated pest for New Zealand (ONZPR 2021)
- o *Pestalotiopsis longiseta* has unwanted status and is a notifiable organism (ONZPR 2021)

Pestalotiopsis longiseta has the potential to establish (and spread) in New Zealand because:

- Avocado which is a known host plant is grown commercially and also in home gardens in most parts of New Zealand.
- It is not likely to be restricted by climate because it occurs in both tropical and temperate climate and in countries with similar climate (CMI ≥ 0.7 (Phillips et al. 2018)) to most parts of New Zealand.
- It can spread by the human movement of infected nursery stock for planting.

Pestalotiopsis longiseta has the potential to cause harm to New Zealand because:

- Having been recently associated with avocado, it can potentially affect avocado crops and cause yield and economic loss.
- It can infect other host plants and cause crop loss, for example, it was reported to cause 10-20% loss of tea (*Camellia sinensis*) in India and Japan and up to 10% loss in crop yield of Persimmons (*Diospyros kaki*) in Brazil.

Pestalotiopsis longiseta is associated with *Persea americana* budwood because:

- It is a known host of *Pestalotiopsis longiseta* (Lin et al. 2018);
- It was recently reported to cause dieback symptoms in young avocado plants after it was isolated from an avocado plant nursery in China (Lin et al. 2018).

Given the arguments and evidence above, *Pestalotiopsis longiseta* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.16.3 Risk assessment

Biology and epidemiology of Pestalotiopsis longiseta

Biology

Pestalotiopsis is a species rich genus known to cause plant diseases (Kang et al. 1999). They have often been isolated as endophytes and are also known to occur as saprobes (Watanabe et al. 2010; Maharachchikumbura et al. 2014). They are common in both tropical and temperate ecosystems (Sutton 1980; Fail and Langenheim 1990). Species from this genus have received attention because they produce various antimicrobial active compounds significant to pharmaceutical science (Uzma et al. 2018; Elkhateeb and Daba 2021).

As pathogens, *Pestalotiopsis* species can cause a variety of diseases such as; cankers, shoot dieback, leaf spots, needle blight, grey blight, scabby canker, chlorosis, fruit rot and other postharvest diseases (Kang et al. 1999; Crous et al. 2011; Maharachchikumbura et al. 2014). They are considered weak pathogens because they are not host specific but opportunistic and are able to infect a wide range of host plants (Elliott 2006; Joshi et al. 2009; Kamhawy et al. 2011). However, they have caused diseases that have resulted in reduced productivity and resulted in economic loss crop plant species such as *Camellia sinensis* (tea), *Magnifera indica* (mango) and *Cocos nucifera* (Coconut) (Joshi et al. 2009; Ismail et al. 2013; Maharachchikumbura et al. 2013).

Pestalotiopsis spp. produce asexual spores (conidiospores) and like most endophytic pathogens, they have been demonstrated to inhabit tree barks and leaves without causing symptoms. Symptoms can eventually develop when the host plant is stressed or wounded (Maharachchikumbura et al. 2011). The conditions that can lead to expression of symptoms are usually hard to predict. Some species can be present on leaves of their host plant all year round and become an inoculum source on mature fruits which can eventually develop postharvest rot (Ullasa and Rawal 1985). Spores also can survive harsh weather conditions and are able to cause infections when they get transferred onto a suitable host plant (Maharachchikumbura et al. 2011).

Host Range

Pestalotiopsis longiseta was recently reported to cause dieback symptoms in young avocado plants after being isolated from an avocado plant nursery in China (Lin et al. 2018). It is known to cause diseases in other crops such as *Diospyros kaki* (Japanese persimmon), *Camellia sinensis* (tea) in Japan and Thailand and leaf spot in *Actinidia deliciosa* (kiwifruit) in Iran (Table 5-17).

Pestalotiopsis species have been associated with *Actinidia deliciosa* (kiwi fruit) (Ushiyama et al. 1996; Ertaș and Karakaya 2018). *Pestalotiopsis longiseta* among other *Pestalotiopsis* species is reported to have been isolated from leaf spots on kiwi fruit leaves in orchards in Iran. In this study, only the morphological characteristics of the isolates were used for identification (Mousakhah et al. 2014). The taxonomy of members of the genus *Pestalotiopsis* is complex and difficult to classify. (Maharachchikumbura et al. 2011). Isolates of *Pestalotiopsis* species can have similar morphological characteristics with slight differences that makes them difficult to group (Keith et al. 2006). As such, a combination of both molecular techniques and morphological characteristics is more reliable for species identification (Maharachchikumbura et al. 2011; Maharachchikumbura et al. 2012).

Table 5-17 The distribution, hosts and diseases caused by Pestalotiopsis longiseta

Continent	Country	State/Region	Host plant	Symptoms/diseases	CMI*	References
Asia	China	Hainan Province	*Persea americana* (avocado)	Dieback	0.4-0.5	(Lin et al. 2018)
	Japan	Torrori Prefecture	*Diospyros kaki* (Japanese persimmon)	Lesions on leaves, ring spot on fruit calyxes	0.7	(Yasuda et al. 2003)

Continent	Country	State/Region	Host plant	Symptoms/diseases	CMI*	References
	Japan	Shizuoka Plateau	*Camellia sinensis* (Tea)	Grey blight	0.7	(Omatsu et al. 2012; Yamada et al. 2016)
	Thailand	Chai Mai	*Camellia sinensis* (Tea)	Grey blight	0.5	(Maharachchikumbura et al. 2013)
	Iran	Guilan Province	*Actinidia deliciosa* (Kiwi fruit)	Leaf spots	0.6-0.8	(Mousakhah et al. 2014)
	India	Arunachal Pradesh and Papum Pare	*Phrynium capitatum* Willd	Endophytic (symptomless) infection	0.5-0.8	(Sharma et al. 2020).

* (Phillips et al. 2018)

Transmission and Spread

Pestalotiopsis longiseta can be transmitted and spread by both natural and artificial means through rain splash and from plucking tools and equipment (Yamada et al. 2016). Natural transmission velocity estimates have shown that fungal spores dispersed by rain splash will normally land <1m from source (Fitt et al. 1989; Fitt et al. 2006). Spores carried by water splash (rain or irrigation) that happen to be deposited on plants with wounds are likely to infect host plants (Yamada et al. 2016).

Hand plucked tea farms were less susceptible to infection by *P. longiseta* as most infections occurred through wounds on leaves and stems caused by machine plucking (Horikawa 1986). *Pestalotiopsis* species can occur in cuticles and intercellular spaces of leaves and cause disease symptoms when it becomes damaged (Fail and Langenheim 1990).

Wounds on plants appears to be necessary for transmission of *Petsalotiopsis* species (Elliott 2006; Chen et al. 2018). For example during a pathogenicity test on tea leaves, wounded leaves developed symptoms in contrast to unwounded leaves that exhibited little or no symptoms (Chen et al. 2018). Wounds caused by insect damage have been reported to enhance infection by *Pestalotiopsis* species. Insect damage was reported to be necessary for disease development in *Elaeis guineensis* (Africa oil palm) (Elliott 2006).

Symptoms on Persea americana (avocado)

Nursery avocado plants infected by *Pestalotiopsis longiseta* exhibited blight on the green shoot initially, it progressed to necrotic lesions on the stems and leaves. This eventually resulted in dieback symptoms of shoots and twigs. During pathogenicity testing, all inoculated 2-year old potted avocado plants started to blight and form necrotic lesions in seven days. The green shoots developed dieback symptoms by day 15 (Lin et al. 2018).

Symptoms on other plant species

Pestalotiopsis longiseta caused ring spots on leaves and calyxes of young fruits of persimmon. It also caused lesions that resulted in defoliation of leaves. During pathogenicity testing, it produced similar symptoms (large lesions on leaves) observed from natural infection 15 days after inoculation (Yasuda et al. 2003). In tea growing areas in Japan, it caused severe lesions in tea crops causing leaves (Hamaya and Horikawa 1982) and shoots to wither, thereby affecting the quality of the tea (Hamaya and Horikawa 1982; Ando et al. 1987). However susceptibility varied among different tea cultivars (Takeda 2002). It has also been reported to cause leafspots of kiwifruit plants (Mousakhah et al. 2014).

5.16.4 Likelihood of entry

Pestalotiopsis longiseta has been reported to be present in five countries and is known to affect some crop plants including avocado (Table 5-17). It was recently reported to cause dieback symptoms in young avocado plants when it was isolated from an avocado plant nursery in China (Lin et al. 2018).

During pathogenicity test, 2-year old potted avocado plants were inoculated with *P. longiseta* and in seven days all inoculated plants started to blight and form necrotic lesions. The green shoots developed dieback symptoms by day 15 (Lin et al. 2018). In a pathogenic phase of infection, symptoms are likely to be expressed on infected avocado budwood during the active growing period at level 3A PEQ (a minimum of three distinct phenological growing events).

Species from the genus *Pestalotiopsis* have been demonstrated to inhabit tree barks and leaves without causing symptoms (latent endophytic infection). They can eventually develop symptoms when the host plant is stressed or wounded (Maharachchikumbura et al. 2011). *P. longiseta* has been isolated as an endophyte from leaves, stems and roots of a wild plant species (Sharma et al. 2020). This suggests that it can potentially be present on leafless avocado budwood and not exhibit symptoms during the growing period in PEQ. However, there is no study that has investigated endophytic infection of *P. longiseta* in crop plant species.

Given the arguments and evidence above, that is:

- *Pestalotiopsis longiseta* is reported to be present in five countries and has been associated with avocado and other plants.
- In its pathogenic phase of infection, it is likely to exhibit symptoms if infected budwood is grafted in PEQ level 3A.
- It could be present on avocado budwood (in a latent or endophytic phase) without exhibiting symptoms especially if the budwood are not physiologically stressed or wounded.

the likelihood of *P. longiseta* entering New Zealand associated with *P. americana* budwood is considered to be **low**, with **moderate** uncertainty.

5.16.5 Likelihood of exposure

This assessment is made on the basis that *P. longiseta* has entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *P. longiseta*.

If avocado plants are infected with *P. longiseta* at the time of their release from PEQ they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *P. longiseta* in New Zealand from *P. americana* budwood is considered **high**, with **moderate** uncertainty.

5.16.6 Likelihood of establishment

This assessment is made on the basis that *P. longiseta* has been successfully exposed to a suitable host plant in the New Zealand environment.

Pestalotiopsis longiseta is recorded to be present mainly in Asia: China, India Japan, Thailand and Iran (Table 5-17). The countries where it has been recorded to be present have a CMI range of 0.4-0.8. CMI of ≥ 0.7 indicates a similar climate to the whole of New Zealand (Phillips et al. 2018). Species from the genus *Pestalotiopsis* are reported to be common in both tropical and temperate regions (Sutton 1980; Fail and Langenheim 1990). This suggests that the climatic conditions in some parts of New Zealand are likely to be suitable for it to establish. About 16 species from the genus *Pestalotiopsis* are recorded to be present in New Zealand (Biota NZ 2022). This indicates that climate is likely not to be a barrier to the establishment of *P. longiseta* in New Zealand.

Pestalotiopsis longiseta has been reported to infect and cause diseases in crop plant species (avocado, Japanese persimmons,tea and kiwifruit). Avocado which is a known host plant is widely grown commercially in orchards and in home gardens in the North Island and some parts of the South Island of New Zealand (Chapter 2). Therefore, suitable hosts are available for *P. longiseta* to establish and spread in New Zealand. Members of the genus *Pestalotipsis* are generally not host specific but opportunistic (Elliott 2006; Joshi et al. 2009; Kamhawy et al. 2011), *P. longiseta* can potentially infect other host plants and establish in New Zealand.

Most *Pestalotiopsis* infections occur through wounds and natural openings on the host plant. Species of the genus *Pestalotiopsis* can occur in cuticles and intercellular spaces of leaves and cause disease symptoms when leaves become damaged (Fail and Langenheim 1990). *Pestalotiopsis longiseta* has been isolated as an endophyte from leaves, stems and roots of wild native plants in India (Sharma et al. 2020; Shukla 2020). As such, it can spread by the human movement of infected nursery stock for planting..

Given the arguments and evidence above, that is:

- *Pestalotiopsis longiseta* has been reported in four countries, some of which have a similar climate to New Zealand. Climate in most parts of New Zealand is suitable for it to occur and spread especially since about 16 species from the same genus are already known to be present in New Zealand.
- Avocado, one of its known host plants, is widely grown commercially and in home gardens in both the North and South Island of New Zealand. Additionally, because it is not host specific, it can potentially infect other host plants and spread.
- It can spread by the human movement of infected nursery stock for planting.

the likelihood of *P. longiseta* establishing in New Zealand is considered **moderate**, with **moderate** uncertainty.

5.16.7 Impacts in New Zealand

Pestalotiopsis longiseta has been reported to cause disease symptoms (dieback, lesions, leaf spot and gery blight) in crops such as persimmon, tea, kiwifruit as well as young avocado plants (Table 5-17). It can be transmitted and spread through spores carried by water splash (rain or irrigation) that happen to be deposited on plants with wounds (Yamada et al. 2016). *Pestalotiopsis longiseta* can establish and spread in New Zealand because avocado, kiwifruit and perssimons are grown commercially in orchards and in home gardens New Zealand. Broad range chemical fungicides such as QoI are commonly can be used to control disease symptoms caused by *Pestalotiopsis longiseta* (Bartlett et al. 2002; Yamada et al. 2016).

Economic impacts

Pestalotiopsis longiseta was recently reported to cause dieback symptoms in young avocado plants after being isolated from an avocado plant nursery in China. It caused dieback symptoms in up to 30% of young avocado plants in China (Lin et al. 2018). This is the first report of *P. longiseta* infecting avocado plants. Because it is considered to be a weak opportunistic pathogen, it is able to infect a wide range of crop plants (Maharachchikumbura et al. 2011). It's association with avocado could have possibly been as a result of an opportunistic infection. The fact that *P. longiseta* is known to affect mainly leaves and result in dieback symptoms (Yasuda et al. 2003; Yamada et al. 2016; Lin et al. 2018) suggests that young plants might be more susceptible to its pathogenic effect. It was observed to cause leaf spots that resulted in dieback symptoms of the young avocado nursery plants in China (Lin et al. 2018). It is possible that young avocado plants are more susceptible than mature established plants. Having been recently discovered to be associated with avocado, the impact it might have on avocado crop yield has not been assessed and quantified. However, the loss of 30% of young plants would have a significant impact on a single nursery grower who could be supplying several orchards.

Pestalotiopsis longiseta and other *Pestalotiopsis* spp. having been reported to cause grey blight disease in tea was estimated to cause b etween 10-20% crop loss in Japan (Horikawa 1986) and 17% crop loss in India (Joshi et al. 2009) as a result of the infection of young tea leaves. Although tea is widely consumed in New Zealand it is only grown on a small commercial scale in the Waikato region (zealong.com). If *P. longiseta* is to establish in New Zealand, the grower is likely to suffer some economic loss. However, the economic impact on tea crop production in New Zealand is not likely to be significant.

Pestalotiopsis longiseta has been reported to cause post-harvest rot in persimmons in Brazil resulting in an estimated 10% loss in crop yield. Persimmons are grown in New Zealand with a value of NZ$1.3 million and NZ$10.7 million in domestic and export markets respectively (Plant & Food Research 2020). It can potentially affect the crop yield of persimmons that might result in some economic impact in New Zealand.

About sixteen species from the genus *Pestalotiopsis* are reported to be present in New Zealand (Biota NZ 2022), two of which have been associated with avocado in Chile (Valencia et al. 2011) and South Africa (Darvas and Wehner 1987). So far none of the *Pestalotipsis* species present in New Zealand has been reported to cause crop loss or economic loss to crop planting in New Zealand.

Given the arguments and evidence above, that is:

- *Pestalotiopsis longiseta* is likely to result in the loss of young avocado nursery plants. Because it was only recently discovered to be associated with avocado, the impact it might have on avocado crop yield has not been assessed and quantified.
- Although it caused 10-20% crop loss of tea in India and Japan, tea crop has little economic significance in New Zealand as such not likely to have a significant impact on the economy
- It caused up to 10% loss in crop yield of Persimmons in Brazil. Persimmons have a value of NZ$12 million in domestic and export market.
- Several species form the genus *Pestalotiopsis* are reported to be present in New Zealand and so far, none has been reported to cause crop loss or economic loss of or other economic crop plants present in New Zealand.

the economic impact of *P longiseta* to New Zealand is considered to be **low**, with **moderate** uncertainty.

Environmental impacts

There is no record of *P. longiseta* having a significant impact on native plants and the environment. However, *Pestalotiopsis* species are ubiquitous and have been isolated as the endophytes and as saprobes from soil, polluted water and from leaves, bark and twigs (Maharachchikumbura et al. 2011). The fact that species from the genus are not host specific but mostly opportunistic (Elliott 2006; Joshi et al. 2009; Kamhawy et al. 2011) suggests that it can potentially infect plants in the environment. However, they are considered weak pathogen (Beresford and Mulholland 1982; Madar et al. 1991) therefore, their impact on the environment might not be significant.

Given the arguments and evidence above, the impact on the environment from the establishment of *P. longiseta* in New Zealand is considered to be **low**, with **low** uncertainty.

Human health impacts

Species from the genus *Pestalotiopsis* are commonly known plant pathogens that rarely cause human diseases (Borgohain et al. 2020). Few human infections caused by *Pestalotiopsis* species have been reported. One report involved a gardener that developed keratitis (eye infection) after sweeping twigs and leaves on a windy day and *Pestalotiopsis clavispora* was identified as causative agent (Monden et al. 2013). Another report of *Pestalotipsis* species affecting human health involved some *Pestalotiopsis* spp. recovered from sinuses, fingernails, bronchi, scalp, foot and eye (associated with onychomycosis disease) of a few individuals working in tea plantations in India. (Sutton 1999; Maharachchikumbura et al. 2011; Borgohain et al. 2020). Infected individuals were exposed to pathogenic fungi such as *Pestalotiopsis* species present in the plantations (Borgohain et al. 2020).

Sixteen *Pestalotiopsis* species are reported as present in New Zealand (Biota NZ 2022). There has been no report on any of *Pestalotiopsis* species infecting humans and causing health problems in New Zealand.

Given the arguments and evidence above, the human health impact of *P. longiseta* to New Zealand is considered to be **low**, with **low** uncertainty.

Sociocultural impacts

Avocado has become popular and is widely consumed in New Zealand. It is commonly grown in home gardens in some parts of the country. The potential impact of *P. longiseta* on avocado crop yield has not been assessed and quantified. However, 16 species from the same genus are reported as present in New Zealand, and two have been has been associated with avocado. This suggests that *P. longiseta* is not likely to cause scarcity of avocado fruits in local markets.

Given the arguments and evidence above, the sociocultural impact of *Pestalotiopsis longiseta* to New Zealand and is considered to be **negligible**, with **low** uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, human health and society is considered to be **low**, with **low** uncertainty.

5.16.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is **low** with **moderate** uncertainty
- assuming entry, the likelihood of exposure is **high** with **moderate** uncertainty
- assuming successful exposure, the likelihood of establishment is **moderate** with **moderate** uncertainty
- the overall impact on the New Zealand economy, environment, human health and society is considered to be **low** with **moderate** uncertainty.

the overall level of assessed risk to New Zealand from *P. longiseta* on imported *P. americana* budwood (as in the commodity description) is considered to be **low**, with **moderate** uncertainty.

5.16.9 Management considerations

Plants infected by *Pestalotiopsis longiseta* can exhibit blight on the green shoot initially, which can progress to form necrotic lesions on the stems and leaves. This can eventually result in dieback symptoms of shoots and twigs. During pathogenicity test, 2-year old potted avocado plants were inoculated with *P. longiseta* and in seven days all inoculated plants started to blight and form necrotic lesions. The green shoots developed dieback symptoms by day 15 (Lin et al. 2018).

Under laboratory conditions, inoculated avocado plants were covered with plastic bags for 48 hours and kept under high humidity at 28℃ and symptoms were observed to develop in seven days (Lin et al. 2018). However environmental conditions condusive for symptom development in field conditions has not been specified.

Broad range chemical fungicides such as QoI are commonly used to control gray blight disease caused by *Pestalotiopsis longiseta* (Bartlett et al. 2002; Yamada et al. 2016). Molecular techniques (genomic DNA using ITS1/ITS4) primers (Lin et al. 2018) can be used to test for asymptomatic infection and any part of the plant (budwood) can be tested for asymptomatic infections. Optimum conditions for conducting testing has not been specified.

5.16.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Plants infected by *Pestalotiopsis longiseta* can exhibit blight on the green shoot, which can progress to form necrotic lesions on the stems and leaves. This can eventually result in dieback symptoms of shoots and twigs. During pathogenicity testing, 2-year old potted avocado plants were inoculated with *P. longiseta* and in seven days all inoculated plants started to blight and form necrotic lesions. The green shoots developed dieback symptoms by day 15 (Lin et al. 2018).

- ***What are the known environmental conditions conducive to symptom expression?***

Under field conditions, environmental conditions condusive for symptom development caused by *P. longiseta* have not been specified. However, under laboratory conditions, inoculated avocado plants were covered with plastic bags for 48 hours and kept under high humidity at 28℃ and symptoms were observed to develop in seven days (Lin et al. 2018).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Any part of the plant (budwood) can be tested for asymptomatic infections.

- ***What is the optimum season for conducting detection testing?***

Optimum conditions for conducting testing have not been specified.

5.16.11 References

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5.17 *Phyllachora gratissima* (tar spot)

Phyllachora gratissima is a fungal leaf pathogen that causes tar spot on the leaves of *Persea americana* (avocado). It is considered a minor pathogen that is more frequent in 'Creole' avocados.

5.17.1 Taxonomic description

Scientific name: *Phyllachora gratissima* Rehm (1892)

Order/Family: Phyllachorales/Phyllachoraceae

5.17.2 Hazard identification

Phyllachora gratissima is not known to be present in New Zealand:

- *Phyllachora gratissima* is listed as absent from New Zealand in Biota NZ (2022) and NZOR (2021)
- *Phyllachora gratissima* is not listed in PPIN (2021) or ONZPR (2021)

Phyllachora gratissima has the potential to establish in New Zealand because:

- *Persea americana* (avocado) is a known host (Spaulding 1961; Molano 2007) that is widely grown in in New Zealand, in home gardens and commercial production, mainly in the north of the North Island and the Hawkes Bay region.
- *Phyllachora gratissima* causes tar spot in avocado and is recorded in countries with a climate match index (CMI) ≥ 0.7 (e.g. Bolivia, Colombia, Ecuador, Mexico, Venezuela) indicating a similar climate to the whole of New Zealand (Phillips et al. 2018).

Phyllachora gratissima has the potential to cause harm to New Zealand because:

- *Phyllachora gratissima* causes tar spot in avocado which can sometimes cause defoliation in trees (Spaulding 1961).
- avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$50.6 million (Plant & Food Research 2020).

Given the arguments and evidence above, *Phyllachora gratissima* meets the criteria to be a hazard for New Zealand

5.17.3 Risk assessment

Biology and epidemiology of Phyllachora gratissima

Hosts

Phyllachora gratissima is specific to *P. americana* (avocado) (Spaulding 1961; Hodges 1969).

Distribution

Phyllachora gratissima has been recorded in North, Central and South America (Table 5-18).

Table 5-18: Distribution of Phyllachora gratissima and the climate match index (CMI) of each country.

Region	Country	CMI*	Reference
North America	Mexico	0.3–0.8	Molano (2007)
Central America and the Caribbean	Costa Rica	0.3–0.6	Spaulding (1961)
	Guatemala	0.3–0.6	Spaulding (1961)
	Puerto Rico (USA)	0.5–0.6	Zentmyer (1961)
South America	Bolivia	0.4–0.8	Zentmyer (1961)
	Colombia	0.3–0.8	Spaulding (1961); Zentmyer (1961)
	Ecuador	0.4–0.8	Spaulding (1961); Zentmyer (1961)
	Venezuela	0.4–0.8	Spaulding (1961); Zentmyer (1961)

*(Phillips et al. 2018)

Symptoms

This disease is readily visible as symptoms resemble droplets of tar on leaf surfaces (Molano 2007; Thaung 2008). These spots look like small domes with a matte to shining black membrane (Thaung 2008). The black spots are surrounded by a chlorotic halo (Molano 2007). The tar spots increase in size and turn a brown colour on both the upper and underside of the leaf (Molano 2007). Infection can sometimes cause defoliation (Spaulding 1961).

Phyllachora species are minor plant pathogens that rarely kill host tissues (i.e. obligate biotrophs); however, the longevity of infection allows pathways for secondary infection by other pathogens (Tamakaew et al. 2017).

Time to symptom development

There is limited information regarding time to symptom development of *P. gratissima* in avocado; however, *Phyllachora* tar spot of bamboo in India forms lesions after 7-9 days (Thaung 2008). Other tar spot pathogens such as *P. maydis* form symptoms within 14 days and has a short latent period in *P. maydis* which ranges from 14 to 20 days post inoculation (Valle-Torres et al. 2020).

Conditions for symptom development have not been described for *P. gratissima*. However, *P. maydis* develops symptoms between 10 to 20℃ and development decreases above 25℃ (Thaung 2008).

Reproduction and transmission

There is limited information regarding reproduction and transmission of *P. gratissima* and thus information is based on other *Phyllachora* species. Ascospores, of *P. maydis*, are released by wind or rainsplash on foliage during periods of moderate temperature (16–23℃), leaf wetness duration of greater than 7 h per night and a relative humidity of 75% (Valle-Torres et al. 2020).

5.17.4 Likelihood of entry:

Phyllachora gratissima has been recorded in tropical climates (Table 5-18) and is predominantly a foliar pathogen (Tamakaew et al. 2017). Green budwood may include some leaf tissue within the buds and therefore, there is potential for *P. gratissima* to be imported into post entry quarantine (PEQ) on *P. americana* budwood.

Phyllachora gratissima appears to be a surface pathogen, so it is expected that disinfection of budwood entering PEQ (1% sodium hypochlorite dip for 2 minutes) would be effective in eliminating any infection on the surface. Although, it is uncertain if the fungus may persist in small crevices within the leaf buds.

If *P. gratissima* is imported into New Zealand, and disinfection were not suffient to remove the pathogen, it is very likely that it would produce visible symptoms in PEQ. There are no results of inoculation experiments available for avocado to determine the period to develop symptoms or the potential to be asymptomatic on this host. However, other tar spot pathogens (i.e. *P. maydis*) forms symptoms within 14 days (Valle-Torres et al. 2020). This suggests that if *P. gratissima* was present on avocado budwood imported to New Zealand, it is highly likely that symptoms would develop within the period of growth in PEQ.

Based on the mode of reproduction and transmission, there is no evidence available to suggest that *P. gratissima* would not be successfully contained within Level 3A PEQ conditions.

Given the arguments and evidence above, that is:

- there is limited potential for the association of *P. gratissima* with avocado budwood cuttings.
- infection by *P. gratissima* is highly likely to be detected during the period of growth in Level 3A PEQ.

Given the arguments and evidence above, the likelihood of *Phyllachora gratissima* entering New Zealand associated with *P. americana* budwood is considered to be negligible, with low uncertainty.

5.17.5 Overall level of associated risk to New Zealand

Based on the assessment of likelihood above, that:

- the likelihood of *P. gratissima* entering New Zealand associated with *P. americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ) is considered negligible, with low uncertainty;

the overall level of risk to New Zealand from *P. gratissima* on imported *P. americana* bedwood is considered to be negligible, with low uncertainty.

5.17.6 Management considerations

Phyllachora gratissima produces highly visible black spots on leaves of avocado which appear within days of infection (Spaulding 1961; Molano 2007; Thaung 2008). It is considered a minor pathogen which, in some cases, can cause defoliation (Spaulding 1961). Literature searches on the species found no evidence for asymptomatic infection of leaves or stem tissues. Therefore, *P. gratissima* is highly unlikely to persist as an asymptomatic infection on susceptible tissues.

Warm and humid conditions (16–23°C and 75% humidity) are conducive to disease expression and ascospore release (Valle-Torres et al. 2020). Detection of *Phyllachora* species can be conducted using PCR primers ITS4 and ITS5 to sequence the ITS region (i.e. common for many fungi) (Ruhl et al. 2016). However, given many *Phyllachora* species are obligate biotroiphs, they are considered unculturable (Ruhl et al. 2016).

5.17.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Phyllachora gratissima causes tar spot on the leaves of *P. americana*. Black dots (that resemble tar) appear on the surface of leaves surrounded by a chlorotic halo (Molano 2007; Thaung 2008). Spots increase in size and turn a brown colour that is seen on both the upper and underside of the leaf (Molano 2007). Infection may cause defoliation (Spaulding 1961).

- ***What are the known environmental conditions conducive to symptom expression?***

Moderate temperature between 10–25℃ and 75% relative humidity seem to be conducive to symptom expression of *Phyllachora* species (Thaung 2008; Valle-Torres et al. 2020).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested?***

Phyllachora gratissima is a foliar pathogen and thus symptomatic leaves should be tested. There is evidence that this genus has a short latency period suggesting that asymptomatic infection is unlikely (Valle-Torres et al. 2020).

- ***What is the optimum season for conducting detection testing?***

Seasons with moderate temperatures and relative humidity (Thaung 2008; Valle-Torres et al. 2020) seem to be conducive to symptom expression and would be optimal for detection testing.

5.17.8 References

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5.18 *Phyllosticta perseae* (leaf spot disease)

Phyllosticta perseae is a fungal pathogen known to cause leaf spot disease in multiple *Persea* species. Affected plants typically have dark spots in lesions at leaf edges and may lead to leaf death.

5.18.1 Taxonomic description

Scientific name: *Phyllosticta perseae* Ellis and G. Martin 1885

Order/Family: Botryosphaeriales/Phyllostictaceae

Other names include: *Phyllosticta micropuncta* Cooke 1878

Taxonomic notes: *Phyllosticta perseae* and *P. micropuncta* are often considered synonymous (Shetty et al. 2016; Farr and Rossman 2021; Ramirez et al. 2021) but some databases consider them as different species (Robert et al. 2005; Index Fungorum 2022). *Phyllosticta perseae* and *P. micropuncta* are likely considered synonymous by majority of literature because the earliest specimens were observed to be the same (Ramirez et al. 2021). A recent taxonomic revision of the genus *Phyllosticta* showed that they often occur as anamorphs of some *Guignardia* species but no evidence was found to show that *P. perseae* or *P. micropuncta* are anamorphs of any *Guignardia* species (Wulandari et al. 2013). There is very little information on *P. micropuncta* in the literature.

5.18.2 Hazard identification

Phyllosticta perseae is not known to be present in New Zealand:

- *Phyllosticta perseae* is not listed in NZOR (2021), Biota NZ (2022) or PPIN (2021).
- *Phyllosticta perseae* is not listed in ONZPR (2021).

Phyllosticta perseae has the potential to establish (and spread) in New Zealand because:

- *Persea americana* (avocado) is a known host (Zentmyer 1961; Rieuf and Teasca 1970; Lara Vazquez 2019) that is widely grown in New Zealand, in home gardens and commercial production, mainly in the north of the North Island.
- *Phyllosticta perseae* causes leaf spot disease in tropical and subtropical countries including Dominican Republic, India and South Africa (Doidge 1950; Zentmyer 1961; Das et al. 2005). Some regions in these areas have a climate match index (CMI) ≥ 0.7 indicating a similar climate to the whole of New Zealand (Phillips et al. 2018).

Phyllosticta perseae has the potential to cause harm to New Zealand because:

- it causes leaf spot on avocado (Blain 1931; Zentmyer 1961; Rieuf and Teasca 1970). The extent of leaf spot disease in avocado is still unknown but Rieuf and Teasca (1970) observed the pathogen on dead leaf tissue of avocado indicating that infection could lead to leaf death.
- avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$50.6 million (Plant & Food Research 2020).

Phyllosticta perseae is associated with *Persea americana* budwood because:

- *Persea americana* is a known host of *P. perseae* (Blain 1931; Zentmyer 1961; Rieuf and Teasca 1970).
- *Phyllosticta perseae* causes leaf spots and seems to strictly parasitise leaves; although there is no evidence to assert that the pathogen also infects stems, it is likely that spores could contaminate the crevices around buds or the surface of budwood .

Given the arguments and evidence above, *Phyllosticta perseae* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.18.3 Risk assessment

Biology and epidemiology of Phyllosticta perseae

Hosts and geographical distribution

Phyllosticta perseae is the causative agent of leaf spot disease in *Persea* spp including *Persea americana* (avocado) (Blain 1931; Zentmyer 1961; Ohr et al. 2003). Leaf spot is considered a minor problem in avocado orchards because it is an uncommon disease and there has been no record of this disease reducing yield (Zentmyer 1961; USDA 2016). *Phyllosticta perseae* has also been isolated from dead leaves of avocado (Rieuf and Teasca 1970).The pathogen has not been recorded to infect other plant genera. *Phyllosticta perseae* is found in both tropical and subtropical regions of the world (Table 5-19).

Table 5-19: Geopgraphic distribution of *Phyllosticta perseae*

Region	Country	CMI*	Host	References
Asia	India	0.2–0.9	*Persea americana* and *P. bombycina*	(Rao 1965; Das et al. 2005; Chattopadhyay et al. 2014; Ray et al. 2019)
Africa	Morocco	0.5–0.9	*P. americana*	(Rieuf and Teasca 1970)
	South Africa	0.6–0.9	*P. americana*	(Doidge 1950)
America	USA	0.5–0.9	*P. americana*, *P. carolinensis*, *P. borbonia,* and *Persea* sp.	(Ellis and Martin 1885; Anderson 1919; Blain 1931; Alfieri Jr 1984; Farr and Rossman 2021)
	Mexico	0.3–0.8	*P. americana*	(Lara Vazquez 2019)
	Dominican Republic	0.5–0.7	*P. americana*	(Zentmyer 1961)

* Climate Match Index (CMI) with all of New Zealand (Phillips et al. 2018).

Symptoms

There was no information regarding the severity of leaf spot disease caused by *Phyllosticta perseae* in *Persea americana*. Therefore, information on symptoms shown here is based on description of infection in *Persea bombycina* which may be slightly different in *Persea americana*.

Phyllosticta perseae causes brownish-gray spots around the edges of host leaves on both the dorsal and ventral surfaces (Ellis and Martin 1885; Chattopadhyay et al. 2014; Ray et al. 2019). Older leaves are more likely to be infected with the pathogen (Ray et al. 2019). Necrotic lesions around the spots on the edges of leaves may result in the eventual death of leaves (Chattopadhyay et al. 2014; Ramirez et al. 2021). The percentage of necrotic lesions on *Persea bombycina* leaves ranges between 19% – 47% of the surface area of the leaf (Chattopadhyay et al. 2014). In experimental inoculations, more than 30% of all leaves on a 3-month old Persea bombycine seedlings could be infected (Das et al. 2005). The total leaf infection in adult trees (8-10 year olds) may exceed 50% in severe cases (Chattopadhyay et al. 2014).

Reproduction and transmission

The sexual morph of *Phyllosticta perseae* is yet to be identified (Chattopadhyay et al. 2014). Rieuf and Teasca (1970) noted that the anamorph (i.e. asexual stage) of *P. perseae* produces pycnidia (i.e. asexual fruiting bodies). Typically, conidia (i.e. asexual spores) of *Phyllosticta* species can spread from infected leaves to uninfected leaves through rain or water splashes (Wikee et al. 2011). By spraying the leaves of 3-month old *P. bomybcina* seedlings with a concentrated solution containing *P. perseae* conidia, leaves became infected suggesting the pathogen can be transmitted by water (Das et al. 2005). Transmission and spread of the disease is favoured by rain, especially during the rainy season in India (Chattopadhyay et al. 2014).

The optimal temperature for the development of *P. persea* on avocado is unknown. However, the pathogen was shown to develop well on *P. bombycina* at a temperature of 25 ℃ (Das et al. 2005). The pathogen's viability on dead leaves of *Persea* spp is not known but *Phyllosticta citricarpa,* which affects *Citrus*, is known to be viable on dying or dead plant tissues for up to 45 days (de Oliveira Silva et al. 2017).

In-field management

There is no known management practice of *Phyllosticta perseae* in avocado orchards. However, management practice of *P. perseae* in *P. bombycina* orchards involves application 0.1% Mancozeb (a broad spectrum fungicide) two times before the peak rainy season (CMERTI 2021).

5.18.4 Likelihood of entry

Phyllosticta perseae causes leaf spots on avocado leaves (Rieuf and Teasca 1970) but has not been reported on stems. There is a possibility that *P. perseae* spores could contaminate leaf buds or the surface of relatively wet, leafless, rootless avocado budwood cuttings due to the transmission of spores through water or rain splash (Das et al. 2005; Chattopadhyay et al. 2014). However, there is currently no evidence to support this association. Furthermore, *P. perseae* is more likely to produce conidia on older leaves or dying and dead material (Rieuf and Teasca 1970; Chattopadhyay et al. 2014; Ray et al. 2019) and thus importing clean rootless budwood would considerably reduce the likelihood of entry on this pathway.

As part of the commodity description, all imported avocado germplasm will undergo a growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ. This period in 3A PEQ will impact on likelihood of entry in the following manner:

- any ungerminated *P. perseae* conidia on the surface of avocado budwood are highly unlikely to remain viable (alive and still capable of infecting the plant), based on the assumption of the known conidial viability of *P. citricarpa* (Korf et al. 2001; de Oliveira Silva et al. 2017), during preparation of the material for export, transit to New Zealand, surface disinfection (cuttings dipped in 1% sodium hypochlorite for 2 minutes) upon arrival in PEQ and subsequent grafting of the buds onto new rootstock.
- if viable conidia remain on a bud once the bud is grafted, infection is likely to occur once the bud breaks dormancy to form leaves. In this case, visible spots and lesions are likely to develop assuming the following conditions occurs, temperatures close to 25 ℃ and high humidity, and thus, be detected during routine inspections.
- Searches of Google Scholar, CAB abstracts, CPC and Google using the search terms '*Phyllosticta perseae*', '*Phyllosticta micropuncta*' and 'asymptomatic' found no evidence for asymptomatic *P. perseae* infections or longer latent periods in avocado plants[48]. Additionally, recent isolation of endophytic fungi of avocado trees in Florida did not find *P. perseae,* suggesting this pathogen may be restricted to the surface of leaves (Shetty et al. 2016). Budwood or grafted plants infected with *P. perseae* are therefore almost certain to develop symptoms and be detected and destroyed before they are released from PEQ.
- Conidia are spread by watersplash and rain (Wikee et al. 2011; CMERTI 2021). Level 3A PEQ has measures in place to manage the risks associated with insects, wastewater, waste and accidental transfer (see section 2.3), therefore viable *P. perseae* propagules are highly unlikely to escape level 3A PEQ to infect a suitable host.

Therefore, if *P. perseae* is imported on avocado, the residual biosecurity risks are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ.

Given the arguments and evidence above, that is:

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[48] Spanish and Portuguese references were also checked when they were found in searches on the species names.

- there is very limited evidence to suggest that *P. perseae* can be associated with leafless, rootless avocado budwood cuttings as the fungus is only known to be associated with leaves of host plants;
- residual biosecurity risks from new infections or ungerminated conidia on the surface of avocado budwood are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ;

the likelihood of *Phyllosticta perseae* entering New Zealand associated with *Persea americana* budwood is considered to be negligible, with low uncertainty.

Therefore, further assessment is considered unnecessary at this time; and likelihoods of exposure and establishment and impacts to New Zealand were not assessed.

5.18.5 Overall level of assessed risk to New Zealand

Based on the assessment of likelihood above, that:

- the likelihood of *P. perseae* entering New Zealand associated with *P. americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush' in 3A PEQ) is considered to be negligible with low uncertainty;
- therefore, continuing the assessment was considered unnecessary; and

the overall level of risk to New Zealand from *P. perseae* on imported *P. americana* budwood is considered to be negligible, with low uncertainty.

5.18.6 Management considerations

Phyllostica perseae produces visible spots and lesions on leaves (Chattopadhyay et al. 2014; Ramirez et al. 2021). Searches on the species name found no evidence for asymptomatic infection of leaves or stem tissues. Therefore, *P. perseae* is highly unlikely to persist as an asymptomatic infection on susceptible tissues.

No information was found on the time to development of symptoms on avocado, but high humidity and moisture conditions, and temperatures close to 25°C are known to induce symptoms in *P. bombycina* (Das et al. 2005). Conidia are more likely to germinate on older leaves or dying and dead material than new leaves (Rieuf and Teasca 1970; Chattopadhyay et al. 2014) and this suggests that symptoms of pre-existing *P. perseae* infections are highly likely to be detected by visual inspection during preparation for export or on arrival in New Zealand.

If viable conidia have remained on the surface of buds, infections are likely to develop when the plant breaks dormancy and produces new shoots. Given the conditions described above, temperatures in PEQ are likely to favour infection, but given that there is no overhead watering in PEQ (MPI 2021), there may not be enough water on the surface of the plant to allow conidia to germinate and infect susceptible tissues. Nevertheless, conidia are highly unlikely to remain viable on *P. americana* budwood through pre- and post-export handling and the PEQ period. Therefore, prophylactic testing in the absence of visible lesions is not considered necessary.

Phyllosticta spp. specific primers (Pca8/ITS4, GCM/GCMR, GCF2/GCR4) for nested polymerase chain reaction (PCR) have been developed, however, these were developed for *Citrus* leaves and fruits (Wang et al. 2012). This suggests that PCR techniques could be developed for diagnostic testing of *P. perseae* if symptoms are detected in PEQ.

5.18.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Phyllosticta perseae produces brownish-gray spots within small necrotic lesions around the ends and sides of host plant leaves (Ellis and Martin 1885; Ramirez et al. 2021). The disease mostly affects leaf quality and sometimes leads to leaf loss (Rieuf and Teasca 1970; Das et al. 2005).

- ***What are the known environmental conditions conducive to symptom expression?***

There is very little information about the conditions for symptom expression in *P. americana* but in *P. bombycina*, symptoms are best expressed in highly humid conditions and at a temperature of 25 ℃ (Das et al. 2005). Although, the pathogen was collected on leaves of *P. carolinensis* during winter in Florida (Ellis and Martin 1885), it is important to note that winter in Florida is mild because the climate ranges from tropical to subtropical. in Florida is largely tropical. In India, the pathogen is best transmitted and expressed during the rainy season, suggesting that wet and humid environmental conditions are most appropriate (Chattopadhyay et al. 2014).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested?***

Phyllosticta perseae is not known to cause asymptomatic infections.

- ***What is the optimum season for conducting detection testing?***

The pathogen is likely to develop and be expressed in the rainy season or warm, humid and wet conditions (Chattopadhyay et al. 2014).

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5.19 *Pleiocarpon algeriense* and *Cylindrocladiella peruviana*

Pleiocarpon algeriense and *Cylindrocladiella peruviana* are causative agents of black-foot disease of grapevines which causes necrosis at the base of root stocks, reduced vigor of plants and occasional mortality. In avocado trees, both of these organisms cause stem and crown rot.

5.19.1 Taxonomic description

Scientific name: *Pleiocarpon algeriense* W. Aigoun-Mouhous, A. Cabral & A. Berraf-Tebbal (2019)

Order/Family: Hypocreales/Nectriaceae

Scientific name: *Cylindrocladiella peruviana* (Bat., J.L. Bezerra & M. P. Herrera) Boesew. (1982)

Other names: *Cylindrocladium peruvianum* Bat., J. L. Bezerra & M. P. Herrera (1965)

Order/Family: Hypocreales/Nectriaceae

5.19.2 Hazard identification

Pleiocarpon algeriense and *Cylindrocladiella peruviana* are not known to be present in New Zealand:

- *Pleiocarpon algeriense* is not listed in Biota NZ (2022), NZOR (2021), PPIN (2021) or ONZPR (2021).
- *Cylindrocladiella peruviana* is listed as absent from New Zealand in Biota NZ (2022) and NZOR (2021) and is not listed in PPIN (2021).
- *Cylindrocladiella peruviana* is listed as regulated in ONZPR (2021).

Pleiocarpon algeriense and *Cylindrocladiella peruviana* have the potential to establish [and spread] in New Zealand because:

- *Persea americana* (avocado) is a known host (Aiello et al. 2020) that is widely grown in New Zealand, in home gardens and commercial production, mainly in the north of the North Island and the Hawkes Bay region.
- *Eucalyptus* spp. and *Vitis* spp. are also known hosts of *C. peruviana* (Crous and Wingfield 1993; van Coller et al. 2005) which are also widely grown in New Zealand.
- *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* have been recorded from countries (e.g. Italy) with a climate match index (CMI) ≥ 0.7 (Phillips et al. 2018) indicating a similar climate to the whole of New Zealand.

Pleiocarpon algeriense and *Cylindrocladiella peruviana* have the potential to cause harm to New Zealand because:

- *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* cause stem and crown rot in *Persea americana* plants which can result in the death of trees (Aiello et al. 2020).
- avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$50.6 million (Plant & Food Research 2020).

Given the arguments and evidence above, *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* meet the criteria to be a hazard for New Zealand

5.19.3 Risk assessment

Biology and epidemiology of Pleiocarpon algeriense and Cylindrocladiella peruviana

Pleiocarpon algeriense is a recently described organism (Aigoun-Mouhous et al. 2019) and was first isolated from the basal part of *Vitis* spp. rootstock in Algeria (Aigoun-Mouhous et al. 2019). It has also been isolated from *Persea americana* plants in Italy (Aiello et al. 2020).

Cylindrocladiella peruviana is associated with root and cutting rot of *Acacia mearnsii*, *Eucalyptus* spp. and *Vitis vinifera* (van Coller et al. 2005) in addition to leaf spot in *Acacia* spp. and *Eucalytus* spp. (Crous and Wingfield 1993). It has also been isolated from *Persea americana* plants in Italy (Aiello et al. 2020).

Symptoms

Both *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* cause wilting of avocado plants as a result of stem and crown rot (Aiello et al. 2020). Infected plants have stunted growth, general wilting and leaf chlorosis; the basal stem and crown of plants become dry, cracked and show external decay (Aiello et al. 2020). Tissue discolouration is observed internally (i.e. infection in the xylem) which can result in the detachment of bark and/or roots from the stem (Aiello et al. 2020). In advanced stages of infection, root rot is also observed (Aiello et al. 2020).

An orchard in Italy, consisting of 2200 plants, showed 40% disease incidence (880 trees infected) where 800 plants had died (Aiello et al. 2020). *Pleiocarpon algeriense* was found co-occuring with *Cylindrocladiella peruviana* and the symptoms observed could be the result of the interaction between the two species; however, pathogenicity trials, where plants were inoculated with *P. algeriense* or *C. peruviana* only, still produced visible symptoms and *P. algeriense* was more virulent than *C. peruviana* (Aiello et al. 2020).

Time and conditons to symptom development

In vivo pathogenicity trials in seedlings and young trees (≥3 years old) resulted in similar symptoms as described above (Aiello et al. 2020). Visible necrotic lesions were visible both externally and internally in the bark above the inoculation site and initial symptoms developed within 2 months after inoculation in seedlings (Aiello et al. 2020). After 4 months post inoculation symptoms in young trees were similar to those as seen in the orchard (described above) (Aiello et al. 2020).

In *in vivo* experiments, plants were held in growth chambers with a 12 hour photoperiod and temperatures of 25℃ ± 1℃ (Aiello et al. 2020). Plants were also watered regularly (Aiello et al. 2020).

In culture, no growth of *P. algeriense* was observed at 10℃; colonies started growing at 15℃ and the optimal temperature for growth is 25–30℃ (Aigoun-Mouhous et al. 2019). The minimum temperature requirements for growth of *C. peruviana* in culture is 5℃ with a maximum of 35℃ and an optimum of 25℃ (Crous and Wingfield 1993).

Reproduction and transmission

Given the recent discovery of *P. algeriense*, there is limited information on the reproduction and transmission of this organism. Affected plants produced in nurseries could represent the primary pathway for the transmission of this pathogen (Aiello et al. 2020). Long lived spores such as chlamydospores are rarely seen in *P. algeriense* (Aiello et al. 2020).

Cylindrocladiella peruviana is dispersed through wind and rain splash from the soil surface or by contact with the ground (Department of Agriculture Fisheries and Forestry 2004). Chlamydospores are extensive and are observed in chains in *Cylindrocladiella* species (Crous and Wingfield 1993).

In addition, Aiello et al. (2020) collected symptomatic plant material (sections of basal stem and crown tissue) which were then surface disinfected for 1 min in 1.5% sodium hypotchorlite. Growth of *P.*

algeriense and *C. peruviana* still occurred from surface disinfected material indicating this organism grows internally and would not be affected by surface disinfection (Aiello et al. 2020).

5.19.4 Likelihood of entry

This assessment is made on the basis that *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* are present in the countries where avocado budwood is being imported from. *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* have been reported on avocado in Italy (Aiello et al. 2020). *Cylindrocladiella peruviana* has been recorded on other host species in many other countries (**Error! Reference source not found.**).

Infection by both cause stem and crown rot and infection is more commonly associated with the base of the plant (Aiello et al. 2020). Severe infection can result in the base of the tree detatching from the roots (Aiello et al. 2020). Infections were observed in trees ≥3 years old, suggesting the pathogens may have a latency period (Aiello et al. 2020). *Cylindrocladiella* species are soil-borne fungi and regarded as saprotrophs and pathogens, suggesting they may remain latent in the environment (i.e. soil) (Lombard et al. 2012). The mode of infection of *C. peruviana* and *P. algeriense* are most likely through roots and wounds as dispersal (particularly for *C. peruviana*) is through wind and rain splash from the soil surface (Crous and Wingfield 1993; Department of Agriculture Fisheries and Forestry 2004).

Infection is most likely associated with the base of stems near the surface of the soil; however, *Cylindrocladiella peruviana* has caused root and cutting rot in other host species (e.g. *Acacia mearnsii*, *Eucalyptus* spp. and *Vitis vinifera*) (van Coller et al. 2005) suggesting a possibilility this pathogen could be associated with avocado budwood; however, given the recent observation of the association of *C. peruviana* with avocado, there is no evidence of the association of this pathogen on avocado budwood. It has been demonstrated through artificial inoculations that *Pleiocarpon algeriense* causes infection in grapevine cuttings (Aigoun-Mouhous et al. 2019); however, similar to *C. peruviana*, there is no evidence of infection on avocado cuttings. Therefore, there is high uncertainty regarding the possibility of infection in avocado budwood.

Given the internal infection of both of these organisms, the prescribed surface disinfection of budwood entering PEQ (1% sodium hypochlorite dip for 2 minutes) would reduce surface populations of these fungi but would not reduce internal infections.

Given the arguments and evidence above, that is:

- *Cylindrocladiella peruviana* and *P. algeriense* are likely soilborne (Lombard et al. 2012; Aiello et al. 2017; Aiello et al. 2020)
- latent infections of *C. peruviana* and *P. algeriense* are observed in avocado orchards as disease has only been observed in 3 year old plants (Aiello et al. 2020),
- *Cylindrocladiella peruviana* and *P. algeriense* infect cuttings of other host species (van Coller et al. 2005; Aigoun-Mouhous et al. 2019)
- routine surface disinfection (1% sodium hypochorite for 2 minutes) would not reduce internal infections

the likelihood of *Pleiocarpon algeriense* entering New Zealand associated with *P. americana* budwood is considered to be low, with high uncertainty.

5.19.5 Likelihood of exposure

This assessment is made on the basis that *Cylindrocladiella peruviana* and *Pleiocarpon algeriense* have entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *C. peruviana* and *P. algeriense*.

If avocado plants are infected with *C. peruviana* and/or *P. algeriense* at the time of their release from PEQ they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *C. peruviana* and *P. algeriense* in New Zealand from *P. americana* budwood is considered to be high, with low uncertainty.

5.19.6 Likelihood of establishment

This assessment is made on the basis that *Cylindrocladiella peruviana* and *Pleiocarpon algeriense* have been successfully exposed to a suitable host plant in the New Zealand environment.

Pleiocarpon algeriense has been recorded on *Persea americana* in Italy (Aiello et al. 2020) and on *Vitis vinifera* in Algeria (Aigoun-Mouhous et al. 2019). Algeria has a climate match index ranging between 0.3–0.8 (Phillips et al. 2018). *Cylindrocladiella peruviana* has a wider geographic and host range in comparison to *P. algeriense* (**Error! Reference source not found.**). Both species are found in countries that have a similar climate to the whole of New Zealand, as well as to the Auckland and Northland regions, as indicated by a climate match index of ≥0.7 (Phillips et al. 2018). Climate suitability is further supported by the fact that other *Cylindrocladiella* species have already established in New Zealand (Biota NZ 2022).

Both species infect hosts that are either grown commericially (e.g. grapes, avocado and *Eucalyptus*) or as backyard plants in many home gardens. Therefore, hosts for the establishment of *C. peruviana* and *P. algeriense* are readily available. In addition, *C. peruviana* has commonly been isolated from soils (Table 5-20).

Table 5-20: Geographic and host range of *Cylindrocladiella peruviana*.

Region	Country	CMI*	Host	Reference
Europe	Spain	0.7–0.9	*Vitis vinifera*	Agustí-Brisach et al. (2012)
	Italy	0.7–0.9	*Persea americana*	Aiello et al. (2020)
North America	USA	0.5–0.8	*Vitis vinifera*	Koike et al. (2016)
Asia	Taiwan	0.4–0.8	*Eriobotrya japonica*	Tsai et al. (2012)
	Thailand	0.3–0.5	Soil	van Coller et al. (2005)
	India	0.2–0.5	*Oryza sativa*	van Coller et al. (2005)
	Japan	0.6–0.8	-	Victor et al. (1998)
	Vietnam	0.4–0.5	Soil	Pham et al. (2018)
South America	Peru	0.4–0.8	Ants	van Coller et al. (2005)
	Brazil	0.3–0.9	Ants	Victor et al. (1998)
			Piptadenia sp.	Lombard et al. (2012)
			Psidium guajava	Lombard et al. (2012)
	Not recorded		Soil	van Coller et al. (2005)
Africa	South Africa	0.5–0.9	*Acacia mearnsii*	Victor et al. (1998)
			Vitis vinifera	van Coller et al. (2005)
			Eucalyptus sp.	Victor et al. (1998)
			Leaf litter	Victor et al. (1998)
			Pelargonium sp.	Crous et al. (2019)
Oceania	Australia	0.4–1	Soil	Lombard et al. (2012)

* Climate match index (CMI) ≥ 0.7 indicates countries with a similar climate to the whole of the New Zealand (Phillips et al. 2018).

Given the arguments and evidence above, that is:

- hosts (in the form of cultivated avocados and other cultivated crops) are available for the establishment of *Cylindrocladiella peruviana* and *Pleiocarpon algeriense*.
- suitable climate (Table 5-20) is available for the establishment of *Cylindrocladiella peruviana* and *Pleiocarpon algeriense*

the likelihood of *C. peruviana* and *P. algeriense* establishing in New Zealand is considered high, with low uncertainty.

5.19.7 Impacts in New Zealand:

The co-occurance of *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* in *Persea americana* has been recorded causing 40% disease incidence in orchards with a high proportion of those causing death of the plant (Aiello et al. 2020). Both of these organisms have also been implicated in causing black-foot disease in grapevines (e.g. van Coller et al. 2005; Aigoun-Mouhous et al. 2019).

Other agents of black-foot disease are recorded as present in New Zealand (e.g. *Ilyonectria* spp., *Cylindrocarpon* spp., *Dactylonectria* spp. and *Cylindrocladiella parva*) and are managed through the use of fungicides (Bleach et al. 2013). Black-foot disease is particularly a problem in nurseries and new vineyards (Berlanas et al. 2020) and death is likely in vines less than 10 years old (Bleach et al. 2021). Fungi causing black-foot disease can live as latent pathogens within grapevine and may become pathogenic under specific conditions such as waterlogging, water stress, nutrient deficiency or soil compaction (Berlanas et al. 2020). Diseased vines need to be removed and fungal inoculum removed (Bleach et al. 2021).

Given *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* are soil-borne pathogens and that both can produce long lived chlamydospores, spread of these pathogens can occur through the movement of infected soils (Crous and Wingfield 1993).

Economic impacts

Pleiocarpon algeriense is likely to affect both the avocado and grape industry whereas *Cylindrocladiella peruviana* is likely to affect avocado, grape and *Eucalyptus* spp. plantations (**Error! Reference source not found.**).

In 2020, the export values for avocado was $112.3 million and the domestic value was $50.6 million (Plant & Food Research 2020). The export value for grapes for wine was $1.9 billion (Plant & Food Research 2020). *Eucalyptus* spp. were valued at $671 million in 2018 with an estimated planted area of 27, 298 ha (Radics et al. 2018).

Incidence of these pathogens in vineyards and plantations are lacking; however, given that other fungi that cause black-foot disease are present in New Zealand and have been for awhile (Bleach et al. 2013; Bleach et al. 2021), current management practices are likely to account for disease caused by *P. algeriense* and *C. peruviana*.

Therefore, given these pathogens have the ability to cause up to 40% disease incidence in avocado orchards, economic impacts are likely (Aiello et al. 2020). Furthermore, cost of plant and inoculum removal would also need to be incorporated into the cost of management (Bleach et al. 2021).

Given the arguments and evidence above, that is:

- some species causing black-foot disease in grapevines are already present in New Zealand (Bleach et al. 2021),
- management practices are used in New Zealand for *Cylindrocladiella* spp. and other black-foot disease causing fungi (Bleach et al. 2013; Bleach et al. 2021),

- the host range of *P. algeriense* and *C. peruviana* indicates not only the avocado but also the grape and *Eucalyptus* industry could be affected which together are valued at $2.7 billion (sum of both domestic and export earnings),

the economic impact of *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* to New Zealand is considered to be moderate, with moderate uncertainty.

Environmental impacts

It is unknown if either *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* have known native New Zealand plant hosts. However, *C. peruviana* is known to infect *Acacia mearnsii* which is present throughout New Zealand (NZPCN 2021). *Cylindrocladiella peruviana* is commonly found in the soil and has been associated with ants (species unknown) in South America (**Error! Reference source not found.**). Ants may also act as a mode of dispersal for this fungus.

Given the possibility of *C. peruviana* species living as pathogens or saprotrophs on plants and in the soil (Lombard et al. 2012), this fungus has the potential wide range of environmental impacts. However, a related species *C. parva* is already present in New Zealand (Biota NZ 2022), suggesting impacts would not differentiate drastically. In contrast, *Pleiocarpon algeriense* is newly described (Aigoun-Mouhous et al. 2019) and thus, environmental impacts are unknown.

Given the arguments and evidence above, the impact on the environment from the establishment of *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* in New Zealand is considered to be very low, with moderate uncertainty.

Health impacts

There is no evidence that *Pleiocarpon algeriense* or *Cylindrocladiella peruviana* cause human health impacts. Search terms [*Cylindrocladiella* OR *Pleiocarpon* AND “human health”] were used in Google, Google Scholar and Web of Science with no results.

Given the arguments and evidence above, the health impacts of *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* to New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

Both *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* have the potential to cause death of plants in avocado orchards and vineyards (Aigoun-Mouhous et al. 2019; Aiello et al. 2020). If left unmanaged, there could be a decline in fruits in markets. Avocado, grapes (and wine) are popular and are widely consumed in New Zealand. They are commonly grown in home gardens in some parts of the country.

Given the arguments and evidence above, the social/cultural impacts of *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* to New Zealand is considered to be low, with low uncertainty.

5.19.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is low
- assuming entry, the likelihood of exposure is high
- assuming successful exposure, the likelihood of establishment is high
- the overall impact on the New Zealand economy, environment, human health and society is considered to be moderate

the overall level of assessed risk to New Zealand from *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* on imported *Persea americana* budwood (as in the commodity description) is considered to be moderate, with moderate uncertainty.

5.19.9 Management considerations

Cylindrocladiella peruviana and *Pleiocarpon algeriense* cause stem and crown rot in *Persea americana* (avocado). Infected plants have stunted growth, general wilting and leaf chlorosis; the basal stem and crown of plants become dry, cracked and show external decay (Aiello et al. 2020). Tissue discolouration is observed internally (i.e. infection in the xylem) which can result in the detachment of bark and/or roots from the stem (Aiello et al. 2020). In advanced stages of infection, root rot is also observed (Aiello et al. 2020).

Experimental conditions for symptom expression a 12 hour photoperiod and temperatures of 25°C ± 1°C (Aiello et al. 2020). In the field, stem and crown rots are exacerbated by specific conditions such as waterlogging, water stress, nutrient deficiency or soil compaction (Berlanas et al. 2020).

Infection seems to be common around the base of trees; however, there is evidence these species can infect cuttings in other host species (van Coller et al. 2005; Aigoun-Mouhous et al. 2019). Thus, the possibility for association with avocado budwood can not be ruled out. Infections are within internal tissues of stems (with external symptoms visible) and thus testing of tissue within stems would be necessary.

PCR testing is available for both *Cylindrocladiella peruviana* and *Pleiocarpon algeriense*. To distinguish among organisms within the complex that causes black-foot disease, multiple gene regions are sequenced to better resolve species identification (Aigoun-Mouhous et al. 2019). Gene regions sequenced include the ITS, *tub2*, *tef1*, *his3*and *rpb2* gene regions (Aigoun-Mouhous et al. 2019; Aiello et al. 2020).

5.19.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Both *Pleiocarpon algeriense* and *Cylindrocladiella peruviana* cause wilting of avocado plants as a result of stem and crown rot (Aiello et al. 2020). Infected plants have stunted growth, general wilting and leaf chlorosis; the basal stem and crown of plants become dry, cracked and show external decay (Aiello et al. 2020). Tissue discolouration is observed internally which can result in the detachment of bark and/or roots from the stem (Aiello et al. 2020). In advanced stages of infection, root rot is also observed (Aiello et al. 2020).

- ***What are the known environmental conditions conducive to symptom expression?***

Experimental conditions for symptom expression are 12 hour photoperiod and temperatures of 25°C ± 1°C (Aiello et al. 2020). In the field, stem and crown rots are exacerbated by specific conditions such as waterlogging, water stress, nutrient deficiency or soil compaction (Berlanas et al. 2020).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested?***

Transverse and longitudinal sections from the grafting point, basal part of the crown and middle part between grafting point and basal point have successfully had the pathogens isolated (Aigoun-Mouhous et al. 2019).

- ***What is the optimum season for conducting detection testing?***

There is no optimum season for conducting detection testing as specific environmental stressors (listed above) exacerbate symptom expression.

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5.20 *Podosphaera perseae-americanae*

Podosphaera perseae-americanae is a fungus causing powdery mildew on leaves of *Persea americana* (avocado) plants.

5.20.1 Taxonomic description

Scientific name: *Podosphaera perseae-americanae* Siahaan & S. Takam.

Order/Family: Erysiphales/Erysiphaceae

Taxonomic notes: Genus *Podosphaera* consists of two subsections *Podosphaera* sect. *Podosphaera* and *Podosphaera* sect. *Sphaerotheca*. However, based on DNA sequences, *Podosphaera perseae-americanae* seems to form an independent lineage, suggesting that this fungus may belong to an undescribed genus. Due to lacking sexual morph description, this fungus was tentatively assigned to genus *Podosphaera* (Siahaan et al. 2016). Powdery mildew on avocado can also be caused by *Pseudoidium perseae-americanae* (synonym *Oidium perseae-americanae*), but these two fungi are distinguishable by morphological characteristics, where *Podosphaera* produces conidia (asexual spores) in chains (catenate-type) while *Pseudoidium* (syn. *Oidium*) produces non-catenate conidia (Siahaan et al. 2016).

5.20.2 Hazard identification

Podosphaera perseae-americanae is not known to be present in New Zealand as it is not listed in Biota NZ (2022) or PPIN (2021). The regulatory status of *Podosphaera perseae-americanae* has not been assessed (ONZPR 2021).

Podosphaera perseae-americanae has the potential to establish (and spread) in New Zealand because:

- *Persea americana* (avocado) is a known host (Siahaan et al. 2016) that is widely grown in New Zealand, in home gardens and commercial production, mainly in the north of the North Island.

Podosphaera perseae-americanae has the potential to cause harm to New Zealand because:

- *Podosphaera perseae-americanae* causes powdery mildew on avocado (Siahaan et al. 2016).

Podosphaera perseae-americanae is associated with *Persea americana* budwood because:

- *Persea americana* is a known host of *Podosphaera perseae-americanae* (Siahaan et al. 2016);
- *Podosphaera perseae-americanae* was isolated from young leaves of *Persea americana.* Powdery mildew fungi stay dormant in buds of infected plants, as dormant mycelia (Glawe 2008). Therefore, mycelium may be present on leafless budwood produced from an infected mother plant.

Given the arguments and evidence above, *Podosphaera perseae-americanae* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.20.3 Risk assessment

Biology and epidemiology of Podosphaera perseae-americanae

Host range

The host range of *Podosphaera perseae-americanae* is restricted to *Persea americana* as it is so far the only host plant and locality recorded (Siahaan et al. 2016). Most other *Podosphaera* species parasitise woody plants from *Rosaceae* family, with the exception of species in *Podosphaera* sect. *Sphaerotheca* subsect. *Magnicellulatae* with herbaceous host plants scattered among 40 plant families that do not include the *Rosaceae*. Some species in the genus *Podosphaera* are plurivorous (with multiple host plants) such as *Podosphaera xanthii* reported from plants in the families *Asteraceae*, *Fabaceae*, *Cucurbitaceae* and *Solanaceae* (Pérez-García et al. 2009). Since *Podosphaera perseae-americanae* was only reported from *Persea americana* in one country, the known host range of *Podosphaera perseae-americanae* is restricted to *P. americana*, although host plants may include other members of the Lauraceae family. The possibility that this species may not be in the genus *Podosphaera* (Siahaan et al. 2016) further introduces uncertainty associated with the assessments.

Symptoms

Powdery mildews in general are characterised by a floury layer of mycelium with conidia (asexual spores) which covers affected leaves, buds, shoots and fruits. Affected developing shoots and leaves may remain short, deformed and lesions and chlorotic spots can develop. *Podosphaera perseae-americanae* causes powdery mildew disease on *Persea americana*, although no detailed description of the symptoms is provided, the depicted symptoms include thick powdery cover on the underside of slightly discoloured and deformed young leaves (Siahaan et al. 2016). Generally, powdery mildew symptoms can become severe in warm, humid, shaded areas such as nurseries. Rain can wash conidia from plant surfaces before they germinate, thus reducing symptom expression (Zentmyer 1953; Glawe 2008).

Life cycle and transmission

Podosphaera perseae-americanae was isolated from young avocado leaves and is likely to only affect above ground tissues similarly to other powdery mildews. Although, some species of the genus *Podosphaera* can be endophytic (e.g. *Podosphaera tridactyla* in buds and leaves of *Eucalyptus* (Miguel et al. 2019)), mycelium of *Podosphaera perseae-americanae* has been described as ectophytic – affecting only the host cells on the surface (Siahaan et al. 2016). *Podosphaera perseae-americanae* was isolated only in its asexual stage, producing large amounts of asexual spores (conidia) on avocado leaves. The conidia of *Podosphaera perseae-americanae* are likely airborne and transmitted by wind or human activity as is the case of other *Podosphaera* species and powdery mildew in general (Glawe 2008; Numminen and Laine 2020). The complete life cycle of *Podosphaera perseae-americanae* could involve an asexual stage as well as a sexual stage, as is the case for other powdery mildews (Glawe 2008). Sexual spores may be released from dormant chasmothecia (sexual ascocarps – fruiting bodies) with spring rains. Powdery mildews are obligate parasites of vascular plants, that means they can survive unfavourable conditions as dormant cleistothecia, endophytes or as dormant hyphae in buds of their host. The latter is most likely in case of *Podosphaera perseae-americanae* as the ascospores produced in chasmothecia have not been observed in *Podosphaera perseae-americanae* so far and mycelium of *Podosphaera perseae-americanae* has been described as ectophytic (Siahaan et al. 2016).

5.20.4 Likelihood of entry

In plants, powdery mildews can colonise all metabolically active above ground plant parts (i.e. stems, twigs, leaves, flowers, buds, and fruit) as epiphytes or endophytically (Glawe 2008). *Podosphaera perseae-americanae* was isolated from young leaves and it is likely to colonise dormant buds similar to other powdery mildews (Glawe 2008). Therefore, *Podosphaera perseae-americanae* can be associated with avocado budwood.

Avocado budwood is dipped in 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility, which is likely to kill surface infections, but may not penetrate into buds where dormant mycelium might be present.

Although there is not much information specifically related to *Podosphaera perseae-americanae*, based on related species of powdery mildew, conidial production typically begins within several days of infecting the host (Glawe 2008). Although some *Podosphaera* species are known to be endophytic, such as *Podosphaera tridactyla* in the leaves of hybrid *Eucalyptus grandis* X *Eucalyptus urophyla (Miguel et al. 2019)*, the only record of *Podosphaera perseae-americanae* describes it as an epiphyte with ectophytic mycelium (Siahaan et al. 2016).

Avocado plants in post entry quarantine L3A must be inspected at least twice per week during periods of active growth and once per week during dormancy (MPI 2021). *Podosphaera perseae-americanae* produces cylindrical spores with a size of 24.5-33.5 x 13.5-20 µm. In the event of rapid sporulation, typical of powdery mildews, airborne spores could escape containment facilities of L3A PEQ through the vents which are screened with stainless steel insect-proof mesh with a maximum opening size of 0.04 mm (40 µm) (MPI 2021). Powdery mildew spores are typically airborne and spread better in low humidity, usually over short distances, although some powdery mildew species other than *Podosphaera* have been recorded to spread over hundreds of kilometres (Glawe 2008). Due to limited information related to *Podosphaera perseae-americanae* the uncertainty associated with this assessment is high.

Given the arguments and evidence above, that is:

- *Podosphaera perseae-americanae* is likely to enter PEQ in dormant buds.
- *Podosphaera perseae-americanae* is not likely to survive as an epiphyte on avocado budwood without causing symptoms throughout the PEQ period. In other words, the likelihood of *Podosphaera perseae-americanae* entering New Zealand on avocado plants from PEQ undetected is negligible with moderate uncertainty.
- If symptoms develop, rapid production of airborne spores may not be detected early enough for adequate containment and measures in PEQ L3A may not prevent spores escaping containment facilities.

the likelihood of *Podosphaera perseae-americanae* entering New Zealand associated with *Persea americana* budwood is considered to be low, with moderate uncertainty.

5.20.5 Likelihood of exposure

Since there is a negligible risk of avocado plants infected with *Podosphaera perseae-americanae* at the time of their release from Post Entry Quarantine, this assessment is made on the basis that *Podosphaera perseae-americanae* has entered New Zealand undetected as airborne spores.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. If suitable hosts were present near the post-entry quarantine facilities, for example, if the facility was located near avocado orchards, private gardens with avocado trees or other native or naturalised plants from the Lauraceae family, the fungus could be exposed to suitable host material. However, the amount of inoculum likely to escape is considered to be small, and the conidia would have to land on a suitable host under suitable conditions to germinate and cause disease. Due to limited information related to *Podosphaera perseae-americanae* the uncertainty associated with this assessment is moderate.

Given the arguments and evidence above, the likelihood of exposure of *Podosphaera perseae-americanae* in New Zealand from *Persea americana* budwood is considered low, with moderate uncertainty.

5.20.6 Likelihood of establishment

This assessment is made on the basis that *Podosphaera perseae-americanae* has been successfully exposed to a suitable host plant in the New Zealand environment.

If the host range of the pathogen is only *Persea americana* then hosts in the form of cultivated avocados will be available for their establishment in Aotearoa New Zealand and especially in the North Island (see section 2.3). If the host range includes other members of the family Lauraceae then the number of potential host species available for the establishment of this fungus is greatly increased – there are five native and four introduced species (in addition to *Persea americana*) from the Lauraceae family found in New Zealand (NZPCN 2021).

Podosphaera perseae-americanae is reported from North Sumatra and West Java in Indonesia (Siahaan et al. 2016). These two regions have a more tropical climate than the whole of New Zealand, or Auckland and Northland regions, as indicated by a climate match index range of 0.3 – 0.6 (Phillips et al. 2018). Therefore, climate consistently suitable for the establishment of *Podosphaera perseae-americanae* is not readily available. However, powdery mildews generally require low humidity to spread (Glawe 2008), for example *Podosphaera xanthii* has spread in Korea with higher temperatures and low moisture (Lee 2012). *Podosphaera perseae-americanae* is so far reported only from two regions in Indonesia, which may be due the fact that the potential to establish has not yet been realised elsewhere. Due to limited information related to *Podosphaera perseae-americanae* the uncertainty associated with this assessment is high.

Given the arguments and evidence above, that is:

- hosts (in the form of cultivated avocados) amd potential hosts(in the form of native and introduced members of the Lauraceae family) are available for the establishment of *Podosphaera perseae-americanae,* and
- climate suitable for the establishment of the *Podosphaera perseae-americanae* is likely not readily available,

the likelihood of *Podosphaera perseae-americanae* establishing in New Zealand is considered low, with moderate uncertainty.

5.20.7 Impacts in New Zealand

Podosphaera perseae-americanae is a recently described species causing powdery mildew on avocado (Siahaan et al. 2016). Fungi related to *Podosphaera perseae-americanae*, causing powdery mildews, can produce airborne conidia in large amounts, within a short space of time (3–7 days post infection under disease-promoting conditions). This can cause rapid spread of the disease.

Powdery mildew typically disperse by airborne conidia over over short distances, although some powdery mildew species other than *Podosphaera* have been recorded to spread over hundreds of kilometres (Glawe 2008). Rain can wash conidia from plant surfaces before they germinate, thus reducing symptom expression or preventing spread (Zentmyer 1953; Glawe 2008). *Podosphaera perseae-americanae* is so far reported to only infect avocado, so it's spread is highly likely to be limited to avocado growing regions due to host availability.

Economic impacts

Because avocados are so far the only known host of *Podosphaera perseae-americanae* which are commercially produced in New Zealand, the assessment of economic impacts is limited to the avocado industry.

There is no specific information on the economic impacts of *Podosphaera perseae-americanae* currently available. However, powdery mildew infection may impact yields, as is the case with species in the genus *Podosphaera*, although yield losses may vary widely between species. For example, *Podosphaera macularis* (hop powdery mildew) caused complete loss of 810 hectares of hops in Washington, USA worth 10,000,000 USD (Mahaffee et al. 2003), whereas *Podosphaera aphanis* on strawberries may not even cause direct yield losses if the powdery mildew symptoms occur after fruit harvest (Carisse and Bouchard 2010).

If there were an outbreak of *Podosphaera perseae-americanae* in one of the major avocado-producing regions in New Zealand, the outbreak might not be contained to that region. Spread within and between regions could be quite rapid, especially if dry, windy conditions conductive for conidia dispersion were followed by rain or a humid period that would be favourable for germination of the conidia

However, there is a wide range of control options for powdery mildews which include synthetic fungicides, wettable sulphur or copper sprays (Mahaffee et al. 2003; Mburu et al. 2014), surfactants (Hislop and Clifford 1976) or cultural practices (Mburu et al. 2014). Although there is no specific information on control of *Podosphaera perseae-americanae*, control measures used against other powdery mildews are likely applicable to control *Podosphaera perseae-americanae*.

Given the arguments and evidence above, that is:

- avocados are the only known host of *Podosphaera perseae-americanae* commercially produced in New Zealand,
- infection of avocado trees with *Podosphaera perseae-americanae* could cause yield losses
- an outbreak would likely spread quickly within and between regions

the economic impact of *Podosphaera perseae*-americanae to New Zealand is considered to be low, with high uncertainty.

Environmental impacts

Podosphaera species have a wide range of hosts including cultivated and wild plants, and some species may cause significant disease in wild plants or natural environments (Takamatsu et al. 2010). *Podosphaera perseae-americanae* was isolated from avocado, but there is no information available to confidently assume whether *Podosphaera perseae-americanae* is a specialist only parasitising *Persea americana* or whether it could be plurivorous (parasitising multiple host plants). The host range of *Podosphaera perseae-americanae* may include other species of the family Lauraceae, which include five native Lauraceae species (*Beilschmiedia taraire*, *Beilschmiedia tawa*, *Cassytha paniculate*, *Cassytha pubescens* and *Litsea calicaris*) found in Aotearoa New Zealand (NZPCN 2021). Some of these native species are endemic, however none of these are threatened Aotearoa New Zealand (NZPCN 2021).

Given the arguments and evidence above, the impact on the environment from the establishment of *Podosphaera perseae-americanae*in New Zealand is considered to be low, with high uncertainty.

Human health impacts

There are no known human health impacts associated with *Podosphaera perseae-americanae*.

Given the arguments and evidence above, the impact on human health from the establishment of *Podosphaera perseae-americanae* in New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

An outbreak of *Podosphaera perseae-americanae* may affect avocado plants grown in home gardens. However, as there is likely a wide range of control options for powdery mildews, this impact is likely to be very low. If native Lauraceae plants were susceptible to the disease, an outbreak could affect sociocultural values especially since *Beilschmiedia* spp. and *Litsea calicaris* are only found in Aotearoa New Zealand (NZPCN 2021).

Given the arguments and evidence above, the sociocultural impact of *Podosphaera perseae-americanae* to New Zealand is considered to be very low, with moderate uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, human health and society is considered to be low, with moderate uncertainty.

5.20.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is low,
- assuming entry, the likelihood of exposure is low,
- assuming successful exposure, the likelihood of establishment is moderate,
- the overall impact on the New Zealand economy, environment, human health and society is considered to be low,

the overall level of assessed risk to New Zealand from *Podosphaera perseae-americanae*on imported *Persea americana* budwood (as in the commodity description) is considered to be very low, with moderate uncertainty.

5.20.9 Management considerations

The limited information on *Podosphaera perseae-americanae* include thick powdery cover on the underside of slightly discoloured and deformed young leaves (Siahaan et al. 2016). Symptoms similar to those described from other powdery mildews could develop after infection by *Podosphaera perseae-americanae*; conidia can be produced quickly and in large amounts (3–7 days post infection under disease-promoting conditions). White mycelium can cover juvenile parts (inflorescence and leaves) causing damage and even necrosis. Old infection sites appear as conspicuous chlorotic areas with net-like markings (McMillan 1976). The pest might not exhibit symptoms if it is present as dormant hyphae in buds of the host plant.

Symptomatic *Podosphaera perseae-americanae* was isolated from avocado plants in Indonesia in September 2013 and January 2015, where mean daily temperatures were greater than 26^{o} C and monthly rainfall was between 50 and 300 mm ("Monthly Climatic Data for the World September 2013" 2013; "Monthly Climatic Data for the World January 2015" 2015). Generally, powdery mildew symptoms can become severe in warm, humid, shaded areas such as nurseries. Rain can wash conidia from plant surfaces before they germinate, thus reducing the conductivity to symptom expression (Zentmyer 1953; Glawe 2008). However, powdery mildews are usually synchronized with host life cycles, thus the conditions might depend more on the etiology of the host (Glawe 2008), such as development of young shoots and leaves.

Powdery mildews can survive as dormant hyphae in buds of their host (Glawe 2008). In case dormant *Podosphaera perseae-americanae* mycelia is unknowingly imported into PEQ, young leaves and shoots should be inspected regularly for developing symptoms. Molecular diagnostics focusing on genetic marker regions 28S rRNA and ITS should be sufficient to differentiate this pathogen from other powdery mildews or allow to test asymptomatic plants (Siahaan et al. 2016).

Symptoms of *Podosphaera perseae-americana* were described in Indonesia which is a tropical country with relatively stable high humidity and temperature. The symptoms were recorded in September and January.

5.20.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

The limited information on *Podosphaera perseae-americanae* include thick powdery cover on the underside of slightly discoloured and deformed young leaves (Siahaan et al. 2016). Symptoms similar to those described from other powdery mildews could develop after infection by *Podosphaera perseae-americanae*; conidia can be produced quickly and in large amounts (3–7 days post infection under disease-promoting conditions). White mycelium can cover juvenile parts (inflorescence and leaves) causing damage and even necrosis. Old infection sites could appear as conspicuous chlorotic areas with net-like markings (McMillan 1976). The pest might not exhibit symptoms if it is present as dormant hyphae in buds of the host plant.

- ***What are the known environmental conditions conducive to symptom expression?***

Symptomatic *Podosphaera perseae-americanae* was isolated from avocado plants in Indonesia in September 2013 and January 2015, where mean daily temperatures were greater than 26° C and monthly rainfall was between 50 and 300 mm ("Monthly Climatic Data for the World September 2013" 2013; "Monthly Climatic Data for the World January 2015" 2015). Generally, powdery mildew symptoms can become severe in warm, humid, shaded areas such as nurseries. Rain can wash conidia from plant surfaces before they germinate, thus reducing symptom expression (Zentmyer 1953; Glawe 2008). However, powdery mildews are usually synchronized with host life cycles, thus the conditions might depend more on the etiology of the host (Glawe 2008), such as development of young shoots and leaves.

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Powdery mildews can survive as dormant hyphae in buds of their host (Glawe 2008). In case dormant *Podosphaera perseae-americanae* mycelia is unknowingly imported into PEQ, young leaves and shoots should be inspected regularly for developing symptoms.

- ***What is the optimum season for conducting detection testing?***

Symptoms of *Podosphaera perseae-americana* were described in Indonesia which is a tropical country with relatively stable high humidity and temperature. The symptoms were described in September and January. This suggests that summer would be the optimum season for detection testing in New Zealand.

5.20.11 References

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5.21 *Pseudocercospora purpurea*

Pseudocercospora purpurea is a fungal plant pathogen causing Pseudocercospora spot disease resulting in leaf, stem, and fruit spot symptoms in *Persea* spp. including avocado (*P. americana*). The disease is a serious pre-harvest disease affecting all cultivars of avocado in warm, humid, and rainy climates.

5.21.1 Taxonomic description

Scientific name: *Pseudocercospora purpurea* (Cooke) Deighton 1976

Order/Family: Capnodiales/Mycosphaerellaceae

Other names include: *Cercospora purpurea*, Cercospora/Pseudocercospora spot; black spot; leaf spot: avocado; spot blotch of avocado; Cercospora spot blotch; mancha angular de la hoja del aguacate (Spanish).

Taxonomic notes: A review of the previous scientific literature relating to the fungus was recently undertaken by Kallideen (2020) and mentions the historic name for the fungus, *Cercospora purpurea* Cooke (1878) (stat. anam.) Kallideen (2020) referred to a previous taxonomic revision of *Cercospora* and allied genera, and found the fungus was renamed *Pseudocercospora purpurea*.

5.21.2 Hazard identification

Pseudocercospora purpurea is not known to be present in New Zealand:

- *Pseudocercospora purpurea* is not listed in PPIN (2021)
- *Pseudocercospora purpurea* is recorded as ‘recorded in error’ in Biota NZ (2022). The sole NZ collection has been redetermined as an unidentified coelomycete.
- *Pseudocercospora purpurea* is a regulated pest in New Zealand, has an unwanted status but is not notifiable (ONZPR 2021).

Pseudocercospora purpurea has the potential to establish (and spread) in New Zealand because:

- The reported host of *Pseudocercospora purpurea* is avocado (*Persea americana*) and other *Persea* spp. (California Department of Food & Agriculture 2016).
- *Pseudocercospora purpurea* is found in subtropical and tropical regions (California Department of Food & Agriculture 2016), some of which have a climate match index (CMI) ≥ 0.7 (Table 5-21) indicating a similar climate to New Zealand (Phillips et al. 2018).

Pseudocercospora purpurea has the potential to cause harm to New Zealand because:

- It causes disease symptoms on leaves, stems, and fruit (California Department of Food & Agriculture 2016).
- Avocados are an important commercial crop in New Zealand. Export sales in 2020 (fresh and oil) earned NZ$122 million. Domestic sales earned NZ$ 50.6 million in 2019/20 (Plant & Food Research 2020).
- *Pseudocercospora purpurea* has been reported to cause significant losses (48%) in avocado production by decreasing fruit quality and making fruits unacceptable for export (Reina-Noreña et al. 2015; Rodríguez-Polanco et al. 2020).

Pseudocercospora purpurea is associated with ***Persea americana*** **budwood** because:

- *Persea americana* is a commonly reported host of *Pseudocercospora purpurea* (EPPO 2021).

- *Pseudocercospora purpurea* occurs in association with leaves, stems, and fruit (Kallideen 2020; USDA 2021). This disease can infect previously uninjured leaves, young stems, and fruit (McMillan 1976).
- *Pseudocercospora purpurea* is known to infect all commercial cultivars of avocado.

Given the arguments and evidence above, *Pseudocercospora purpurea* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

5.21.3 Risk assessment

Biology and epidemiology of Pseudocercospora purpurea

Host range

Pseudocercospora purpurea has a limited known range of plant hosts. All are *Persea* spp., in the family Lauraceae, namely, *P. americana* (syn. *P. gratissima*, avocado), *P. borbonia* (redbay), *P. drymifolia* (Mexican avocado) and *P. palustris* (swamp bay) (California Department of Food & Agriculture 2016).

Host susceptibilities differ between *Persea americana* cultivars. Late maturing avocado cultivars are more susceptible as they have longer period during which they can be infected (Menge and Ploetz 2003). For example, Fuerte and Ryan cultivars were found to be considerably more susceptible to *P. purpurea* than Edranol and Hass (Darvas and Kotze 1987). Hass avocados are known to be less susceptible to *P. purpurea* (Willis and Mavuso 2007). In Florida, Waldin, Booth 7 & 8, Taylor, Linda, Lula, Nabal, Trapp and Wagner are the most susceptible while Collinson, Fuchsia and Pollock are moderately susceptible (Darvas 1982). Cultivars may also be considered less susceptible for several seasons but later become severely infected after P. purpurea becomes firmly established in trees (Stevens and Piper 1941). If exposed to conidia early in the critical period (when the climate is wet and warm) then fruit (such as later maturing fruit) have significantly more *P. purpurea* spots than fruit only exposed later (Darvas and Kotzé 1979). Avocado fruit exposed to natural infection early in the critical period (when the climate is wet and warm) develop significantly more *p. purpurea* spots than fruit exposed later

All commercial cultivars of *P. americana* are affected (Everett and Siebert 2018). However, there has been studies undertaken in Westfalia, South Africa to develop new disease resistant cultivars. Cultivars with a natural tolerance or resistance to *P. purpurea* have been identified but none have succeeded to commercial production due to other characteristics which were not desirable (Bruwer et al. 2015). As different cultivars vary in their susceptibility to *P. purpurea* in different geographical regions it has been suggested by Menge and Ploetz (2003) that there may be different physiological races of this pathogen.

Geographical distribution

Pseudocercospora purpurea is widely spread in subtropical and tropical countries (Table 5-21). The fungus is widely reported in avocado growing countries and is considered one of the most common disease problems.

Table 5-21: Geographical distribution of Pseudocercospora purpurea (Dann et al. 2013; CABI 2021) and climate similarity (CMI) to New Zealand.

Region	**Country**	**State/Province**	**CMI***
Asia	India	Sikkim	0.6-**0.7**
	Japan		0.6-**0.8**
	Philippines		0.3-0.4
Africa	Cameroon		0.4-0.5
	Congo Democratic Republic		0.5-**0.8**
	Cote d'Ivoire		0.5
	Guinea		0.4-0.5
	Kenya		0.4-**0.8**
	South Africa		**0.5-0.9**
North America	USA	Florida	0.5-**0.7**
		Georgia, Mississippi	0.7-**0.8**
	Bermuda		Not determined
	Mexico		0.4-**0.8**
Central America & Caribbean	Dominica		Not determined
	El Salvador		0.4-0.5
	Honduras		0.3-0.6
	Jamaica		0.5-0.6
	Nicaragua		0.4-0.5
	Panama		0.3-0.5
	Puerto Rico		0.5-0.6
	Trinidad and Tobago		0.5
	U.S. Virgin Islands		Not determined
South America	Colombia	Northern Tolima	0.3-**0.8** 0.5-**0.8**
	Argentina		0.6-**0.9**
	Bolivia		0.4-**0.8**
	Brazil	Sao Paulo	0.3-**0.9** **0.7**
	Chile		0.5-**0.9**
	Guyana		0.4-0.5
	Peru		0.4-**0.8**
	Venezuela		0.3-**0.7**
Oceania	Palau	Belau	Not determined
	Australia	Queensland	0.4-**0.8**
Europe	Spain		0.4**-0.8**

Note: Dann et al. (2013) lists West Indies, Atherton (Tablelands), Queensland, Australia, and Spain. CABI (2021) does not list Spain.

***(Phillips et al. 2018)**

Symptoms

Pseudocercospora purpurea can infect the leaves, stem, and fruit of *Persea americana* at any time during the growing season (Schoeman and Kallideen 2018). On leaves, the fungus causes small lesions (1-6mm) that are angular, purple-purplish brown, and look like flecks, or spots, near the leaf margin (Perez-Jimenez 2008; California Department of Food & Agriculture 2016; Kallideen 2020). The angular appearance of the lesions is highly diagnostic for the disease (Marais 2004). After the appearance of lesions, chlorotic halos become visible around older spots (Schoeman and Kallideen 2018). Eventually, lesions are seen on both leaf surfaces (Schoeman and Kallideen 2018). The infection may progress up the stems (Perez-Jimenez 2008). The pathogen survives on old lesions. In high humidity conditions, sporulation causes a grey, felty mycelium growth in the centre of lesion. Large areas of infection may appear as necrotic tissue. Leaves curl, deform, and may fall of the plant (California Department of Food & Agriculture 2016). On fruit stems and green twigs, lesions (2 – 10 mm in diameter) may become dark brown to black (Schoeman and Kallideen 2018). These infections may precede those on fruit by several weeks. Lesions producing spores being present on stems long before the fruit (Stevens and Piper 1941).

Reproduction and transmission

Under high humidity and warm environmental conditions infection is mainly through asexual spores (conidia) which grow on specialised stalks called conidiophores and develop in clusters onto the surface of the leaves, stems (including twigs and fruit stems) and fruit (Schoeman and Kallideen 2018).

Conidia are present year-round when moisture (such as rainfall or heavy dew) and humidity is suitable (Kallideen 2020) or the pathogen can overwinter as mycelium (stromata) in old, infected leaves (Stevens and Piper 1941; Marais 2004; California Department of Food & Agriculture 2016). The conidia are carried by wind, rain, insects, and water (Menge and Ploetz 2003). Spread can also occur through contaminated orchard equipment and tools (Kallideen 2020). The pathogen penetrates host tissue either directly (thru uninjured leaves, stems, or fruit) or through wounds (McMillan 1976; California Department of Food & Agriculture 2016).

Darvas (1982) determined the critical infection period for *P. purpurea* on avocado fruit. The critical infection period is the time or latent phase which must elapse between infection and symptom development (Darvas 1982). During his experiment he enclosed Fuerte fruit, on avocado trees in bags. Artificial inoculation of Fuerte fruit with *P. purpurea* was undertaken and these were then reisolated from spots and symptoms analysed. He also used spore traps to determine daily spore numbers in the environment against weather data. The findings determined that in South Africa (which has a similar climate to New Zealand with a CMI of 0.5-0.9; Phillips et al. 2018), *Pseudocercospora purpurea* remains latent for 3 months (Darvas 1982; Menge and Ploetz 2003).

Environmental conditions for infection

Warm, wet, and humid conditions are the environmental conditions most suited to infection. Spore release is highest at 18℃ and declines as temperatures increase to 25℃ (Darvas 1982). No infection occurs below 15℃ (Kallideen 2020).

Significant numbers of spores are produced with the onset of rainy, warm temperatures. The most important environmental parameter is rainfall which has a significant influence on production of *P. purpurea* conidia. Low temperature however will supress spore production or release despite substantial rainfall (Darvas and Kotzé 1979). When comparing temperature, rainfall and relative humidity in South Africa in October, Darvas and Kotzé (1979) found fewer conidia were seen as temperatures decreased from 20.8℃ to 19.9℃ even with rainfall increasing from 7.5mm to 31.3mm.

Relative humidity and leaf wetness are not important in conidia release but can influence pathogen development (Kallideen 2020). There is a positive correlation of symptom development with increasing humidity up to at least 84% (Darvas 1982).

In a laboratory setting most of the fresh *P. purpurea* isolates produced conidia on artificial media when kept continuously under near-ultra-violet light (Darvas and Kotze 1987). Sporulation began ca

10 days after inoculation and a fair amount of conidia was produced for about another 10 days, after which the fungus became sterile. These cultures regained the ability to sporulate if transferred to sterile, freshly cut, avocado pieces, but again only for a limited period (Darvas and Kotze 1987). The implications of this for avocado budwood may be that under ideal conditions conidia can continue to be produced and may resporulate on freshly cut avocado plants.

Spread

Host tissue is penetrated by the fungus through uninjured leaves, stems or fruit or thorough wounds (McMillan 1976; California Department of Food & Agriculture 2016; Schoeman and Kallideen 2018).

The fungus then develops small lesions which appear first on the underside of infected leaves and increase abundantly to eventually cover both sides of infected leaves. The infection may progress along the stem and cause defoliation of the tree (Perez-Jimenez 2008). New shoot tissues are infected wherever this disease occurs (California Department of Food & Agriculture 2016). The disease can infect young stems (McMillan 1976).

Movement of spore-infected fruit and/or plant material can result in rapid spread of the disease over long distances to new areas (Schoeman and Kallideen 2018).

Spores are produced and spread by nursery stock plants, rain and irrigation water, infected leaves, fruit, and insects (California Department of Food & Agriculture 2016). Contaminated tools, machinery and equipment can also spread the fungus to new sites (Everett and Siebert 2018; Kallideen 2020). Spores easily detach and are wind dispersed over long distances (Darvas 1982).

Factors influencing symptom expression (environmental)

Factors influencing symptom expression include:

- Availability of conida leading to high risk of infection, in addition to favourable weather conditions for infection.
- Lapse between infection and symptom development due to latent period or interval.
- Increasing length of exposure time increases severity of symptoms. Susceptibility to *P. purpurea* differs between different cultivars (Kallideen 2020). Avocado exposed earlier in the season have more infection than those later in the season (Schoeman and Kallideen 2018). Late maturing cultivars are more susceptible due to longer time they can be infected (Menge and Ploetz 2003).
- As lesions age the infectious tissue decreases (Everett and Siebert 2018; Rodríguez-Polanco et al. 2020).
- The pathogen can be asymptomatic and overwinter as mycelium in old, infected leaves or shoots (Mavuso et al. 2015).

5.21.4 Likelihood of entry

Pseudocercospora purpurea is a common pathogen of avocado with a wide distribution in most regions around the world and is considered one of the most serious pre-harvest diseases in many avocado growing countries (Darvas 1977).

The disease affects *Persea* spp., in the family Lauraceae, namely, *P. americana* (syn. *P. gratissima*, avocado), along with all cultivars of avocado (California Department of Food & Agriculture 2016). Symptoms are visible on leaves, fruit, and stems during all growth stages (Schoeman and Kallideen 2018) and is as a result associated with budwood. In South Africa, the pathogen is latent on Fuerte avocados for at least 3 months after penetration (Darvas and Kotzé 1979). Therefore, inspection by the NPPO prior to export and on arrival might not detect non-visible infection.

Routine disinfection of the imported budwood on arrival in PEQ with 1% sodium hypochlorite for 2 minutes would reduce surface spore populations. Environmental conditions (assuming warm temperature and high humidity are met) within the PEQ facility are likely to cause symptom expression. Moist conditions such as that provided by misting could provide optimal conditions given that rainfall is considered the most important factor in symptom expression in the field.

Although P. purpurea can be latent for 3 months on imported budwood, given the length of time in the PEQ facility (minimum of 12 months) the pathogen will eventually be symptomatic and detectable by PEQ inspection.

Given the arguments and evidence above, that is:

- Wide distribution in many regions of the world where it is considered one of the most serious pre-harvest diseases in many avocado growing countries.
- Infection is associated with budwood.
- pathogen is latent for at least 3 months after penetration.
- Inspection by NPPO prior to export and on arrival will not detect non-visible infection. However, disinfection will reduce surface spore populations on imported budwood on arrival in PEQ. Optimisation of environmental conditions (temperature, humidity, misting) within the PEQ facility will maximise symptom expression on budwood.
- *Persea* nursery stock must be held in an approved Level 3A greenhouse facility and grown for 12 months providing enough time for any *P. purpurea* infection to become symptomatic and detected by inspection.

the likelihood of *P. purpurea* entering New Zealand associated with *P. americana* budwood is considered to be negligible, with low uncertainty.

5.21.5 Overall level of assessed risk to New Zealand

Based on the assessment of likelihood above, that:

- the likelihood of *P. purpurea* entering New Zealand associated with *Persea americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ) is considered to be negligible with very low uncertainty;
- therefore, continuing the assessment was considered unnecessary; and

the overall level of risk to New Zealand from *P. purpurea* on imported *Persea americana* budwood (as in the commodity description) is considered to be negligible, with moderate uncertainty.

5.21.6 Management considerations

Pseudocercospora purpurea can infect leaves, stems, and fruits (Benson 2018). The pathogen can remain dormant (as mycelium (stromata) in old, infected leaves or shoots for about three months following penetration (Kallideen 2020). Once optimal environmental conditions (warm (over 18℃), humid (up to 84%), and wet climate) are available conidia will be released and initiate infection (Stevens and Piper 1941; California Department of Food & Agriculture 2016; Kallideen 2020). The most important environmental parameter is moisture which has a significant influence on production of *P. purpurea* conidia. Upon landing on the surface of a plant host, moisture is required for conidia to germinate and penetrate the host (Darvas 1982). Once infected under optimal environmental conditions and symptomatic the fungus reproduces abundantly. Leaves become the primary source of continuing disease (Stevens and Piper 1941).

The latent period appears to be longer than 3 months in South Africa on Fuerte avocados according to Darvas and Kotzé (1979). However, no study mentions the maximum period of latency. Darvas and Kotzé (1979) did however record only a few conidia were seen in late October even with substantial rain due to the supressing effects of low temperatures on spore production. Spore traps recorded significant numbers of conidia beginning in November. Darvas (1982) recorded no conidia in April. Given South Africa has a similar climate to New Zealand, the period in PEQ for 12 months will allow time to cover any latency period which for this pathogen is around 3 months.

The fungus is isolated from fresh symptomatic tissue and inoculated on PDA (potato dextrose agar) (Kallideen 2020). The fungus is difficult to isolate initially, but once isolated it grows readily on ordinary nutrient media, producing tufted grey leathery growth which later becomes brown or blackish-brown (Marais 2004; Perez-Jimenez 2008).

As the lesions age the infectious tissue decreases making spores more difficult to isolate and eventually no longer produces spores (Rodríguez-Polanco et al. 2020). It can be difficult to isolate from older lesions as other fungi can outcompete *P. purpurea* such as *Colletotrichum gloesporioides* which grows faster (Kallideen 2020). Surface disinfection of plant material is crucial to eliminating other fungi (Menge and Ploetz 2003).

In a laboratory setting, most of the fresh *P. purpurea* isolates produced conidia on artificial media when kept continuously under near-ultra-violet light. Sporulation begins 10 days after inoculation and a fair amount of conidia was produced for about another 10 days, after which the fungus became sterile. These cultures regained the ability to sporulate if transferred to sterile, freshly cut, avocado pieces, but again only for a limited period (Darvas and Kotze 1987).

5.21.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit? What is the optimum season for conducting detection testing?***

Symptoms are visible on leaves, fruit, and stems during all growth stages (Rodríguez-Polanco et al. 2020). New infections produce lesions which become infectious and results in further lesions. As the lesions age the infectious tissue decreases and no longer produces spores (Rodríguez-Polanco et al. 2020). Specifically, the disease appears on leaves as individual spots. Small lesions initially (1-6mm) form, are angular in shape, purple-purplish brown in colour, and appear as flecks, or spots, near the leaf margin. Chlorotic halos are visible on both leaf surfaces following the appearance of the lesions.

In high humidity conditions sporulation causes a grey, felty mycelium growth in the centre of the lesions. Lesions may coalesce, forming areas of infection that may appear as brown coloured necrotic tissue and may form irregular patches. Leaves curl, deform, and may fall off the plant (Darvas 1982; California Department of Food & Agriculture 2016).

Dark brown to black lesion of 2-10mm may appear on stems and fruit pedicels of the plant (Darvas 1982). Cracks are formed only in older spots and don't always develop in lenticels (or the surface of stems where gas is exchanged between the atmosphere and underlying tissue) (fruit stems).

- ***What are the known environmental conditions conducive to symptom expression?***

Spore release is highest at 18°C (no infection will occur below 15°C) (Darvas 1982; Everett and Siebert 2018). Spore release declines as temperatures increases to 25°C (Darvas 1982; Everett and Siebert 2018). Spore abundance exposes avocados to a greater chance of infection and when this occurs early in the critical (wet and warm) period this can lead to significantly more *P. purpurea* spots (Darvas and Kotzé 1979). Moist conditions are conducive to symptom expression (Darvas 1982; Kallideen 2020). There is a positive correlation of symptom development with increasing humidity up to at least 84% (Darvas 1982). Relative humidity and leaf wetness are not important in conidia release but can influence pathogen development (Kallideen 2020). Outside the PEQ environment the most important environmental parameter is moisture which has a significant influence on production of *P. purpurea* conidia. Therefore, within the PEQ facility, moisture will be the most significant factor for creating the optimal environment for symptom expression. Low temperature (below 18°C) however will supress spore production or spore release despite substantial moisture (Darvas and Kotzé 1979). Significant numbers of spores are produced with the onset of rainy, warm temperatures.

- ***What are the limitations to taking samples for potential testing?***

Detection would have to include symptomatic tissue. As the pathogen can be latent for at least three months after penetration (Darvas and Kotzé 1979), the best time for detection testing is after three months in PEQ. The pathogen is likely to spread to further plant parts when the plants break dormancy and produce new shoots and leaves. New shoot tissue is infected wherever the disease occurs (California Department of Food & Agriculture 2016). Samples must be isolated from fresh young lesions and the surface disinfected to eliminate other contaminating fungi that can outgrow *P. purpurea* (Menge and Ploetz 2003).

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Lesions on leaves produce the most conidia and therefore this plant part constitutes the main source of inoculum for primary and secondary infections and the continuation of the disease from one season to another (Benson 2018; Kallideen 2020).

5.21.8 References

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5.22 *Pseudoidium perseae-americanae* (powdery mildew of avocado)

Pseudoidium perseae-americanae is a fungus that causes powdery mildew on the leaves, buds and herbaceous stems of *Persea americana* (avocado) plants.

5.22.1 Taxonomic description

Scientific name: *Pseudoidium persea-americanae* (Liberato & R.W. Barreto) Liberato & R.W. Barreto 2012

Order/Family: Erysiphales, Erysiphaceae

Other names include: *Oidium perseae-americanae* Liberato & R.W. Barreto 2006

Taxonomic notes: There have been occasional records of *Pseudoidium* spp. causing powdery mildew on avocado plants in the literature but *Pseudoidium perseae-americanae* is the first to be described to species level (Liberato and Barreto 2006). In some records, there is a degree of uncertainty about the identity of the causative agent of powdery mildew on avocado plants because it is referred to only by the common name 'powdery mildew' or only described to the genus level, as *Oidium* species. However, the literature suggests that *Pseudoidium perseae-americanae* is the most prevalent cause of powdery mildew in avocado as there are very few records of powdery mildew being caused by other species, such as *Podosphaeria persea-americanae*. *Podosphaera perseae-americanae* is distinguishable from *Pseudoidium persea-americanae* by morphological characteristics, where *Podosphaera* produces conidia (asexual spores) in chains (catenate-type), and *Pseudoidium* (syn. *Oidium*) produces non-catenate conidia (Siahaan et al., 2016).

The USDA, Species Fungorum, and much of the recent literature use *Pseudoidium persea-americanae* as the preferred name, and regard *Oidium persea-americanae* to be an obsolete name.

5.22.2 Hazard identification

Pseudoidium perseae-americanae is not known to be present in New Zealand:

- it is not listed in either Biota NZ (2022) or PPIN (2021),
- the regulatory status of *P. perseae-americanae* has not been assessed (ONZPR 2021).

Pseudoidium perseae-americanae has the potential to establish (and spread) in New Zealand because:

- avocado is the only recorded host plant (Farr and Rossman 2021), and it is grown commercially and domestically in northern parts of New Zealand (Plant & Food Research 2019).

Pseudoidium perseae-americanae has the potential to cause harm to New Zealand because:

- it is a minor disease problem on avocado plants under prolonged warm and humid conditions with low rainfall (Stevens and Piper 1941a; Zentmyer 1953; McMillan 1976). Disease promoting conditions may occasionally be met in New Zealand's avocado growing regions, and frequently met in protected environments such as nurseries (Chappell 2013c, 2013b; MPI 2021b).

Pseudoidium perseae-americanae is associated with avocado budwood because:

- avocado is the only known host plant of *P. perseae-americanae* (Liberato and Barreto 2006),

- *Pseudoidium perseae-americanae* colonises the epidermis of susceptible host plant tissues (leaves, herbaceous stems, buds, shoots and inflorescence) and overwinter on hosts as dormant mycelia (Plantwise 2015),

Given the arguments and evidence above, *P. perseae-americanae* meets the criteria to be a hazard on avocado budwood (as in the commodity description) imported to New Zealand.

5.22.3 Risk assessment

Biology and epidemiology of Pseudoidium perseae-americanae

Fungi in the order Erysiphales are commonly referred to as powdery mildews (Glawe 2008). They colonise the epidermis of susceptible host plant tissues (leaves, herbaceous stems, buds, shoots and inflorescence) (Plantwise 2015) and obtain nutrients via haustoria (feeding organs) that grow into the epidermal cells of the plant organs (Agrios 2005).

Host range

Avocado is the only recorded host of *P. perseae-americanae* (Farr and Rossman 2021). It is common for species that cause powdery mildews to be host specific (Liberato and Barreto 2006).

Distribution

The recorded distribution of *P. perseae-americanae* and climate similarity with New Zealand is provided in Table 5-22.

Table 5-22. The geographic distribution of *Pseudoidium perseae-americanae*.

Continent/Region	**Country (CMI range)** (Phillips et al. 2018; Farr and Rossman 2021).	**CMI***
Africa	Kenya (0.4-0.8), Togo (0.4-0.5), Uganda (0.5-0.8), Guinea (0.3-0.5)	0.3-0.8
Asia	Israel (0.6-0.7)	0.6-0.7
North, Central and South America	Barbados, Bermuda, Brazil (0.3-0.9), Costa Rica (0.3-0.6), Cuba (0.5), Honduras (0.3-0.6), Panama (0.3-0.5), Peru (0.4-0.8), United States of America (0.5-0.8), Venezuela (0.4-0.8), West Indies (0.4-0.7).	0.3-0.9
Oceania	New Caledonia (0.6-0.7)	0.6-0.7

* Climate Match Index (CMI) with all of New Zealand (Phillips et al. 2018)

Symptoms

Young flushes including leaves, inflorescence and buds are most susceptible to infection (Plantwise 2015). Dark green to purplish-brown discolorations develop on the upper surface of leaves along the midribs. Leaves may be distorted and curled. These discoloured infection sites produce abundant conidia that appear as characteristic white-grey powder on the underside of the leaves (Zentmyer 1953). Conidia are produced quickly (3–7 days post infection under disease-promoting conditions) and in large amounts. White mycelium can cover juvenile parts (inflorescence and leaves) causing damage and even necrosis. Old infection sites appear as conspicuous chlorotic areas with net-like markings (McMillan 1976).

Optimal conditions for growth and symptom development

Pseudoidium perseae-americanae can become severe in humid, damp, shaded areas, especially nurseries. Rain can prevent spread because it washes conidia from plant surfaces before they germinate (Stevens and Piper 1941a; Zentmyer 1984; Plantwise 2015).

Reproduction and transmission

The complete life cycle of *P. perseae-americanae* may involve an asexual stage as well as a sexual stage (Glawe 2008). During asexual reproduction, conidia (asexual spores) are produced on the host. The conidia have a white, powdery appearance, and the conidia production stage is the state that is referred to as 'powdery mildew'. Conidia are produced in large amounts, within a short space of time (3–7 days post infection under disease-promoting conditions) and therefore can cause rapid spread of the disease. They are airborne and mostly transmitted by wind, but also by human activity on people (clothing, hands), on equipment (pruning tools, mechanical harvesters or hedgers) or on vehicles (Agrios 2005; Business Queensland 2019). The conidia of *Pseudoidium (Oidium)* spp. do not require free water on plant surfaces to initiate infection (Agrios 2005). Powdery mildews can overwinter as dormant asexual mycelia in buds of host plants or as sexual ascospores independent of a host (Glawe 2008). The teleomorph (sexual reproductive stage) of *P. perseae-americanae* has not been observed (Liberato and Barreto 2006).

5.22.4 Likelihood of entry

There is potential for *P. perseae-americanae* to be imported into post-entry quarantine on avocado budwood, in which case it would be highly likely to produce visible symptoms in PEQ:

- *Pseudoidium perseae-americanae* has a tropical/subtropical distribution and is reported from reported from 17 countries in 5 continents (Table 5-22),
- it can be associated with leafless avocado budwood because it is known to infect buds and herbaceous stems of avocado plants (Zentmyer 1953; McMillan 1976; Plantwise 2015),
- there is a low likelihood that severely infected plants would be used to produce budwood material. However, trees that were only mildly infected can appear healthy but harbour mycelium in dormant buds (Glawe 2008). Therefore, during times of mild disease infection, there is the potential for some contaminated material to be used for leafless budwood,
- budwood/leafless cuttings are dipped in 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility, which is likely to kill surface infections, but may not penetrate into buds where dormant mycelium has been shown to overwinter (Glawe 2008),
- *Pseudoidium perseae-americanae* causes disease symptoms under warm and humid conditions and especially in nurseries (Zentmyer 1953; McMillan 1976; Plantwise 2015), suggesting that conditions in PEQ are likely to promote the expression of symptoms,

Therefore, the likelihood of infected plants being released from PEQ is considered to be negligible.

It is likely that *P. perseae-americanae* would produce characteristic powdery conidia on infected plants in PEQ, which may not be contained in level 3A:

- all plants must be inspected at least twice per week during periods of active growth and once per week during dormancy (MPI 2021a). *Pseudoidium perseae-americanae* could produce inoculum between inspections because conidia are produced in large amounts, within a short space of time (3–7 days post infection under disease-promoting conditions) (Glawe 2008),
- airborne conidia may escape from level 3A facilities because the mesh screen over the vents is required to have a maximum opening size of 0.04 mm^2 (MPI 2021a), which may be too large to contain the conidia of *P. perseae-americanae*, which are 0.027-0.04 × 0.01-0.02 mm (Liberato and Barreto 2006),
- other measures in place within level 3A glasshouses (including protective clothing and hand washing) (MPI 2021a) would reduce the likelihood of any spores/conidia leaving the glasshouse, to a very low level,

Given the arguments and evidence above, that is:

- *Pseudoidium perseae-americanae* has a reasonably wide geographical distribution but is mostly limited to tropical/subtropical climates,
- It could be unknowingly imported into PEQ on leafless avocado budwood because mycelium can overwinter on dormant buds,

- it is likely that *P. perseae-americanae* would produce symptoms on infected plants under suitable conditions and be detected in PEQ. Therefore, the likelihood of infected plants being released from PEQ is considered to be negligible, but,
- airborne conidia may not be contained in level 3A PEQ because they could be produced between inspections and are small enough to fit though the vents. However, the amount of conidia likely to escape is small because only a portion of spores would escape through the vents and plant inspections are likely to detect the fungus at early stages of sporulation,

the likelihood of *P. perseae-americanae* entering New Zealand associated with avocado budwood is considered to be very low, with low uncertainty.

5.22.5 Likelihood of exposure

Since there is a negligible risk of avocado plants infected with *P. perseae-americanae* at the time of their release from Post Entry Quarantine, this assessment is made on the basis that airborne spores of *P. perseae-americanae* have escaped PEQ and entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. If suitable hosts were present near the post-entry quarantine facilities, for example, if the facility was located near avocado orchards, private gardens with avocado trees orother native or naturalised plants from the Lauraceae family, the fungus could be exposed to suitable host material. However, the amount of inoculum likely to escape is considered to be small, and the conidia would have to land on a suitable host under suitable conditions to germinate and cause disease. Due to limited information related to *P. perseae-americanae* the uncertainty associated with this assessment is high.

Given the arguments and evidence above, the likelihood of exposure of *P. perseae-americanae* in New Zealand after escaping from Level 3A PEQ is considered very low, with low uncertainty.

5.22.6 Likelihood of establishment

This assessment is made on the basis that *P. perseae-americanae* has been successfully exposed to a suitable host plant in the New Zealand environment.

- *Pseudoidium perseae-americanae* is mostly distributed in tropical/subtropical regions that are located in the tropical rain belt, have a rainy and dry season, and do not share a high level of climatic similarity to the whole of New Zealand (Table 5-22) (Phillips et al. 2018).
- Powdery mildew of avocado is most prevalent during the dry season because rainfall can wash conidia off the leaves before they germinate (Crane et al. 2019). It can become severe in warm, humid and protected areas, especially in nurseries (Stevens and Piper 1941b; Zentmyer 1984).
- Northern parts of the North Island (where most of the commercial avocado is grown (Plant & Food Research 2019)) experience high humidity and warm temperatures during the summer, which are likely to be suitable for the establishment of *P. perseae-americanae.* However, these regions also receive abundant rainfall within the range of 1000-2200mm median annual total rainfall, which may limit the ability of *P. perseae-americanae* to establish and spread (Chappell 2013c, 2013a).
- There is limited information in the literature regarding optimal temperatures for *P. perseae-americanae,* but it is reported to grow in mild temperatures (< 24℃) (Plantwise 2015). The optimal temperature for conidia germination of other *Oidium* species that cause powdery mildew is between 18-25℃ (Morrison 1964; Sastra-Hidayat 1992).
- The conidia of *Pseudoidium (Oidium)* spp. do not require free water on plant surfaces to initiate infection (Agrios 2005).
- Avocado is the only reported host of *P. perseae-americanae* so it's establishment is highly likely to be limited to avocado growing regions due to host availability and climate,
- Powdery mildew can overwinter as dormant asexual mycelia in buds of host plants or as sexual ascospores independent of a host (Glawe 2008), however, the teleomorph (sexual

reproductive stage) of *P. perseae-americanae* has not been observed (Liberato and Barreto 2006) suggesting that it is most likely to reproduce asexually in New Zealand.

Given the arguments and evidence above, that is:

- *Pseudoidium perseae-americanae* is mostly distributed in tropical/subtropical climates, indicating that the northern avocado growing regions of New Zealand may have suitable conditions for establishment (warm and humid), especially during summer,
- the pathogen could overwinter as dormant asexual mycelia in buds,
- *Pseudoidium (Oidium)* spp. can spread easily because they produce large amounts of airborne conidia within a short period of time (3–7 days) and do not require free water on plant surfaces to initiate infection,

the likelihood of *P. perseae-americanae* establishing in New Zealand is considered moderate, with moderate uncertainty.

5.22.7 Impacts in New Zealand

Airborne conidia of *P. perseae-americanae* are produced in large amounts, within a short space of time (3–7 days post infection under disease-promoting conditions) and can cause rapid spread of the disease. Conidial dispersal appears to occur mostly over short distances, but they can be transmitted long distances by wind (Glawe 2008) and human activity on people (clothing, hands), on equipment (pruning tools, mechanical harvesters or hedgers) or on vehicles (Agrios 2005; Business Queensland 2019). Rain can prevent spread because it washes conidia from plant surfaces before they germinate (Stevens and Piper 1941b; Zentmyer 1984; Plantwise 2015).

Avocado is the only reported host of *P. perseae-americanae* so it’s spread is highly likely to be limited to avocado growing regions due to host availability.

Economic impacts

In New Zealand, the severity of disease, spread and consequently the economic impact is highly likely to be dependent on host availability and climatic conditions.

- Avocado is the only known host of *P. perseae-americanae* (Farr and Rossman 2021) and any economic impacts are highly likely to be limited to the avocado industry.
- Powdery mildew caused by *P. perseae-americanae* is typically a minor disease problem that does not require control measures.
- It can become severe in warm, humid and protected areas, such as nurseries (Stevens and Piper 1941b; Zentmyer 1984), and impacts may be exaggerated in these environments.
- Infections can damage young flushes including leaves, inflorescence and buds (McMillan 1976; Plantwise 2015), which can weaken the host plant, reduce plant growth and fruit yield.
- If favourable conditions persist and the disease becomes severe, fungicide sprays such as wettable sulphur can be effective at controlling the fungus (McMillan 1976).
- The impact of *P. perseae-americanae* on avocado may be mitigated by fungicide sprays used to control other fungal disease that already occur on avocado plants in New Zealand, such as *Colletotrichum* spp., *Botryosphaeria dothidea*, *Neofusicoccum parvum* and *Phomopsis* sp. (Everett and Siebert 2018).
- *Pseudoidium perseae-americanae* is unlikely to have serious impacts on trade because it has not been reported to infect mature fruit and can be effectively managed by fungicide treatments (McMillan 1976).

Given the arguments and evidence above, that is:

- *Pseudoidium perseae-americanae* is host specific to avocado and is only reported to cause disease in countries with tropical/subtropical climates. It is usually a minor disease problem but can become severe in prolonged warm, humid and shaded conditions, especially in nurseries,

- *Pseudoidium perseae-americanae* has not been reported to be associated with fruit and can be effectively managed by fungicide treatments, suggesting that it is unlikely to have major disease or trade impacts,

the economic impact of *P. perseae-americanae* to New Zealand is considered to be low, with low uncertainty.

Environmental impacts

Avocado is the only known host of *P. perseae-americanae* (Farr and Rossman 2021) and it is common for powdery mildews to be host specific (Liberato and Barreto 2006), indicating that it is unlikely that there would be any impacts to native flora in New Zealand.

Given the arguments and evidence above, the impact on the environment from the establishment of *P. perseae-americanae* in New Zealand is considered to be negligible, with low uncertainty.

Human health impacts

Pseudoidium perseae-americanae is unlikely to cause unwanted impacts on human health. No evidence of such impacts has been found.

Given the evidence above, the health impact of *P. perseae-americanae* to New Zealand is considered to be negligible with low uncertainty.

Sociocultural impacts

Pseudoidium perseae-americanae is reported as a minor disease problem on avocado (McMillan 1976), indicating that it is unlikely to cause significant sociocultural impacts in New Zealand.

Given the arguments and evidence above, the sociocultural impact of *P. perseae-americanae* to New Zealand is considered to be negligible, with low uncertainty.

Overall impact to New Zealand

The overall impact of *P. perseae-americanae* on the New Zealand economy, environment, health and society is considered to be low, with low uncertainty.

5.22.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is very low with low uncertainty,
- assuming entry, the likelihood of exposure is very low, with low uncertainty,
- assuming successful exposure, the likelihood of establishment is moderate with moderate uncertainty,
- the impacts on the New Zealand economy, environment, health and society are low with low uncertainty,

the overall level of assessed risk to New Zealand from *P. perseae-americanae* on imported avocado budwood (as in the commodity description) is considered to be very low, with low uncertainty.

5.22.9 Management considerations

Pseudoidium perseae-americanae infects young flushes including leaves, inflorescence and buds (Plantwise 2015), resulting in dark discolorations, leaf distortions and the production of characteristic white-grey powder conidia on the surface of infected tissue (Zentmyer 1953). Airborne conidia are produced quickly and in large amounts (3–7 days post infection under disease-promoting conditions), allowing the fungus to spread widely and rapidly.

Powdery mildew symptoms can become severe in warm (< 24℃), humid, damp, shaded areas, especially in nurseries. Rain/water can wash conidia from plant surfaces before they germinate (Stevens and Piper 1941a; Zentmyer 1984; Plantwise 2015). The optimal temperature for conidia production of *Pseudoidium (Oidium)* spp. is between 18-25℃ (Morrison 1964; Sastra-Hidayat 1992), suggesting that it could grow and spread in summer in New Zealand.

Powdery mildews can overwinter as dormant asexual mycelia in buds of healthy looking host plants (Glawe 2008). In case *P. perseae-americanae* is unknowingly imported into PEQ, plants should be inspected regularly for symptoms, as airborne conidia are produced rapidly and in large amounts (Zentmyer 1953; McMillan 1976), and may not be contained in level 3A PEQ due to their small size (Liberato and Barreto 2006; MPI 2021a).

Protective and eradicant fungicide treatments are effective against powdery mildew, although the fungus' development of resistance to fungicides is a growing concern. Rotation of different fungicides is recommended to slow the development of resistance. Protective fungicides must be applied to healthy tissues before infection takes place, and include tebuconazole, cyprodinil/fludioxonil, boscalid/pyraclostrobin, fluoxastrobin, wettable sulfur, myclobutanil, azoxystrobin, triadimefon, thiophanate-methyl, propiconazole, Reynoutria sachalinensis and potassium bicarbonate. Eradicant fungicides are used to treat existing infections, and include lime sulfur 28%, neem oil, piperalin, stylet oil and tebuconazole (Koike et al. 2020).

5.22.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Young flushes including leaves, inflorescence and buds are most susceptible to infection (Plantwise 2015). Dark green to purplish-brown discolorations develop on the upper surface of leaves along the midribs. Leaves may be distorted and curled. Infection sites produce abundant conidia which appear as characteristic white-grey powder on the underside of the leaves (Zentmyer 1953). Conidia are produced quickly and in large amounts (3–7 days post infection under disease-promoting conditions). White mycelium can cover juvenile parts (inflorescence and leaves) causing damage and even necrosis. Old infection sites appear as conspicuous chlorotic areas with net-like markings (McMillan 1976).

- ***What are the known environmental conditions conducive to symptom expression?***

Powdery mildew symptoms can become severe in warm (< 24℃), humid, damp, shaded areas, especially in nurseries. Rain/water can wash conidia from plant surfaces before they germinate (Stevens and Piper 1941a; Zentmyer 1984; Plantwise 2015). There is limited information in the literature regarding the optimal temperature for *P. perseae-americanae*, but the optimal temperature for conidia production of other *Pseudoidium (Oidium)* spp. is between 18-25℃ (Morrison 1964; Sastra-Hidayat 1992).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Powdery mildew overwinters as dormant asexual mycelia in buds of healthy looking host plants (Glawe 2008). In case dormant *P. perseae-americanae* mycelia is unknowingly imported into PEQ, plants should be inspected regularly for symptoms, as airborne conidia can be produced rapidly and in large amounts under disease promoting conditions (Zentmyer 1953; McMillan 1976), and may not be contained in level 3A PEQ due to their small size (Liberato and Barreto 2006; MPI 2021a)

- ***What is the optimum season for conducting detection testing?***

Pseudoidium perseae-americanae infections begin in spring when conditions are warm (< 24°C) and humid (Plantwise 2015). In its natural geographical distribution (tropical/subtropical climates) (Farr and Rossman 2021) it is most prevalent in the dry season. This suggests that summer would be the optimum season for detection testing in New Zealand.

5.22.11 References

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5.23 *Pyrrhoderma noxium* (brown root rot)

Pyrrhoderma noxium is a tropical and sub-tropical fungus that causes root rot in a wide range of host species, frequently resulting in plant death. Avocado is reported to be a highly susceptible host of this fungus.

5.23.1 Taxonomic description

Scientific name: *Pyrrhoderma noxium* (Corner) L.W. Zhou & Y. C. Dai

Order/Family: Hymenochaetales / Hymenochataceae

Other names include: *Fomes noxius, Phellinus noxius*

Taxonomic notes:

Fomes noxius is the basionym of this species (Robert et al. 2005). It was reclassified to *Phellinus noxius* in 1965 and this is the predominant name found in the literature (Stewart et al. 2020). Recent molecular phylogenetic analysis reassigned this fungus to the genus *Pyrrhoderma* under the name *P. noxium* (Zhou et al. 2018). The currently recognised name is *Pyrrhoderma noxium* (Robert et al. 2005), but the phylogeny and taxonomy of this species continues to be disputed (Stewart et al. 2020). *Pyrrhoderma noxium* is a diverse species that likely represent several distinct genetic groups (Garfinkel et al. 2020).

5.23.2 Hazard identification

Pyrrhoderma noxium is not known to be present in New Zealand:

- The species is listed in the NZOR (2021) and Biota NZ (2022) as being absent for New Zealand. This species is listed as a “regulated” and “unwanted” organism in ONZPR (2021) under the synonym *Phillinus noxius*.

Pyrrhoderma noxium has the potential to establish (and spread) in New Zealand because:

- This fungus can be spread though root to root contact of hosts, or through airborne spores infecting fresh wounds of live plants (Dann et al. 2012; Leung et al. 2020).
- Known hosts of this fungus are prevalent in New Zealand, including horticultural (e.g. grape and avocado) and forestry (e.g. eucalyptus) species.
- *P. noxium* is a tropical or subtropical disease and optimal growth occurs at high temperatures. Therefore, establishment and spread of this fungus is likely to be limited to warmer regions of New Zealand (Everett and Siebert 2018).

Pyrrhoderma noxium has the potential to cause harm to New Zealand because:

- Many economically significant hosts, such as avocado, grapevine, and citrus, are grown in northern regions of New Zealand where conditions are most suitable for establishment.
- Infections of this fungus frequently cause rapid death of established trees and may cause a significant loss of yield for some horticultural species.
- Artificial inoculation trials with *P. noxium* reported 100% loss of avocado seedlings within 3 months (Dann 2016), and 10% death of trees in Australian avocado orchards had an estimated cost of AUD$5400 per hectare (Everett and Siebert 2018).

Pyrrhoderma noxium is associated with *P. americana* budwood because:

- Avocado has been reported as a host of *P. noxium* in Taiwan, Australia, Papua New Guinea and the Malay Peninsula (Ann et al. 2002; Dann et al. 2012; Farr and Rossman 2021).
- Airborne basiodiospores may be present on the surface of avocado budwood.

Given the arguments and evidence above, *Pyrrhoderma noxium* meets the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.23.3 Risk assessment

Biology and epidemiology of P. noxium

Host range

Pyrrhoderma noxium is reported infecting at least 200 different species across 59 plant families (Ann et al. 2002). This includes many economically import species such as lemon (*Citrus limon*), apricot (*Prunus armeniaca*), grapevine (*Vitis vinifera*), and avocado (*Persea americana*) (Ann et al. 2002; CABI 2021). This fungus appears to be a non-host specific opportunistic pathogen (CABI 2021). Infections of avocado have been reported in Taiwan, Australia, Papua New Guinea and the Malay Peninsula (Ann et al. 2002; Dann et al. 2012; Farr and Rossman 2021).

Distribution

This pathogen is widespread in tropical and subtropical regions around the world (CABI 2021; Farr and Rossman 2021) as shown in Table 5-23.

Table 5-23: The geographical distribution of Pyrrhoderma noxium.

Region	Country	CMI*
Africa	Angola, Benin, Burkina Faso, Cameroon, Central African Republic, Democratic Republic of Congo, Côte d'Ivoire, Gabon, Ghana, Kenya, Nigeria, Sierra Leone, Tanzania, Togo, Uganda	0.4-0.7
Asia	China, Hong Kong, India, Indonesia, Japan, Malaysia, Myanmar, Pakistan, Philippines, Singapore, Sri Lanka, Thailand, Taiwan, Vietnam	0.4-0.8
Americas	Brazil, Costa Rica, Cuba, Puerto Rico	0.4-0.6
Oceania	American Samoa, Australia, Federated States of Micronesia, Fiji, Niue, Northern Mariana Islands, Papua New Guinea, Samoa, Solomon Islands, Vanuatu	0.4-1.0

* Climate Match Index (CMI) with all of New Zealand (Phillips et al. 2018)

Symptoms

Infection by *P. noxium* causes brown root rot in a wide range of hosts, usually in woody species (Dann et al. 2012; Zhou et al. 2018). Symptoms are typically characterised by a yellow/brown/black mycelial mat developing on exposed roots and at the base of the trunk or stem, often referred to as a “stocking” (Ota and Sahashi 2018). Also causes white rot of wood, which is spongy, soft, and whiter than normal. Conspicuous brown lines are often observed permeating through the rotted wood. Early stages of the infection can be symptomless, but once the disease is established it effectively girdles the tree, causing foliar necrosis, defoliation and rapid plant death (Leung et al. 2020). This decline typically occurs within a few months of infection, but may take several years in some hosts (Ota and Sahashi 2018).

Symptoms of *P. noxium* reported on avocado are severe (Dann et al. 2009; Dann 2016; Everett and Siebert 2018). As is typically observed in other hosts, infections develop a brown mycelial “stocking” around the base of the plant (Everett and Siebert 2018). Underneath the stocking the wood is brown with white patches. The infection causes rapid decline and death of avocado, often leaving leaves and fruit hanging on the tree (Dann et al. 2009). Dann (2016) reported death of avocado seedlings occurring within 3 months of inoculation.

Optimal conditions for growth and symptom development

The optimum temperature for growth of *P. noxium* is 30℃, and it is reported that is does not survive or spread at low temperatures (Everett and Siebert 2018). The fungus also prefers acidic conditions below pH 5.0, and it is reported that it will not grow when pH values are greater than 7.5 (Ann et al. 1999).

Reproduction and transmission

The primary means of spread for this fungus is root-to-root contact (Dann et al. 2012). The fungus may remain viable in fragments of infected roots within soil for more than ten years (Chung et al. 2015; CABI 2021). However, soil without root fragments is not a reservoir of disease (Wu et al. 2020). The fungus may also be spread through airborne basidiospores, allowing for long-distance dispersal (Chung et al. 2015; Leung et al. 2020). Fruiting bodies producing basidiospores are typically observed on dead and fallen trees (Ota and Sahashi 2018). These spores may then enter living hosts through wounds (CABI 2021) or survive for 3 to 4.5 months within soil (Chung et al. 2015).

5.23.4 Likelihood of entry

Pyrrhoderma noxium is a disease infecting the roots and trunk of trees. While the disease does cause leaf wilting and defoliation symptoms, mycelia are not known to be present in leaves, branches, or shoots (Ann et al. 1999; MPI 2016). Therefore, it is considered unlikely that the shoots and new growth from which avocado budwood cuttings are taken would be contaminated with *P. noxium* mycelia.

It is suggested that airborne basidiospores may play a role in the establishment of *P. noxium* at new sites (Chung et al. 2015; Leung et al. 2020), so may contaminate the surface of avocado budwood. If basiodiospores were present on the surface of avocado budwood, it is expected that the surface disinfection procedure (1% sodium hypochlorite dip for 2 minutes) would be effective in making the spores inviable.

Avocado plants are highly susceptible to *P. noxium* infection. Dann (2016) found that avocado seedlings (Reed variety) artificially inoculated with a *P. noxium* isolate originating from avocado all died within 3 months of inoculation. It is not reported when the visible symptoms of infection first occurred. A second trial found that seedlings of both Reed and Hass avocados died within 2 months of inoculation. Given the rapid deterioration of avocado exhibited in these trials, it appears likely that symptoms of the disease would be evident during the period of PEQ if environmental conditions are suitable.

Pyrrhoderma noxium is a tropical and subtropical disease that requires warm temperatures to develop. Therefore, conditions in PEQ may not be suitable for development of the disease. *Pyrrhoderma noxium* is only thought to persist long-term within wood material and root debris, so if fungal mycelia were present on grafted buds it is not expected to survive for a significant period at low to moderate temperatures (Everett and Siebert 2018). Basiodiospores may survive up to 4.5 months within soil (Chung et al. 2015), but are not expected to survive an extensive period without optimal conditions.

The primary means of transmission for this fungus is root-to-root contact, which is not a risk within a Level 3A PEQ facility. Basidiocarps of *P. noxium* are uncommon in the field, but may be produced on the remnants of fallen or dead trees (Ota and Sahashi 2018). Basiodiocarps have not been reported on avocado (Dann et al. 2012). Therefore, if infection of *P. noxium* were to occur within the PEQ facility, it is highly likely that it would be sufficiently contained.

Given the arguments and evidence above, that is:

- *P. noxium* is not reported to be associated with shoots, stems or leaves of its hosts so it is unlikely that budwood cuttings would contain infected tissue,
- surface sterilisation of avocado budwood is likely to make any basiodiospores that may be present on the surface of the cuttings inviable,
- symptoms on avocado are severe and develop rapidly, so are likely to be observed within the period of PEQ is environmental conditions are suitable,
- if environmental conditions in PEQ are not suitable for disease development, the fungal mycelia or basiodiospores are not expected to remain viable for a significant period of time, and
- outbreaks of *P. noxium* would be effectively contained within Level 3A PEQ,

the likelihood of *P. noxium* entering New Zealand associated with *P. americana* budwood is considered to be negligible, with low uncertainty. Therefore, further assessment is considered unnecessary at this time; likelihoods of exposure and establishment and impacts to New Zealand are not further assessed.

5.23.5 Overall level of associated risk to New Zealand

Based on the assessment of likelihood above, that:

- the likelihood of *P. noxium* entering New Zealand associated with *P. americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ) is considered negligible, with low uncertainty;

the overall level of risk to New Zealand from *P. noxium* on imported *P. americana* bedwood is considered to be negligible, with low uncertainty.

5.23.6 Management considerations

Symptoms of *P. noxium* on avocado are reported to be severe (Dann et al. 2009; Dann 2016; Everett and Siebert 2018). Infections develop a brown mycelial “stocking” around the base of the plant (Everett and Siebert 2018). Underneath the stocking the wood is brown with white patches. The infection causes rapid decline and death of avocado, often leaving leaves and fruit hanging on the tree (Dann et al. 2009). This is consistent with the symptoms reported on many other host species. The disease may be symptomless in the early stages of infection (Leung et al. 2020), but 100% dieback within 3 months is reported for avocado seedlings (Dann 2016). Therefore, it appears highly likely that infection would be observed within the period of PEQ.

Pyrrhoderma noxium is a tropical and sub-tropical disease with a strong preference for warm temparatures and the optimal temperature for growth is 30℃ (Everett and Siebert 2018; Zhou et al. 2018). Fungal mycelia can survive extended periods of time in wood tissue and debris (Chung et al. 2015; CABI 2021), but does not survive for extended periods in soil without root fragments (Wu et al. 2020). Long term persistence of the fungus at low to moderate temperatures is not expected to occur in avocado budwood in PEQ.

This fungus can be readily isolated using cultures from roots or the lower parts of basal stems exhibiting rot symptoms and mycelial masses. The fungus is not known to be present in shoots or leaves, so positive cultures are not expected from these plant parts, even they may be exhibiting typical disease symptoms. *Pyrrhoderma noxium* can be identified through morphological or molecular methods (Leung et al. 2020). Molecular methods may use sequencing of the ITS or 28S regions, or microsatellites (Chung et al. 2015; Leung et al. 2020).

5.23.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

In avocado, *P. noxium* infection is reported to have severe symptoms (Dann et al. 2009; Dann 2016; Everett and Siebert 2018). As is typically observed in other hosts, infections cause root rot and the development of a brown mycelial “stocking” around the base of the plant (Everett and Siebert 2018). Underneath the stocking the wood is brown with white patches. The infection causes rapid decline and death of avocado, often leaving leaves and fruit hanging on the tree (Dann et al. 2009). Dann (2016) reported death of avocado seedlings occurring within 3 months of inoculation.

- ***What are the known environmental conditions conducive to symptom expression?***

The optimum temperature for growth of *P. noxium* is 30℃, and it is reported that is does not survive or spread at low temperatures (Everett and Siebert 2018). The fungus also prefers acidic soil conditions below pH 5.0, and it is reported that it will not grow when pH values are greater than 7.5 (Ann et al. 1999).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

This fungus can be readily isolated using cultures from roots or lower parts of basal stems exhibiting rot symptoms and mycelial masses. The fungus is not known to be present in shoots or leaves, so positive cultures are not expected from these plant parts, even though they may be exhibiting disease symptoms. Asymptomatic infection has not been reported in avocado.

- ***What is the optimum season for conducting detection testing?***

Given that high temperature requirements of *P. noxium*, detection will be most likely during summer.

5.23.8 References

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5.24 *Raffaelea* spp. (laurel wilt disease)

The following *Raffaelea* species will be assessed in this PRA: *R. aguacate*, *R. campbellii*, *R. canadensis* and *R. lauricola*.

Raffaelea spp. are typically saprotrophic fungi symbiotically associated with ambrosia beetles. However, *Raffaelea lauricola* is unique in that it causes the vascular wilt disease in trees and shrubs in the Lauraceae family including *Persea americana*. Similarly, *R. canadensis* has also been associated with wilt disease.

5.24.1 Taxonomic description

Scientific name: *Raffaelea aguacate* D. R. Simmons, Dreaden & Ploetz (2016)

Scientific name: *Raffaelea campbellii* D. R. Simmons, A. Campbell & Ploetz (2016)

Other names include: *Raffaelea campbelliorum* D. R. Simmons, A. Campbell & Ploetz (2016)

Scientific name: *Raffaelea canadensis* L. R. Batra (1967)

Other names include: *Tuberculariella ambrosiae* A. Funk (1965)

Scientific name: *Raffaelea lauricola* T. C. Harr., Fraedrich & Aghayeva (2008)

Order/Family: Ophiostomatales/Ophiostomataceae

5.24.2 Hazard identification

Raffaelea spp. (*R. aguacate*, *R. campbellii*, *R. canadensis* and *R. lauricola*) are not known to be present in New Zealand:

- *Raffaelea* spp. are not listed in either Biota NZ (2022), NZOR (2021) or PPIN (2021).
- *Raffaelea lauricola* is a regulated pest for New Zealand (ONZPR 2021).
- *Raffaelea aguacate*, *R. campbellii*, *R. canadensis* are not listed in ONZPR (2021).

Raffaelea spp. have the potential to establish (and spread) in New Zealand because:

- *Persea americana* is a known host (Eskalen and McDonald 2011; Simmons et al. 2016) that is widely grown in New Zealand, in home gardens and commercial production, mainly in the north of the North Island. Other hosts in the Lauraceae family (e.g. *Cinnamomum camphora*) are also reported hosts of *R. lauricola* (Table 5-25) and are widespread is gardens, parks and street verges (NZPCN 2021).
- *Raffaelea* spp. cause wilting and tree death in regions such as the United States (Ploetz et al. 2017a) that have a climate match index (CMI) ≥ 0.7 indicating a similar climate to the whole of New Zealand (Phillips et al. 2018). Specifically, *R. lauricola* has the potential to spread through vectors that are present in New Zealand (*Xyleborinus saxesenii* and *Xylosandrus crassiusculus*); however, spread would most likely be limited as these beetle species are not the primary vector. The primary vector, *Xyleborus glabratus*, is not present in New Zealand (NZOR 2021).

Raffaelea spp. have the potential to cause harm to New Zealand because:

- *Raffaelea lauricola* and *R. canadensis* cause vascular wilt disease in avocado trees typically exhibited by wilting of foliage, sapwood discolouration, vascular dysfunction and in severe cases, tree death (Ploetz et al. 2012; Ploetz et al. 2017a). *Raffaelea* spp. have a symbiotic relationship with ambrosia beetle vectors; these vectors weaken or damage host trees and spread the disease to new hosts.,

- avocados are an important commercial crop in New Zealand. In the 2019-2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$ 50.6 million (Plant & Food Research 2020).
- In Florida alone, the impacts of *R. lauricola* specifically caused economic impacts between $183 million (if control measures were 50% effective) to $356 million (if no controls at all) (Olatinwo et al. 2021).
    - In 2017, approximately 3% of avocados grown in commercial production areas in Florida were lost to the disease and farmers had to incur the cost of tree removal and treatments to protect residual trees (Ploetz et al. 2017a; Olatinwo et al. 2021).

Raffaelea spp. is associated with *P. americana* budwood because:

- *Persea americana* is a known host of *Raffaelea* spp. (Eskalen and McDonald 2011; Ploetz et al. 2012; Simmons et al. 2016; Ploetz et al. 2017a);
- *Raffaelea* spp. have been isolated from branch tissue (Ploetz et al. 2012).

Given the arguments and evidence above, *Raffaelea* spp. meet the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.24.3 Risk assessment

Biology and epidemiology of Raffaelea spp.

Vectors

Raffaelea species are vectored by ambrosia beetles (Table 5-24). The most pathogenic species, *R. lauricola* is vectored by the ambrosia beetle *Xyleborus glabratus* (Hughes et al. 2017). *Raffaelea campbellii* and *R. canadensis* are also vectored by *X. glabratus* (Table 5-25). *Raffaelea aguacate* has been isolated from *Xyleborus bispinatus* (Table 5-25).

Raffaelea lauricola has been isolated from other beetle species (Table 5-24); however, *X. glabratus* contains 10-1000 times more colony forming units than other beetles (Ploetz et al. 2017c). Evidence suggests that a complex of beetles is involved in disease dissemination in avocado given that susceptible host species within the vicinity of *Raffaelea*-carrying *X. glabratus* don't necessarily succumb to infection when colonised by *X. glabratus* alone (Ploetz et al. 2017a).

Xyleborus glabratus tends to colonise and reproduce in stressed or dead trees and tends to have a strong preference for trees in the Lauraceae family (Ploetz et al. 2017a). Stressed trees release a metabolite sesquiterpene which attracts the beetles (Ploetz et al. 2017a). However, there is evidence that *X. glabratus* can also attacks healthy trees (Olatinwo et al. 2021). Ambrosia beetles carry fungi in specialised organs, mycangia (Mayers et al. 2020). Adult female beetles disperse to new trees and bore brood galleries into the host xylem (Ploetz et al. 2017a). Beetles tend to attack stems of larger diameter and the probability of beetle attack decreases with decreasing stem diameter (Olatinwo et al. 2021). Within the galleries beetles cultivate and feed on 'fungal gardens' along the walls of their galleries in tree sapwood (Mayers et al. 2020).

Table 5-24: List of ambrosia beetles from which Raffaelea lauricola has been isolated from and the mean percent of isolations. Data adapted from Ploetz et al. (2017c)

Ambrosia beetle species	Percent of individuals with *Raffaelea lauricola*	Status in New Zealand (NZOR 2021)
Xyleborus glabratus	73	Not listed in NZOR, **regulated**
Xyleborus bispinatus	63	Not listed in NZOR or ONZPR
Xyleborus ferrugineus	57	Not listed in NZOR, **regulated**
Xyleborus affinis	13	Not listed in NZOR, **regulated**
Xyleborus volvulus	32	Not listed in NZOR or ONZPR
Ambrosiodmus devexulus	0	Not listed in NZOR or ONZPR

Ambrosia beetle species	Percent of individuals with *Raffaelea lauricola*	Status in New Zealand (NZOR 2021)
Ambrosiodmus lecontei	2	Not listed in NZOR or ONZPR
Xyleborinus andrewesi	13	Not listed in NZOR or ONZPR
Xyleborinus gracilis	50	Not listed in NZOR, not assessed in ONZPR
Xyleborinus saxesenii	2*	Present, non-regulated
Xylosandrus compactus	0	Not listed in NZOR, **regulated**
Xylosandrus crassiusculus	4*	Present (Ministry for Primary Industries 2020), **regulated**

*Saucedo-Carabez et al. (2018) did not isolate *Raffaelea lauricola* from *X. saxesenii* or *X. crassiusculus*

Symptoms

At early stages of infection, trees exhibit wilting of green foliage of branch tips in sections of the tree canopy and wilting of individual branches (Fraedrich et al. 2008). A dark black discolouration is seen in the outermost sapwood and occurs as localised streak in branches and stems in the early stages of wilting (Fraedrich et al. 2008). Small beetle entrance holes (c. 0.75 mm in diameter) can be seen in branches and stems with sapwood discolouration; sapwood that is normally cream-white may have dark blueish-black streaks. Desiccation and browning of leaves soon follows (Fraedrich et al. 2008). Eventually, stem and limb dieback occur which can lead to tree decline and death. Following wilting and tree death, trees are repeatedly attacked by *X. glabratus* and other ambrosia beetles during spring, summer and autumn (Fraedrich et al. 2008). Wilting occurs because the woody xylem becomes discoloured and non-functional which causes foliage to die due to lack of water (Ploetz et al. 2017a).

Symptoms tend not to spread further than the hilum (scar on the seed where it was once attached to the mother plant) of fruit, but the fungus does colonise pedicels/peduncles (fruit stems) and hila (Eskalen and McDonald 2011; Ploetz et al. 2017a). *Raffaelea* infection does not spread into fruit pulp or seed (Ploetz et al. 2012).

Raffaelea lauricola is a unique ambrosia fungus in that it systemically infects tree xylem and a single infection can be fatal (Ploetz et al. 2017a). The fungus is able to move rapidly in the xylem of trees (Fraedrich 2020). *Raffaelea canadensis* has also been reported to cause wilt on avocado (Eskalen and McDonald 2011). In comparison, *R. aguacate* is not systemic nor pathogenic on avocado, but is closely related to *R. lauricola* (Ploetz et al. 2017a; Saucedo-Carabez et al. 2018). The primary vector of *R. aguacate* is not known, but it has been isolated from *X. bispinatus* infrequently suggesting *R. aguacate* plays a secondary role in the life cycle of *X. bispinatus* (Saucedo-Carabez et al. 2018). *Raffaelea campbellii* has been isolated from *X. glabratus* but its pathogenicity on *Persea americana* is not known (Simmons et al. 2016).

Time to symptom expression

Inoculated seedlings (through drill hole or slits) in the glasshouse exhibited sapwood discolouration and wilted or died within 8 weeks of inoculation (Fraedrich et al. 2008). External symptoms can develop after 3 weeks inoculation (Dreaden et al. 2017). In comparison, inoculated trees in the field wilted within 12 to 14 weeks after inoculation; sapwood discolouration was observed in all inoculated trees in the stems and branches (Fraedrich et al. 2008). Time to symptom development depends on age of the tree, growth conditions (glasshouse vs. field) and tree species (Dreaden et al. 2017). Latent infection of *R. lauricola*, of artificially inoculated avocado, is uncommon as the fungus is almost always isolated on media or detected via PCR from symptomatic sapwood (Ploetz et al. 2017b).

Reproduction and transmission

First inoculation is by an ambrosia beetle (Ploetz et al. 2017a). *Raffaelea* are asexual fungi that typically produces small conidiophores in sporodochia (clusters of conidiophores) within beetle galleries (Global Invasive Species Database 2021).

The pathogen is able to spread to other trees through root-grafts (Ploetz et al. 2017a). Mechanical transmission is possible through infested handsaws on potted plants; however, the pathogen does not survive on circular saws commonly used to prune avocado trees in commercial setting, most likely due to the high heat that is generated from these blades (Ploetz et al. 2017a). Transmission of the fungus is also human-mediated through the transport of wood or wood products (Ploetz et al. 2017a; Fraedrich 2020).

Table 5-25: Host range and geographic distribution of Raffaelea lauricola, R. campbellii, R. canadensi and R. aguacate

Region	Country	CMI	*Raffaelea* species	Vector	Host species	Reference
North America	USA	0.5-0.8	*Raffaelea lauricola*	*Xyleborus glabratus*	*Cinnamomum camphora*	Ploetz et al. (2017a)
					Persea americana	Ploetz et al. (2012)
					Persea borbonia	Harrington et al. (2011)
					Persea palustris	Harrington et al. (2011)
					Sassafras albidum	Harrington et al. (2011)
					Laurus nobilis	Hughes et al. (2014)
					Lindera benzoin	Ploetz et al. (2017a)
					Lindera melissifolia	Hughes et al. (2017)
					Litsea aestivalis	Hughes et al. (2017)
					Persea humilis	Ploetz et al. (2017a)
					Persea indica	Ploetz et al. (2017a)
					Umbellularia californica	Ploetz et al. (2017a)
					Licaria triandra	Ploetz et al. (2017a)
					Nectandra coriacea	Ploetz et al. (2017a)
			Raffaelea aguacate	*Xyleborus bispinatus*	*Persea americana*	Simmons et al. (2016); Saucedo-Carabez et al. (2018)
			Raffaelea campbellii	*Xyleborus glabratus*	*Persea palustris*	Simmons et al. (2016)
					Persea americana	(Simmons et al. 2016)
			Raffaelea canadensi	*Xyleborus glabratus*	*Persea americana*	Eskalen and McDonald (2011)
Asia*	Japan	0.6-0.8	*Raffaelea lauricola*	*Xyleborus glabratus*	Association recorded from beetle	Harrington et al. (2011)
	Myanmar	0.2-0.7	*Raffaelea lauricola*	*Xyleborus glabratus*	*Persea americana*	Ploetz et al. (2016)
	Taiwan	0.4-0.8	*Raffaelea lauricola*	*Xyleborus glabratus*	Association recorded from beetle	Harrington et al. (2011)

* *Xyleborus glabratus* is also found in India and China but no attempt to isolate *Raffaelea lauricola* has been made (Fraedrich 2020).

5.24.4 Likelihood of entry

Raffaelea spp. are found throughout southeast United States (Fraedrich 2020), Japan and Taiwan (Table 5-25). However, epidemics of laurel wilt disease caused by *R. lauricola* and *R. canadensi* have mainly been reported in the United States (Fraedrich et al. 2008; Eskalen and McDonald 2011;

Harrington et al. 2011; Hughes et al. 2014; Mayfield et al. 2019; Fraedrich 2020) and recently in Myanmar (Ploetz et al. 2016).

Infection of *Raffaelea* spp. is associated with ambrosia beetle infestation and with beetle galleries (Ploetz et al. 2017a). Specifically, *Raffaelea lauricola* is the fungal symbiont of the redbay ambrosia beetle, *Xyleborus glabratus* (Mayfield et al. 2019). *Raffaelea lauricola* and other *Raffaelea* spp. are also associated with other ambrosia beetles (Table 5-24, Table 5-25). *Raffaelea aguacate* is reported as not being pathogenic or systemic (Ploetz et al. 2017a; Saucedo-Carabez et al. 2018) and information regarding the pathogenicity of *R. campbellii* is lacking (Simmons et al. 2016). However, these species have been isolated from ambrosia beetles (Table 5-25).

The evidence demonstrates that *Raffaelea* species assessed here are ambrosia fungi and are associated with ambrosia beetle galleries (Olatinwo et al. 2021). Ambrosia fungi depend on and are cultivated by ambrosia beetles (Reverchon et al. 2021). However, there is some evidence that *Raffaelea* can also be mechanically transmitted through infected tools (Ploetz et al. 2017a). Irrespective of how *Raffaelea* is transmitted, symptoms appear within weeks (~3-8 weeks) of infection (Fraedrich et al. 2008; Dreaden et al. 2017). Therefore, if avocado budwood cuttings have an established *Raffaelea* infection, it is likely that beetle galleries and symptoms will be detected, and infected material removed or destroyed, during preparation for export, in phytosanitary inspections pre-export or on arrival in New Zealand.

The routine surface disinfection of budwood entering PEQ (dipping in 1% sodium hypochlorite for 2 minutes) will most likely not eliminate infection as some *Raffaelea* species are systemic (e.g. *R. lauricola*) (Ploetz et al. 2017a).

As part of the commodity description, all imported avocado germplasm will undergo a growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ. This period in 3A PEQ will impact on likelihood of entry in the following manner:

- ambrosia beetle galleries will be visible during inspection and thus infected budwood would be detected and destroyed before they are released from PEQ.
- *Raffaelea* is spread by ambrosia beetles. Budwood will be treated with miticides and insecticides (see section 2.2). Furthermore, Level 3A PEQ has measures in place to manage the risks associated with insects, wastewater, waste and accidental transfer (see section 2.3), therefore ambrosia beetles carrying *Raffaelea* are highly unlikely to escape level 3A PEQ to infect a suitable host.

Given the arguments and evidence above, that is:

- established *Raffaelea* infections are highly likely to be detected, and infected material removed or destroyed, during preparation for export, in phytosanitary inspections pre-export or on arrival in New Zealand;
- residual biosecurity risks from ambrosia entry points or recent infections on the surface of avocado budwood are likely to be managed by the growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ;

the likelihood of *Raffaelea* species entering New Zealand associated with *P. americana* budwood is considered to be negligible, with low uncertainty.

Therefore, further assessment is considered unnecessary at this time, and likelihoods of exposure and establishment, and impacts to New Zealand were not assessed.

5.24.5 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of *Raffaelea* species entering New Zealand associated with *P. americana* budwood (as described in the commodity description, and taking into account the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ) is considered to be negligible with low uncertainty;

- therefore, continuing the assessment was considered unnecessary; and

the overall level of assessed risk to New Zealand from *Raffaelea* species on imported *P. americana* budwood (as in the commodity description) is considered to be negligible, with low uncertainty.

5.24.6 Management considerations

At early stages of infection, trees exhibit wilting of green foliage of branch tips in sections of the tree canopy and wilting of individual branches (Fraedrich et al. 2008). A dark black discolouration is seen in the outermost sapwood and occurs as localised streak in branches and stems in the early stages of wilting (Fraedrich et al. 2008). Small beetle entrance holes (c. 0.75 mm in diameter) can be seen in branches and stems with sapwood discolouration; sapwood that is normally cream-white may have dark blueish-black streaks. Desiccation and browning of leaves soon follows (Fraedrich et al. 2008). Eventually, stem and limb dieback occur which can lead to tree decline and death. Following wilting and tree death, trees are repeatedly attacked by *X. glabratus* and other ambrosia beetles during spring, summer and autumn (Fraedrich et al. 2008). Wilting occurs because the woody xylem becomes discoloured and nonfunctional which causes foliage to die due to lack of water (Ploetz et al. 2017a).

Warmer temperatures, such as those in spring and summer, seem to exacerbate symptom development (Fraedrich 2020). *Raffaelea lauricola* can be detected with species specific primers (Dreaden et al. 2014; Wuest et al. 2017). Sequencing of rDNA has been used for other *Raffaelea* species such as *R. canadensis* (Eskalen and McDonald 2011).

5.24.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

At early stages of infection, trees exhibit wilting of green foliage of branch tips in sections of the tree canopy and wilting of individual branches (Fraedrich et al. 2008). A dark black discolouration is seen in the outermost sapwood and occurs as localised streak in branches and stems in the early stages of wilting (Fraedrich et al. 2008). Small beetle entrance holes (c. 0.75 mm in diameter) can be seen in branches and stems with sapwood discolouration; sapwood that is normally cream-white may have dark blueish-black streaks. Desiccation and browning of leaves soon follows (Fraedrich et al. 2008). Eventually, stem and limb dieback occur which can lead to tree decline and death. Following wilting and tree death, trees are repeatedly attacked by *Xyleborus glabratus* and other ambrosia beetles during spring, summer and autumn (Fraedrich et al. 2008). Wilting occurs because the woody xylem becomes discoloured and non-functional which causes foliage to die due to lack of water (Ploetz et al. 2017a).

Symptoms tend not to spread further than the hilum of fruit, but the fungus does colonise pedicels/peduncles and hila (Eskalen and McDonald 2011; Ploetz et al. 2017a). *Raffaelea* infection does not spread into fruit pulp or seed (Ploetz et al. 2012).

- ***What are the known environmental conditions conducive to symptom expression?***

Temperature and moisture conditions influence the rate of development of disease and subsequent symptoms (Fraedrich 2020). Late in the growing season, disease seems to progress relatively slowly. For example, trees with partial crown wilt (i.e., on only a few branches) are observed during winter months (Fraedrich 2020). In the spring and summer, when trees are actively transpiring and growing, the disease progresses much more rapidly and trees die quickly (Fraedrich 2020).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

There is no evidence that *R. lauricola* or *R. canadensis* cause asymptomatic infections; both species have been reported to cause laurel wilt disease (Eskalen and McDonald 2011). *Raffaelea aguacate* seems to behave as a saprotroph and is not pathogenic; it also does not occur systemically in the plant (Ploetz et al. 2017a; Saucedo-Carabez et al. 2018).

Raffaelea (specifically *R. lauricola*) is a systemic pathogen, thus sampling directly from wood tissue (after debarking) has been used to extract crude DNA from plant tissues (Hamilton et al. 2020).

Given all species will be associated with beetle galleries (Ploetz et al. 2017a), plant material from these areas can be used to be tested for infection.

- ***What is the optimum season for conducting detection testing?***

Rapid disease progression tends to occur in spring and summer, suggesting these seasons would be optimal for testing; in comparison, disease progression slows in winter (Fraedrich 2020).

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5.25 *Rhizoctonia noxia* (thread blight)

Rhizoctonia noxia is a fungal pathogen causing thread blight of coffee and many other plants including avocado. Damage from the disease typically occurrs on all parts of the plant except the roots.

5.25.1 Taxonomic description

Scientific name: *Rhizoctonia noxia* (Donk) Oberw., R. Bauer, Garnica, R. Kirschner 2013

Order/Family: Cantharalles/Ceratobasidiaceae

Other names include: *Ceratobasidium noxium* (Donk) P. Roberts 1999, *Corticium koleroga* (Cooke) Höhn. 1910, *Koleroga noxia* Donk 1958, *Pellicularia koleroga* Cooke 1876

Taxonomic notes: The species name *Pellicularia koleroga,* originally described by Cooke in 1876, was revisited von Höhnel in 1910, who assigned the taxon to genus *Corticium.* This was corroborated by Burt (1918). Later, Donk (1954) who disagreed with Cooke's genus *Pellicularia* assigned a new scientific name (*Koleroga noxia*) to the taxon. The reason for these shifts was likely due to the incorrect description of spores by Cooke (Burt 1918; Donk 1954), who probably observed basidia (spore bearing cells) and mistook them for basidiospores. Thus, the spore morphology descriptions in literature might be conflicting. Nevertheless, it is assumed that above mentioned names as used in the literature refer to the same taxon under the name *Rhizoctonia noxia*. However, there is a lack of DNA sequencing data available to confirm this (B. Weir, pers. comm.).

5.25.2 Hazard identification

Rhizoctonia noxia is not present in New Zealand:

- *Rhizoctonia noxia* is not listed in NZOR (2021), Biota NZ (2022) or PPIN (2021).
- The regulatory status of *R. noxia* has not been assessed (ONZPR 2021)

Rhizoctonia noxia has the potential to establish in New Zealand because:

- There are records of *R. noxia* in association with avocado, citrus and persimmon plants. These hosts are grown in New Zealand.
- the climate in New Zealand is unlikely to limit its development and spread because *R. noxia* has been reported to associate with plant hosts in areas that have a Climate Match Index (McMillan 1976) up to 0.7 indicating a similar climate to the whole of New Zealand (Phillips et al. 2018).

Rhizoctonia noxia has the potential to cause harm to New Zealand because:

- *Rhizoctonia noxia* causes thread blight in *P. americana* and other important crops such as *Citrus* spp.
- Avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$50.6 million (Plant & Food Research 2020).

Rhizoctonia noxia is associated with *P. americana* budwood because:

- *Rhizoctonia noxia* has been isolated from the leaves and braches of *P. americana* (avocado) plants.

Given the arguments and evidence above, *R. noxia* meets the criteria to be a hazard for New Zealand.

5.25.3 Risk assessment

Biology and epidemiology of Rhizoctonia noxia

Rhizoctonia noxia is known to cause thread blight on avocado in Brazil and Honduras, but details about the symptoms or the disease are not provided (Muller and Roberts 1961; McGuire and Crandall 1967; Lourd 1993). Nevertheless, thread blight in coffee and other tree hosts has been reviewed in detail by Dechassa (2019). Assuming the synonymy of *Corticium koleroga* with *Rhizoctonia noxia* (Biota NZ 2022), the description of the disease, including symptoms and transmission, is likely to apply to *P. americana* as a host too, albeit with increased uncertainty. Damage from the disease on coffee is described as occurring on all parts of the plant except the roots. It can occur in shaded and non-shaded coffee plantations at high and low altitudes. The disease appears to be supported by periods of high rainfall, mid altitudes and high humidity as well as shade provided through overhanging branches (Dechassa 2019).

Host range and distribution

The fungus has a cosmopolitan distribution and disease caused by the fungus has been described from many economically important crops such as coffee (Belachew et al. 2015), coconut (Benchimol et al. 2001), citrus, persimmon, tea, cacao, mango and many other species ranging from herbaceous monocts to woody fruit trees (Dechassa 2019).

The areas with reported disease on avocado caused by *R. noxia* have a lower climate matching index (Phillips et al. 2018) such as Brazil's amazon (Lourd 1993) with a CMI <0.5 and 0.5 in Honduras (Muller and Roberts 1961). However, the pathogen has a global distribution and in some regions where it is associated with coffee (e.g. Ethiopia), the climate matching index reaches 0.7 (Belachew et al. 2015; Phillips et al. 2018). Dechassa et al. (2020) report that the optimal temperature for growth of the pathogen is 25℃.

Symptoms

Rhizoctonia noxia typically infects plant tissues through stomata or lenticels but also through wounds. The fungus forms long, slender, white or pale mycelial strands (rhizomorphs) that usually spread along veins or ribs of leaves or along branches. These rhizomorphs then infect more leaves and branches. Infected leaves turn black initially at the petiole and the blackening later spreads to the leaf blade, predominantly on the underside. Affected leaves often wilt and sometimes separate at the petiole, but usually remain attached to the stem by fungal rhizomoprhs. Most mycelial formation is on the underside of leaves and between the cells of the leaf parenchyma (inner spongy leaf tissue) with minute fruiting bodies emerging at many points giving a mottled appearance to the leaf (Dechassa 2019). The symptoms can be quite severe and incidence high, especially if the disease is left unmanaged as was the case recently in Ethiopia. In experimental conditions symptoms develop within 10 days when infecting coffee twigs and leaves either by mycelial plugs or basidiospores (Dechassa et al. 2020).

No records were found in the available literature to suggest that *R. noxia* can infect asymptomatically. Searches were conducted in Google Scholar and Google with the search terms: “Ceratobasidium noxium”, “Corticium koleroga”, “Koleroga noxia”, “Rhizoctonia noxia”, “Pellicularia koleroga” each in combination with “endophyte” or “asymptomatic” e.g. [“Pellicularia koleroga” + endophyt*].

Transmission and lifecycle

Rhizoctonia noxia (*Corticium koleroga*) is an aerial pathogen that is transmitted by free water and splashing over short distances. The fungus can penetrate the host tissues through stomata in leaves, lenticels in bark or wounds and spread in the form of rhizomoprhs (mycelial threads) to infect more host tissue (Dechassa 2019).

The spread of the disease is assisted by wind, water, insects as well as mechanical means (Dechassa 2019). Dispersal over longer distances is assisted by humans, through the use of infected planting material. Over short distances the fungus spreads by growing mycelium, which allows it to infect new leaves and branches on the same tree or even between trees (Dechassa 2019).

Furthermore the fungus can form basidia during rainy weather and this leads to airborne release of basidiospores. Expected transmission distances through airborne basidiospores were not provided (Dechassa 2019). The fungus could also be spread through insect vectors in the genera *Antestiopsis* and *Usingeria* (Dechassa 2019), but neither of these are known to be present in New Zealand (NZOR 2021).

5.25.4 Likelihood of entry:

Rhizoctonia noxia is not likely to be imported into post-entry quarantine (PEQ) on avocado budwood due to conspicuous symptoms, unless an emerging infection was not visually detected prior to export, in which case the fungus would be likely to produce visible symptoms in PEQ.

Rhizoctonia noxia can infect stems through lenticels and wounds. *Persea americana* is a known host of *Rhizoctonia noxia* (Muller and Roberts 1961; McGuire and Crandall 1967; Lourd 1993), so association with leafless avocado budwood is considered to be possible.

Budwood cuttings are dipped in 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility, which would reduce the risk of importing spores and rhizomorphs of *R. noxia* into PEQ. However, mycelium growing in inner tissues might not be affected.

If the fungus is imported into PEQ, then three distinct phenological growing events referred to as ‘shoot flush’ is likely to be a sufficient time period for the fungus to develop visually detectable colonies on the surface infected budwood. No records of asymptomatic infections were found in the literature.

Basidiospores are small (9–13×3– 5 µm) (Dechassa 2019) and may fit through the mesh screen over the level 3A PEQ vents is required to have a maximum opening size of 0.04 mm2 (Chapter 2). However, it is likely that *R. noxia* would be detected by plant inspections before spores are produced because *R. noxia* typically produces visible symptoms such as bundles of mycelial strands growing on the surfaces of infected plant tissues, discolouration of plant tissues. In addition, production of spores is supported by rainy weather which and the spores are spread by wind and rain, and such conditions are unlikely to occur in PEQ.

Given the arguments and evidence above, the likelihood of *R. noxia* entering New Zealand associated with *P. americana* (avocado) budwood is considered to be negligible, with moderate uncertainty.

5.25.5 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of *R. noxia* entering New Zealand associated with imported *P. americana* (avocado) budwood is considered to be negligible with moderate uncertainty. Therefore, continuing the assessment is considered unnecessary; and

the overall level of risk to New Zealand from *R. noxia* on imported *P. americana* (avocado) budwood is considered to be negligible, with moderate uncertainty.

5.25.6 Management considerations

Rhizoctonia noxia produces consipcuous symptoms such as leaf wilting, tissue discolouration (blackening) mottleded appearance of underside of leaves upon fruiting, and mycelial strands (rhizomorphs) running along leaf ribs, petioles or branches .

Symptoms caused by *R. noxia* occur typically after rain. Although the fungus occurs in almost all continents, it has mostly been reported to associate with plant hosts in tropical climates (Dechassa 2019). No records of asymptomatic infections were found in the literature.

Information from avocado is rather scarce and most information about the pathogen comes from literature on coffee. Furthermore, Lourd (1993) reports the disease caused by *R. noxia* (in avocado in Brazil as not a major problem and when controlled by cultural practices (e.g. removing crop residues, good aeration in the orchards)).

5.25.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Rhizoctonia noxia is likely to produce white or pale mycelial strands (rhizomorphs) that usually spread along the veins or ribs of leaves or along branches, infecting leaves and causing discouloration (blackening) and rotting of tissues (Dechassa 2019).

- ***What are the known environmental conditions conducive to symptom expression?***

Humid conditions, rain and sheltering of plant material from sun are likely to induce symptom expression (Dechassa 2019).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested?***

Rhizoctonia noxia is likely to infect branches and leaves of avocado through lenticels or stomata respectively. The pathogen could also infect the plant tissue through fresh wounds so testing of these tissues is recommended.

- ***What is the optimum season for conducting detection testing?***

High rainfall and humidity is likely to promote growth of the pathogen (Dechassa 2019), increasing the chances of detection.

5.25.8 References

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5.26 *Rhizoctonia theobromae* (vascular-streak dieback)

Rhizoctonia theobromae is a leaf-penetrating, xylem-infecting, fungal pathogen that is the causal agent of vascular-streak dieback of cacao. One report was found of it causing vascular-streak dieback in avocado plants.

5.26.1 Taxonomic description

Scientific name: *Rhizoctonia theobromae* (P.H.B. Talbot & Keane) Oberw., R. Bauer, Garnica & R. Kirschner

Order/Family: Cantharalles/Ceratobasidiaceae

Other names include: *Oncobasidium theobromae; Ceratobasidium theobromae; Thanatephorus theobromae*

Common names: Vascular-streak dieback (VSD) of cocoa

Taxonomic notes: In a revision of the *Rhizoctonia* group, Roberts (1999) moved *Oncobasidium theobromae* into the genus *Thanatephorus*, therefore, renaming the fungus *Thanatephorus theobromae*. Although Samuels et al. (2012) agreed that the name *Oncobasidium theobromae* was not justified, they argued that *Oncobasidium theobromae* should instead be moved into the genus, *Ceratobasidium*, with a new name, *Ceratobasidium theobromae*. This new positioning was challenged by Oberwinkler et al. (2013) who argued that *Ceratobasidium theobromae* belonged rightfully in *Thanatephorus* (anamorph *Rhizoctonia*). Furthermore, Oberwinkler et al. (2013) suggested that the widely used name *Rhizoctonia* should be used in place of *Thanatephorus* and therefore suggested renaming *Oncobasidium theobromae*, *Rhizoctonia theobromae*.

5.26.2 Hazard identification

Rhizoctonia theobromae is not known to be present in New Zealand:

- *Rhizoctonia theobromae* is not listed in NZOR (2021), Biota NZ (2022) or PPIN (2021).
- The regulatory status of *Rhizoctonia theobromae* has not been assessed (ONZPR 2021).

Rhizoctonia theobromae is associated with *P. americana* budwood because:

- *Rhizoctonia theobromae* has been isolated from *P. americana* plants.
- *Rhizoctonia theobromae* is known to infect the vascular tissue inside stems.

Rhizoctonia theobromae has the potential to establish [and spread] in New Zealand because:

- *Rhizoctonia theobromae* is a well-known pathogen of cacao, and one record was found of it associating with avocado plants (Anderson 1989; Samuels et al. 2012). Avocado is grown mainly in the north of the North Island, and avocado plants are in commercial production in Northland and Bay of Plenty.
- The climate in New Zealand is unlikely to limit its development and spread because *R. theobromae* has been reported in areas that have a Climate Match Index (CMI) ≥ 0.7 (There are multiple, reliable records of *Akaropeltopsis* sp. causing sooty blotch on avocado in South Africa (Theron et al. 1981; Smith et al. 1985; Smith et al. 1987; Korsten et al. 1997).

There is little information on the distribution of *Akaropeltopsis* sp. elsewhere in the world, and the records of its distribution are largely unsubstantiated or unconfirmed (Theron et al. 1981; Smith et al. 1987; Queensland Government 2001), as follows:

- Sooty blotch on avocado in Australia has been reported to be caused by a range of causative agents, including *Stomiopeltis* sp. (Coates et al. 2001; Willingham 2002; Cooke et al. 2009), *Akaropeltopsis* spp./*Stomiopeltis* spp. complex (Queensland Government 2001), and multiple non-specified fungi (Allen 2004). Given that the causative agents of sooty blotch on avocado have been confused in the past due to the high level of morphological similarity between

Akaropeltopsis and *Stomiopeltis* (Theron et al. 1981; Smith et al. 1987), and that there were no records found describing *Akaropeltopsis* sp. on avocado in Australia, there is uncertainty regarding the presence of *Akaropeltopsis* sp. in Australia.

- *Akaropeltopsis* sp. has been reported to be present and associated with avocado in Colombia, but this is based on international literature compilation and not from direct observation. According to the USDA (2016), the presence of *Akaropeltopsis* sp. in Colombia is 'doubtful'.
    - *Akaropeltopsis* sp. appears to be similar to *A. marchaeriifolii* found on leaves of a *Machaerium* sp. in Brazil, but these species have not been confirmed to be conspecific (Theron et al. 1981; Smith et al. 1987). No other records were found in the literature of *Akaropeltopsis* sp. causing sooty blotch in Brazil.
    - Table 5-1), indicating a similar climate to the whole of New Zealand (Phillips et al. 2018).
    - *Rhizoctonia theobromae* needs a living host plant to survive and spreads via basidiospores. Basidiospores are short-lived (killed by direct and indirect sunlight), dispersed in the air for only short distances (~100m), and initiate the infection cycle when they land on young susceptible leaves (McMahon and Purwantara 2016).
- *Rhizoctonia theobromae* has the potential to cause harm to New Zealand because:
    - *Rhizoctonia theobromae* damages leaves and vascular tissue, resulting in leaf, stem and plant death. Although infections of avocado are apparently rare, avocados are an important commercial crop in New Zealand. In the 2018–2019 financial year, export sales of fresh avocados earned NZ$110 million and domestic sales earned NZ$ 56.5 million (Plant & Food Research 2019).

Given the arguments and evidence above, *R. theobromae* meets the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.26.3 Risk assessment

Biology and epidemiology

Rhizoctonia theobromae is a basidiomycete (Ceratobasidiales) fungal pathogen that is the causal agent of vascular-streak dieback of cacao. *Rhizoctonia theobromae* infects the leaves, and spreads into the vascular tissue of host plants and needs a living host plant to survive (McMahon and Purwantara 2016).

Hosts and Distribution

Rhizoctonia theobromae causes vascular-streak dieback in cacao (Malvaceae) plants in Asia and parts of Melanesia, but one report was found of it causing similar disease symptoms in avocado (Lauraceae) plants in Papua New Guinea (Anderson 1989; Samuels et al. 2012). Anderson (1989) and McMahon and Purwantara (2016) suggested that the fungus may occur as an innocuous endophyte or weak pathogen in unknown native host species in its natural distribution (Southeast Asia and parts of Melanesia), where cacao and avocado are introduced.

Table 5-26: Distribution of *Rhizoctonia theobromae* (EPPO 2011).

Continent	Country	Status	CMI*
Asia	China (Guangdong, Henan)	restricted distribution	0.7
Asia	India	restricted distribution	0.2-0.8
Asia	Indonesia	widespread	0.4-0.6
Asia	Malaysia		0.4-0.5
Asia	Myanmar		0.2-0.6
Asia	Philippines		0.3-0.6
Asia	Thailand		0.3-0.5
Oceania	Papua New Guinea		0.4-0.7

* (Phillips et al. 2018)

Disease symptoms

The disease symptoms of *R. theobromae* appear to be similar in avocado and cacao (Anderson 1989). Initial symptoms in avocado include chlorotic and necrotic spots on leaves. The spores of the fungus infect young unhardened leaves but leaf symptoms usually occur 3-5 months after infection, by which time the infected leaves will have aged and hardened, and young healthy looking flushes of leaves will have developed on the shoot (Samuels et al. 2012). The apparent absence of green spots on avocado leaves is a notable difference to the green spots that present on cacao leaves (Samuels et al. 2012; Perrine-Walker et al. 2018).

As the infection progresses, the vascular tissue becomes blackened and is visible in the leaf scars of abscised leaves, or as dark streaks in split stems. The bark can become rough due to swollen lenticels in response to stress. Severe infections result in defoliation, the death of branches and stems and often kill young seedings. White fruiting bodies develop on leaf scars and stems after several days of wet weather (Anderson 1989; Samuels et al. 2012; Perrine-Walker et al. 2018).

Wet conditions promote symptom development, and the disease is most common in tropical climates and wetter regions where annual rainfall exceeds 2,500 mm (Guest and Keane 2007).

Although there is no evidence that *R. theobromae* can infect avocado asymptomatically, Anderson (1989) and McMahon and Purwantara (2016) suggested that the fungus may occur as an innocuous endophyte or weak pathogen in unknown native host species in its natural range, where cacao and avocado are introduced.

Transmission and reproduction

Rhizoctonia theobromae is wind dispersed through basidiospores, which are the only form of inoculum. Basidiocarps only develop on fresh leaf scars during wet weather and release basidiospores in relatively low numbers during the night. Basidiospores are airborne but dispersal is limited because they lose viability after a few hours or after exposure to direct and indirect sunlight, indicating that spores are only capable of spreading the disease a short distance. Consequently, very few new infections occur beyond <80m from diseased trees (Perrine-Walker et al. 2018). The basidiospores land on and penetrate young, unhardened leaves, and from there the infection spreads into the vascular tissue (Samuels et al. 2012). *Rhizoctonia theobromae* causes defoliation and cracks in leaves and stems several months after the initial infection, and basidiocarps develop on the fresh scars during periods of high rainfall or humidity (Samuels et al. 2012).

The fungus is only capable of surviving in association with a living plant host and does not continue to sporulate or survive on dead branches (Samuels et al. 2012). There is strong evidence that *R. theobromae* is not seedborne (Anderson 1989). In cacao, basidiocarps do not form on budwood taken from infected branches, and grafts prepared from infected budwood do not take. Therefore, it is highly unlikely that infection will spread through grafting in cacao (Guest and Keane 2007), however, this has not been tested in avocado plants.

5.26.4 Likelihood of entry

Rhizoctonia theobromae is well-known to infect cacao plants but one report was also found of it infecting avocado plants (Anderson 1989).

New infections may not be detected prior to export as symptoms take several months to appear (Samuels et al. 2012). Furthermore, *R. theobromae* colonises the vascular tissue of hosts (Samuels et al. 2012), indicating that it could be present inside leafless avocado budwood in the absence of visible external symptoms. Surface disinfection (dipping in 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility) is not considered to be effective at managing internal infections in leafless avocado budwood cuttings.

In cacao, basidiocarps do not form on budwood taken from infected branches, and grafts prepared from infected budwood do not take. Therefore, it is highly unlikely that infection is spread through grafting in cacao (Guest and Keane 2007). However, this has not been tested in avocado.

If *R. theobromae* is imported into PEQ, then three distinct phenological growing events referred to as 'shoot flush' is likely to be a sufficient time period for the fungus to produce symptoms because symptoms have been reported to occur 3-5 months after infection (Samuels et al. 2012). The recommendation for cacao is to quarantine nursery stock for six months before distributing to other regions to avoid spread of *R. theobromae* (McMahon and Purwantara 2016). Although there is no evidence that *R. theobromae* can infect avocado asymptomatically, Anderson (1989) and McMahon and Purwantara (2016) suggested that the fungus may occur as an innocuous endophyte or weak pathogen in unknown native host species within its natural range, where cacao and avocado are introduced.

Fruiting bodies develop on leaf scars and cracks in leaves and stems, which are symptoms of the disease (Samuels et al. 2012). The fungus would not be given the opportunity to sporulate in PEQ because it is highly likely that symptoms such as leaf scars and cracks in leaves and stems would be detected during visual inspections, prior to the development of fruiting bodies. Furthermore, the lack of overhead watering in PEQ (MPI 2021) will prevent sporulation because the growth of fruiting bodies is dependent on wet foliage (Guest and Keane 2007). Therefore, it is considered highly unlikely that airborne spores would be produced in PEQ.

Given the arguments and evidence above, that is:

- *Rhizoctonia theobromae* has only been reported to infect cacao plants with the exception of one report found of it infecting avocado plants*,*
- new infections may not be detected prior to export as symptoms take 3-5 months to occur,
- *Rhizoctonia theobromae* could be present inside leafless avocado budwood and such infections are unlikely to be manged by surface disinfection,
- cacao budwood harvested from infected branches cannot be used for grafting because grafts do not take, meaning that infections are highly unlikely to be spread through grafting in cacao, however, this has not been tested in avocado,
- if *R. theobromae* is imported into PEQ then it is likely to produce visible symptoms during the period of containment,
- it is considered highly unlikely that airborne spores would be produced and escape PEQ via air vents,

the likelihood of *R. theobromae* entering New Zealand associated with *P. americana* (avocado) budwood is considered to be negligible, with low uncertainty.

5.26.5 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of *R. theobromae* entering New Zealand associated with *P. americana* budwood is considered to be negligible with low uncertainty. Therefore, continuing the assessment is considered unnecessary; and

the overall level of risk to New Zealand from *R. theobromae* on imported *P. americana* budwood is considered to be negligible, with low uncertainty.

5.26.6 Management considerations

Vascular-streak dieback, caused by *R. theobromae* is characterised by chlorotic leaf spots, defoliation, cracks in leaves and stems showing blackened vascular tissue and mortality in young seedlings. No evidence or records of asymptomatic *R. theobromae* infections were found in the literature, although Anderson (1989) and McMahon and Purwantara (2016) suggested that the fungus may occur as an innocuous endophyte or weak pathogen in unknown native host species in its natural range. These unknown species may act as an inoculum reservoir.

Symptoms are a key diagnostic indicator for *R. theobromae* infection because its fastidious nature makes it very hard to isolate and maintain in pure culture, and difficult to study (Samuels et al. 2012; Ali et al. 2019).

Currently, *R. theobromae* is identified based on PCR/sequencing of internal transcribed spacer (ITS) regions (Samuels et al. 2012; McMahon and Purwantara 2016). Sampling is likely best performed during prolonged warm and wet weather. The best plant material to test is the leaf petiole and the midrib of mature leaves, or vascular tissue from symptomatic plant material (Samuels et al. 2012).

Wet conditions are important factors for symptom development, reproduction and natural dispersal. The disease is most common in tropical climates and wetter regions where annual rainfall exceeds 2,500 mm (Guest and Keane 2007). The main commercial avocado production is centred within Northland, Auckland and Bay of Plenty where average annual rainfall ranges from about 1350 mm in Kaitaia to 1180 mm in Tauranga (NIWA 2021). The fungus requires a living host to survive and does not survive on dead plant material. The disease is not transmissible via seed, cuttings or grafting in cacao, but this has not been tested in avocado (Guest and Keane 2007).

No information was found in the literature on the management of vascular-streak dieback disease in avocado plantations but the management methods used in cacao plantations are likely to apply. Monitoring cacao planting materials for symptoms of vascular-streak disease before distributing is recommended to prevent the spread of the disease. The international movement of cocoa germplasm requires a pre-export quarantine period of 6 months (Perrine-Walker et al. 2018). In cacao, the disease can be effectively managed by implementing phytosanitary measures such as nursery hygiene, planting in new locations distant from older diseased plants or covering of plants in nurseries to exclude inoculum. Pruning to remove infected branches and to increase aeration and sunlight on the foliage will decrease sporulation and new infections. Some systemic fungicides have been shown to control vascular-streak dieback but are likely to be uneconomic compared to other management methods (Guest and Keane 2007; Perrine-Walker et al. 2018).

5.26.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

The disease symptoms of *R. theobromae* appear to be similar in avocado and cacao (Anderson 1989). Initial symptoms in avocado include chlorotic and necrotic spots on leaves. The spores of the fungus infect young unhardened leaves but leaf symptoms usually occur 3-5 months after infection, by which time the infected leaves will have aged and hardened, and young healthy looking flushes of leaves will have developed on the shoot (Samuels et al. 2012). The apparent absence of green spots on avocado leaves is a notable difference to the green spots that present on cacao leaves (Samuels et al. 2012; Perrine-Walker et al. 2018).

As the infection progresses, the vascular tissue becomes blackened and is visible in the leaf scars of abscised leaves, or as dark streaks in split stems. The bark can become rough due to swollen lenticels in response to stress. Severe infections result in defoliation, the death of branches and stems and often kill young seedings. White fruiting bodies develop on leaf scars and stems after several days of wet weather (Anderson 1989; Samuels et al. 2012; Perrine-Walker et al. 2018).

Although there is no evidence that *R. theobromae* can infect cacao or avocado asymptomatically, the fungus was assumed to live in unknown native host species in its natural range, in which it perhaps occurs as an innocuous endophyte or weak pathogen (Anderson 1989; McMahon and Purwantara 2016).

- ***What are the known environmental conditions conducive to symptom expression?***

Rhizoctonia theobromae causes disease in tropical climates in Asia and parts of Melanesia (Table 5-26). The rate of symptom development is closely correlated with rainfall during a few prior months and, the disease is most common in wetter regions where annual rainfall exceeds 2,500 mm (Guest and Keane 2007). The main commercial avocado production is centred within Northland, Auckland and Bay of Plenty where average annual rainfall ranges from about 1350 mm in Kaitaia to 1180 mm in Tauranga (NIWA 2021). Periods of high rainfall and humidity are required for the development of white fruiting bodies around leaf scars and on stems (Samuels et al. 2012).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

In cacao, vascular tissue may be colonised early on during infection (Samuels et al. 2012), therefore, testing vascular tissue might yield a better chance of detection than other parts of the plant. The emergence of spores appears to happen most frequently through fresh leaf scars and cracks in the petiole or midrib (McMahon and Purwantara 2016). This fastidious[49] pathogen is very hard to isolate and maintain in pure culture (Samuels et al. 2012; Ali et al. 2019).

- ***What is the optimum season for conducting detection testing?***

Rhizoctonia theobromae occurs in tropical climates in South East Asia (Table 5-26), suggesting that in New Zealand, the fungus is most likely to grow during summer, under warm and wet conditions.

High rainfall and humidity are required for the production of white fruiting bodies around leaf scars and on stems (Samuels et al. 2012), but in the absence of overhead irrigation the fungus is unlikely to sporulate in PEQ.

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[49] A fastidious organism has very complicated nutritional and/or environmental requirements and will not grow without specific factors present or in specific conditions.

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5.27 *Scolecobasidium musae* (sooty blotch and flyspeck (SBFS))

Scolecobasidium musae is an epiphytic fungus that has occasionally been reported to colonise the surface of banana and avocado plants. It is considered to be a part of the sooty blotch and flyspeck complex (SBFS) which contains more than 100 species in 30 genera of Ascomycota and Basidiomycota.

5.27.1 Taxonomic description

Scientific name: *Scolecobasidium musae* (G.Y. Sun & Lu Hao) Samerp. & de Hoog **Order/Family:** Venturiales / Sympoventuriaceae

Other names include: *Ochroconis musae* G.Y. Sun & Lu Hao and *Ochroconis mirabilis* Samerp. & de Hoog

Taxonomic notes: The genus *Scolecobasidium* was historically separated from *Ochroconis* based on morphological differences, but this distinction was not supported by subsequent molecular phylogenetic analyses. Therefore, *Scolecobasidium* and *Ochroconis* are currently considered to be synonymous (Shen et al. 2020).

5.27.2 Hazard identification

Scolecobasidium musae is not known to be present in New Zealand:

- *Scolecobasidium musae* is not listed in NZOR (2021), Biota NZ (2022) or PPIN (2021).
- The regulatory status of *S. musae* has not been assessed (ONZPR 2021).

Scolecobasidium musae has the potential to establish (and spread) in New Zealand because:

- There are two records of *S. musae* in association with banana plants, and one record where it is associated with an avocado plant. Both of these hosts are grown mainly in the north of the North Island, and avocado plants are in commercial production in Northland and Bay of Plenty.
- The climate in New Zealand is unlikely to limit its development and spread because *S. musae* has been reported in areas that have a Climate Match Index (CMI) ≥ 0.9, and reported to associate with plant hosts in areas that have a Climate Match Index (CMI) ≥ 0.7 (There are multiple, reliable records of *Akaropeltopsis* sp. causing sooty blotch on avocado in South Africa (Theron et al. 1981; Smith et al. 1985; Smith et al. 1987; Korsten et al. 1997).

There is little information on the distribution of *Akaropeltopsis* sp. elsewhere in the world, and the records of its distribution are largely unsubstantiated or unconfirmed (Theron et al. 1981; Smith et al. 1987; Queensland Government 2001), as follows:

- Sooty blotch on avocado in Australia has been reported to be caused by a range of causative agents, including *Stomiopeltis* sp. (Coates et al. 2001; Willingham 2002; Cooke et al. 2009), *Akaropeltopsis* spp./*Stomiopeltis* spp. complex (Queensland Government 2001), and multiple non-specified fungi (Allen 2004). Given that the causative agents of sooty blotch on avocado have been confused in the past due to the high level of morphological similarity between *Akaropeltopsis* and *Stomiopeltis* (Theron et al. 1981; Smith et al. 1987), and that there were no records found describing *Akaropeltopsis* sp. on avocado in Australia, there is uncertainty regarding the presence of *Akaropeltopsis* sp. in Australia.
- *Akaropeltopsis* sp. has been reported to be present and associated with avocado in Colombia, but this is based on international literature compilation and not from direct observation. According to the USDA (2016), the presence of *Akaropeltopsis* sp. in Colombia is 'doubtful'.
- *Akaropeltopsis* sp. appears to be similar to *A. marchaeriifolii* found on leaves of a *Machaerium* sp. in Brazil, but these species have not been confirmed to be conspecific

(Theron et al. 1981; Smith et al. 1987). No other records were found in the literature of *Akaropeltopsis* sp. causing sooty blotch in Brazil.

- Table 5-1) indicating a similar climate to the whole of New Zealand (Phillips et al. 2018).

Scolecobasidium musae has the potential to cause harm to New Zealand because:

- SBFS results in dark blemishes on the fruit, leaves and stems of host plants, but it does not cause any physiological damage to the underlying tissue (Hao et al. 2013). However, if symptoms occurred on avocado fruit, it could reduce the marketability and value of avocado fruit,
- avocados are an important commercial crop in New Zealand. In 2020, export sales of fresh avocados earned NZ$100.1 million and domestic sales earned NZ$50.6 million, while export sales of avocado oil earned $NZ9.3 million (Plant & Food Research 2020).

Scolecobasidium musae is associated with *P. americana* budwood because:

- it has been isolated from a trichome[50] of a *P. americana* leaf,
- no records were found of *S. musae* on stems of host plants, but SBFS fungi complex (of which is *S. musae* is a part of) is known to occur on stems,

Given the arguments and evidence above, *S. musae* meets the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.27.3 Risk assessment

Biology and epidemiology

Scolecobasidium musae is an epiphytic fungus that has been occasionally reported to colonise the surface of plants. For this reason, it is considered to be a part of the sooty blotch and flyspeck complex (SBFS), which contains more than 100 species in 30 genera of Ascomycota and Basidiomycota (Gleason et al. 2019). For this assessment, where specific information on *S. musae* was not available in the literature, information regarding SBFS complex in general was applied to *S. musae* as it is considered to be a member of the SBFS complex.

Species in the genus *Scolecobasidium* (syn. *Ochroconis*) are oligotrophs[51] and opportunistic pathogens. *Scolecobasidium* spp. have been isolated from water, soil, plants, domestic environments (bathrooms, laundries), skin on a human foot, cold blooded animals such as salmon and trout, and the cuticle of an ant species (Hao et al. 2013; Samerpitak et al. 2014).

Distribution and Host range

Scolecobasidium musae appears to be widespread in parts of Asia and Europe (Table 5-27). However, it has only been reported in association with plants under tropical conditions in China and Thailand (Table 5-27), and only reported to cause symptoms on the leaves (Crous et al. 2021) and fruit (Hao et al. 2013) of *Musa basjoo*. The limited information in the literature suggests that *S. musae* is either not common, or common but not frequently reported.

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[50] Trichomes are unicellular or multicellular hair-like appendages, that grow from the epidermal cells of a plant (Wang et al. 2021).

[51] Oligotrophs are organisms that can live in nutrient deficient environments.

Table 5-27. The potential geographic distribution of *Scolecobasidium musae*.

Region	Country	CMI[52] range	Host	References
Asia	China	0.4 – 0.9	*Musa* sp.	Hao et al. (2013); Crous et al. (2021)
	Thailand	0.2 – 0.5	*Persea americana*	Crous et al. (2019)
	India	0.2 – 0.9	unknown	Institute (2021)
Africa	Egypt	0.4 – 0.6	soil	Crous et al. (2021)
America	USA	0.5 – 0.9	unknown	Institute (2021)
Europe	Austria	0.7 – 0.9	unknown	Institute (2021)
	Denmark	0.8 – 0.9	unknown	Institute (2021)
	Germany	0.8 – 0.9	domestic	Crous et al. (2021)
	Netherlands	0.9	unknown	Institute (2021)
	Sweden	0.6 – 0.9	unknown	Institute (2021)

Symptoms

The fungi that cause SBFS are epiphytic and colonise the epicuticular wax layer of stems, twigs, leaves, and fruit (Gleason et al. 2019).

Scolecobasidium musae is slow growing and does not damage the underlying tissue (Gleason et al. 2019; Crous et al. 2021). It produces shiny, black, sclerotium-like mycelial colonies (Hao et al. 2013) that appear as a dark speckle on the leaves (Crous et al. 2021) and fruit (Hao et al. 2013) of *Musa basjoo*.

SBFS causes surface blemishes on hosts plants, however, there are no records in the literature of any significant disease problems caused by *S. musae*. Although *S. musae* was isolated from leaf trichomes of an avocado plant (Crous et al. 2019), no information was found in the literature on the symptoms, suggesting that it does not cause significant disease on avocado either.

Optimal conditions for growth and symptom development

SBFS occurs in summer in warm and moist conditions (Hao et al. 2013). *Scolecobasidium musae* has only been isolated from plant hosts in tropical climates in China and Thailand (Table 5-27). When cultured in-vitro, *S. musae* formed colonies 30mm in diameter after 20 days at 25 ℃ (Hao et al. 2013). No records of asymptomatic *S. musae* infections were found in the literature.

Reproduction and transmission

Scolecobasidium musae produces small conidia (6.5–8.5×2.0– 3.0 µm) that are most likely transported by wind and rain during the summer (Gleason et al. 2019). *Scolecobasidium* spp. are known to colonise a wide range of substrates including soil (Hao et al. 2013), indicating their potential to overwinter and spread on these substrates.

5.27.4 Likelihood of entry

There is potential for *S. musae* to be imported into post-entry quarantine (PEQ) on avocado budwood, in which case it would be likely to produce visible symptoms in PEQ.

Scolecobasidium musae has been isolated from plants in tropical regions including Thailand and China (Table 5-27).

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[52] Climate Match Index (CMI) with all of New Zealand (Phillips et al. 2018).

Scolecobasidium musae has only been recorded on leaves and fruit of host plants, but SBFS (caused by a large fungi complex that includes *S. musae*) is reported to occur on the leaves, stems and fruit of host plants (Gleason et al. 2019), so association with leafless avocado budwood is considered to be possible.

Budwood cuttings are dipped in 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility, which is considered to substantially reduce the risk of importing *S. musae* into PEQ.

If the fungus is imported into PEQ, then three distinct phenological growing events referred to as 'shoot flush' is likely to be a sufficient time period for the fungus to develop visually detectable colonies on the surface infected budwood. No records of asymptomatic infections were found in the literature.

Conidia are small (6.5–8.5×2.0– 3.0 µm) and may fit through the mesh screen over the level 3A PEQ vents is required to have a maximum opening size of 0.04 mm^2 (MPI 2021). However, it is likely that *S. musae* would be detected by plant inspections before spores are produced because SBFS fungi are typically slow growing and not likely to sporulate rapidly (Gleason et al. 2019). Furthermore, the literature suggests that conidia are spread by wind and rain, and such conditions are unlikely to occur in PEQ.

Given the arguments and evidence above, that is:

- *Scolecobasidium musae* has only been reported in association with plants in tropical climates (Table 5-27).
- *Scolecobasidium musae* could be present on the surface of budwood but such infections are highly likely to be manged by 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility.
- if *S. musae* is imported into PEQ then it is likely to produce visible symptoms during the period of containment.
- *Scolecobasidium musae* is unlikely to produce spores between inspections in PEQ. Even though *S. musae* spores are small enough to fit through the mesh over the vents of level 3A PEQ, windy and rainy conditions are likely required for transmission through the air and such conditions are unlikely to occur in PEQ.

the likelihood of *S. musae* entering New Zealand associated with *P. americana* (avocado) budwood is considered to be negligible, with low uncertainty.

5.27.5 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of *S. musae* entering New Zealand associated with *P. americana* budwood is considered to be negligible with low uncertainty. Therefore, continuing the assessment is considered unnecessary; and

the overall level of risk to New Zealand from *S. musae* on imported *P. americana* budwood is considered to be negligible, with low uncertainty.

5.27.6 Management considerations

Scolecobasidium musae is an epiphytic fungus that produces shiny, black, sclerotium-like mycelial colonies (Hao et al. 2013). They have been reported to appear as a conspicuous dark speckle on the leaves (Crous et al. 2021) and fruit (Hao et al. 2013) of *Musa basjoo*. Although *S. musae* was isolated from leaf trichomes of an avocado plant (Crous et al. 2019), no information was found in the literature on the symptoms on avocado, suggesting that it is not a major disease of avocado.

SBFS occurs in summer in warm and moist conditions (Hao et al. 2013). Although *Scolecobasidium musae* is distributed in both temperate to tropical climates, it has only been reported to associate with plant hosts in tropical climates (Table 5-27). No records of asymptomatic *S. musae* infections were found in the literature.

Field applications of strobilurin and succinate dehydrogenase inhibitor fungicides have been shown to be very effective against SBFS fungi (Gleason et al. 2019).

Post-harvest dip solutions, from chlorine bleach to sodium bicarbonate and various fruit soaps, followed by brushing, are effective at removing most sooty blotch and flyspeck colonies and symptoms (Gleason et al. 2019).

5.27.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

The fungi that cause sooty blotch and flyspeck (SBFS) are epiphytic53 and colonise the epicuticular wax layer[54] of stems, twigs, leaves, and fruit. Their dark-coloured colonies are conspicuous to the naked eye and for this reason, fungi in the SBFS complex are considered to be highly noticeable plant-associated microbes (Gleason et al. 2019). In particular, *S. musae* produces shiny, black, sclerotium[55]-like mycelial colonies (Hao et al. 2013) that appear as a dark speckle on the leaves (Crous et al. 2021) and fruit (Hao et al. 2013) of *Musa basjoo*. Although *S. musae* was isolated from leaf trichomes of an avocado plant (Crous et al. 2019), no information was found in the literature on the symptoms on avocado, suggesting that it is not a major disease in avocado.

- ***What are the known environmental conditions conducive to symptom expression?***

SBFS occurs in summer in warm and moist conditions. *Scolecobasidium musae* is distributed in a range of countries, from temperate to tropical climates, but it has only been reported in association with plants in tropical climates (Table 5-27). When cultured in-vitro, *S. musae* formed colonies 30mm in diameter after 20 days at 25℃ (Hao et al. 2013).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

SBFS typically occurs on the fruit, leaves and stems of hosts (Gleason et al. 2019). Only one record was found of *S. musae* on an avocado plant in the literature, where it was isolated from leaf trichomes in 2008 (Crous et al. 2019). No records of asymptomatic *S. musae* infections were found.

- ***What is the optimum season for conducting detection testing?***

SBFS is a late-season disease which occurs in moist and warm growing conditions during the summer (Hao et al. 2013).

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[53] An epiphytic fungus is a fungus that grows on the surface of a living plant.
[54] An epicuticular wax is a coating of wax covering the outer surface of the plant cuticle in land plants.
[55] A sclerotium is a hard, dense, compact mass of mycelium that varies in form and has a dark-coloured covering.

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5.28 Sooty moulds

Sooty mould is a generic name used to describe a wide range of fungus species that cover the surfaces of above ground plant parts with a black, sooty coating and are generally associated with the sugary exudates of sap-sucking insects. Heavy infestation may inhibit photosynthesis and the unsightly mould may degrade the market value of produce.

5.28.1 Taxonomic description

Table 5-28: Taxonomy of sooty mould species that may be associated with avocado budwood. Taxonomy determined from Index Fungorum (2022).

Scientific name	Order/Family	Other names
Antennulariella batistae S. Hughes	Capnodiales/ Antennulariellaceae	*Capnodendron trichomericola, Septonema perseae, Antennariella perseae, Capnodium capsuliferum*
Calothyrium apiahynum (Spegazzini) F. Stevens	Microthyriales/ Microthyriaceae	*Calothyriolum apiahynum, Calothyriolum aphiaphynum*
Capnodium spp. Mont.	Capnodiales/ Capnodiaceae	
Lembosia perseae Orejuela	Asterinales/ Asterinaceae	
Meliola antioquensis Orejuela	Meliolales/ Meliolaceae	
Asteridiella perseae (F. Stevens) Hansf.	Meliolales/ Meliolaceae	*Irene perseae*, *Meliola perseae*, *Irenina perseae*, *Meliola perseae* f. *setulifera*, *Appendiculella perseae*
Periconiella perseae McKenzie	Mycosphaerellales/ Mycosphaerellaceae	

Taxonomic notes:

The term "sooty moulds" is not clearly defined and refers to a polyphyletic group of fungi. Initially the term was used to described fungi in the genus *Fumago* (Chomnunti et al. 2014). The classification of these fungi varies considerably in the literature. Therefore, it is difficult to determine which species should be considered as sooty moulds. Sooty moulds are generally considered to be of the orders Capnodiales and Chaetothyriales (Chomnunti et al. 2014; Abdollahzadeh et al. 2020). Hongsanan et al. (2015) state that Asterdales and Meliolales are distinct from sooty moulds as some species may penetrate host cells. However, Bernal and Diaz (2006) consider that *Capnodium* spp., *Calothyrium aphiahynum*, *Lembosia perseae*, *Meliola antioquensis* and *Asteridiella perseae* found on avocado all share the characteristics of sooty moulds. The description of *Periconiella perseae* indicates this fungi is also superficial (McKenzie 1990) and is considered similar to sooty fungi. For the purposes of this PRA all the species listed in Table 5-28 are considered to be similar enough in biology and epidemiology to be assessed as a single group.

5.28.2 Hazard identification

The sooty mould species listed in Table 5-28 are not known to be present in New Zealand:

- *Antennulariella batistae, Calothyrium apiahynum, Lembosia perseae, Meliola antioquensis* and *Asteridiella perseae* are not listed in NZOR (2021), Biota NZ (2022), or ONZPR (2021).

- *Periconiella perseae* is recorded as absent from New Zealand in NZOR (2021) and Biota NZ (2022) and is listed as “regulated” in ONZPR (2021).
- It is unknown if species of *Capnodium* that infect avocado are present in New Zealand. *Capnodium* is a large genus of approximately 150 species, only six of which are reported present in New Zealand (*C. annonae*, *C. australe, C. citri, C. fibrosum, C. walteri* and *C. uniseptatum*) and one species has unknown biostatus (*C. sphaeroideum*).(NZOR 2021; Biota NZ 2022). Six species in the genus *Capnodium* are listed as “non-regulated” (*C. annonae, C. australe, C. citricolum, C. salicinum, C. uniseptatum,* and *C. walteri*), four species are “regulated” (*C. elaeophilum, C. mangiferum, C. citri* and *C. tanakae*) and one species is “under assessment” (*C. y fumaco*) in ONZPR (2021).

The sooty mould species listed in Table 5-28 have the potential to establish (and spread) in New Zealand because:

- These sooty mould species are typically distributed in tropical and sub-tropical regions (Chomnunti et al. 2014), so may find suitable environments in some regions of New Zealand.
- Avocado trees are grown commercially and in home gardens in some parts of New Zealand and would provide a host for mould establishment.

The sooty mould species listed in Table 5-28 have the potential to cause harm to New Zealand because:

- Infections by sooty moulds are predominantly superficial and rarely damage plant tissues, but they can reduce the photosynthetic capabilities of host plants (Chomnunti et al. 2014).
- Their presence on fruit may reduce market value of avocado and other potential hosts (Chomnunti et al. 2014).

The sooty mould species listed in Table 5-28 are associated with *P. americana* budwood because:

- They have been reported associated with above ground plant parts of avocado so propagules may be present on stems and around the bud on avocado budwood (Tamayo and Julián 2007; Unites States Department of Agriculture 2016).

Given the arguments and evidence above, the sooty mould species listed in Table 5-28 meet the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

5.28.3 Risk assessment

Biology and epidemiology of sooty moulds

Host range and Distribution

Some sooty moulds reported on avocados appear to have a limited host range, while the host range of other species may be much broader. These fungi typically develop on the exudates of sap sucking insects and it is unclear if association are insect or plant host specific (Chomnunti et al. 2014). Some sooty moulds are not found in association with insects and may instead grow on sap released from the leaves or stems through structures such as glandular trichomes (Hughes 1976). However, the sooty mould species investigated in this assessment have not been reported on avocado in the absence of insects.

Table 5-29: Host range of sooty moulds reported in association with avocado.

Species	Known hosts	References
Antennulariella batistae	35 host species are reported in 17 plant families. Hosts significant to New Zealand include *Persea americana* and *Citrus sinensis* (orange).	Hughes (2000)
Calothyrium apiahynum	*Persea americana, Persea sp., P. palustris*	Tamayo and Julián (2007)

Species	Known hosts	References
Capnodium spp.	*Persea americana* *Capnodium* species are reported on a wide range of host plants.	Tamayo and Julián (2007); Ramírez-Gil and Morales (2019)
Lembosia perseae	*Persea americana*, *Persea sp.*	Buritica (1999); Tamayo and Julián (2007)
Meliola antioquensis	*Persea americana*, *P. petiolaris*	Buritica (1999); Tamayo and Julián (2007)
Asteridiella perseae	*Persea americana*	Buritica (1999); Tamayo and Julián (2007)
Periconiella perseae	*Persea americana*	McKenzie (1990)

These sooty mould species are typically distributed in tropical and sub-tropical regions (Chomnunti et al. 2014), so may find suitable environments in some regions of New Zealand.

Table 5-30: Distribution of sooty moulds associated with avocado.

Species	Known distribution	References	CMI range
Antennulariella batistae	Americas: Bahamas, Belize, Brazil, Cuba, Jamaica, Panama, Puerto Rico, USA (Florida, Hawaii) Asia: Philippines Africa: South Africa	Hughes (2000)	0.4-0.8
Calothyrium apiahynum	Americas: Brazil, Colombia, USA (Florida)	Farr and Rossman (2021)	0.4-0.7
Capnodium spp.	Genus distributed worldwide. Reported on *P. americana* in Cuba, Venezuela and Zimbabwe.	Whiteside (1966); (Urtiaga 1986); Ramírez-Gil and Morales (2019); Farr and Rossman (2021)	0.5-0.7
Lembosia perseae	Colombia	Buritica (1999); Farr and Rossman (2021)	0.4-0.8
Meliola antioquensis	Colombia	Spaulding (1961); Buritica (1999); Farr and Rossman (2021)	0.4-0.8
Asteridiella perseae	Americas: Colombia, Dominican Republic, Panama, Puerto Rico, USA (Florida), Venezuela, Virgin Islands	Buritica (1999); Tamayo and Julián (2007); Farr and Rossman (2021)	0.3-0.8
Periconiella perseae	Oceania: Australia, Samoa, Vanuatu, Palau	McKenzie (1990); Farr and Rossman (2021); Biota NZ (2022)	0.4-0.9

Symptoms

The specific symptoms produced by sooty mould species are difficult to ascertain, as they are frequently comprised of a mixture of various fungal species (Hughes 2000; Chomnunti et al. 2014). Information regarding the symptoms observed on avocado are also limited:

- *Antennulariella batistae* predominantly occurs on the leaves of host plants, but specific symptoms on avocado are not reported (Hughes 2000).
- The symptoms of *C. apiahynum* and *L. perseae* on avocado are not specifically reported but sooty mould are generally described as causing a thin, black layer on the leaves, stems and fruits of avocado (Tamayo and Julián 2007). This layer is easily scrapped from the plant surface. It may result in stains to the surface of leaves or fruits.
- *Capnodium sp.* are reported causing sooty mould on *P. americana* in Zimbabwe following scale infestation (Mwenje et al. 2010). Infection is characterised by a black layer of mycelia on the upper surface of the leaf, or on stems and fruit (Ramírez-Gil and Morales 2019; Ramírez-Gil and Peterson 2019). Outbreaks are typically observed on adult avocado trees

but may also occur on seedlings (Ramírez-Gil and Morales 2019). This genus has also been implicated in causing pre-harvest soft rot of avocado fruit (Mwenje et al. 2010).

  - *Meliola antioquensis* is reported as causing large black patches on the leaves of *P. petiolaris* in Colombia (Spaulding 1961). Symptoms on *P. americana* have not been reported however it has been associated with this host (Table 5-30).
  - *Asteridiella perseae* is reported to cause a highly visible, dense, velvety covering on the surface of avocado fruit (Unites States Department of Agriculture 2016).
  - McKenzie (1990) described *P. perseae* as a superficial black mould growth on the under surface of avocado leaves. The leaves showed some reddish-brown discolouration. The conidiophores were observed to be concentrated to the mid-rib and veins of the leaves.

Optimal conditions for growth and symptom development

Sooty moulds are most prevalent in tropical, subtropical, and warm temperate regions (Chomnunti et al. 2014) and growth is favoured by humid conditions and the presence of sap-sucking insects (Tamayo and Julián 2007).

Reproduction and transmission

Sooty moulds are transmitted through wind, rain or insect dispersal of ascospores, conidia and hyphal fragments (Ploetz 2003; Chomnunti et al. 2014). The spores germinate once in contact with insect exudates (Chomnunti et al. 2014).

5.28.4 Likelihood of entry

Sooty moulds usually infect the leaves of hosts but can sometimes colonise the surface of leaf petioles and plant stems (Callan and Carris 2004), indicating they may be associated with leafless avocado budwood. New infections by sooty moulds may not be detected prior to export as the development of symptoms may be slow and dependent on environmental conditions (Callan and Carris 2004). Therefore, it is possible that propagules of sooty mould on the surface of avocado budwood may be imported into New Zealand and may not be evident during initial inspection.

Sooty mould infections are superficial and endophytism is not known for any of these species on avocado. Dalvi et al. (2002) showed that washing mango fruits with 0.05-0.1% sodium hypochlorite was effective at removing sooty moulds (Chomnunti et al. 2014). Treatment with sodium hypochlorite is also generally considered an effective method for removal of sooty moulds from avocado fruits (Ploetz 2003). Therefore, the risk of importing sooty mould propagules into PEQ after dipping avocado budwood in 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility is considered to be very low.

Sooty moulds are strongly associated with the presence of sap-sucking insects and proliferate in warm, humid conditions (Chomnunti et al. 2014). While quarantine facilities are like to maintain high temperatures, insects will not be present, and humidity may be variable. In the absence of suitable growth conditions, propagules are likely to die out (Prakash and Misra 2001), and would not be viable upon release of the plants from PEQ. If environmental conditions within PEQ are suitable, the period of PEQ is likely to be sufficient to produce conspicuous symptoms of sooty moulds on infected budwood.

Ascospores of *Capnodium* species may be as small as 10µm (Ramírez-Gil and Morales 2019) and spores of *Meliola* species are usually no longer than 10µm (Callan and Carris 2004). This indicates that spores of sooty moulds would likely fit through the mesh screen of Level 3A PEQ vents, which are required to have a maximum opening size of 40µm (MPI 2021). However, growth of sooty moulds is conspicuous and likely to be detected before spores are produced. Furthermore, windy and wet conditions, and potentially insect dispersal, are required to transport spores (Ploetz 2003; Chomnunti et al. 2014), and such conditions are unlikely to occur in PEQ.

Given the arguments and evidence above, that is:

- sooty moulds may be present on the surface of budwood imported into New Zealand, but such infections are highly likely to be manged by 1% sodium hypochlorite for 2 minutes upon entry into the quarantine facility,
- if sooty moulds are imported into PEQ, they may not produce visible symptoms during the period of containment but would become inviable,
- if growth of sooty moulds occurs the symptoms are highly likely to be detected and it is unlikely that spores will be produced prior to detection, and
- conditions aiding spore dispersal are unlikely to occur in PEQ,

the likelihood of the sooty mould species listed in Table 5-28 entering New Zealand associated with *P. americana* budwood is considered to be negligible, with low uncertainty. Therefore, further assessment is considered unnecessary at this time; likelihoods of exposure and establishment and impacts to New Zealand are not further assessed.

5.28.5 Overall level of associated risk to New Zealand

Based on the assessment of likelihood above, that:

- the likelihood of the sooty mould species listed in Table 5-28 entering New Zealand associated with *P. americana* budwood (as described in the commodity description, and taking into account the growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ) is considered negligible, with low uncertainty;

the overall level of risk to New Zealand from the sooty mould species listed in Table 5-28 on imported *P. americana* budwood is considered to be negligible, with low uncertainty.

5.28.6 Management considerations

Information is limited regarding the specific symptoms of the sooty mould species to deemed them as hazard on avocado. Generally, sooty moulds produce a conspicuous dense, black covering on the surfaces of above-ground plant parts (Tamayo and Julián 2007). The symptoms of the sooty mould species being assessed is not expected to differ from typical symptoms.

No reports were found in the literature of sooty moulds infecting hosts asymptomatically or endophytically. As these fungi are primarily superficial colonisers, they are only expected to be undetectable as propagules and during the earliest stages of infection. These fungi predominantly colonise leaves but may also be observed on stems, branches or petioles (Tamayo and Julián 2007). Mould growth may be observed on young or mature growth, but more growth may occur on the lower regions of plants where shading and humidity is greater (Tamayo and Julián 2007).

Any plant surface exhibiting the previously described symptoms can be sampled to identify the infection. Identification of sooty moulds is made primarily through morphological identification, and there are few sequences of sooty moulds available for molecular identification (Chomnunti et al. 2014). There are no sequences recorded in GenBank for any of the species of this assessment, but there are sequences of various *Capnodium* species available (Clark et al. 2016).

The sooty moulds in this assessment are distributed in warm and wet climates (Chomnunti et al. 2014). Mycelial growth is promoted by moisture (dew, rain, high humidity), shade and warm temperatures, as well as the presence of sap-sucking insects (Tamayo and Julián 2007; Chomnunti et al. 2014). The conditions conducive to symptom expression are the same conditions that promote the growth of the fungus, because the main symptom is the formation of black fungal colonies on the surface of the host. The environmental conditions of a PEQ facility may not be suitable for growth of sooty moulds. Humidity may be lower than is necessary, and sap-sucking insects are not expected to be present. However, if conditions are not conducive, it is expected that any propagules that are present would become inviable (Prakash and Misra 2001). Summer is likely the optimum season for detection of sooty moulds (Chomnunti et al. 2014). However, given the conspicuous symptoms of sooty moulds, growth at any time of year is likely to be detected by visual inspection.

Spores of sooty mould species are likely small enough to fit through the mesh screen of Level 3A PEQ vents (Callan and Carris 2004; Ramírez-Gil and Morales 2019), which are required to have a maximum opening size of 40µm (Ramírez-Gil and Morales 2019; MPI 2021). However, growth of sooty moulds is conspicuous and likely to be detected before spores are produced. Furthermore, windy and wet conditions, and potentially insect dispersal, are required to transport spores (Ploetz 2003; Chomnunti et al. 2014), and such conditions are unlikely to occur in PEQ.

5.28.7 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Information is limited regarding the specific symptoms of the sooty mould species deemed a hazard on avocado. Generally, sooty moulds produce a conspicuous dense, black, mycelial covering on the surfaces of above-ground plant parts (Chomnunti et al. 2014; Unites States Department of Agriculture 2016). The symptoms of the sooty mould species being assessed is not expected to differ from typical symptoms.

- ***What are the known environmental conditions conducive to symptom expression?***

The sooty moulds in this assessment are distributed in warm and wet climates (Chomnunti et al. 2014). Mycelial growth is promoted by moisture (dew, rain, high humidity), shade and warm temperatures, as well as the presence of sap-sucking insects (Chomnunti et al. 2014). The conditions conducive to symptom expression are the same conditions that promote the growth of the fungus, because the main symptom is the formation of black fungal colonies on the surface of the host.

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

No reports were found in the literature of sooty moulds infecting hosts asymptomatically. As these fungi are superficial colonisers, they are only expected to be undetectable as propagules and during the earliest stages on infection. These fungi predominantly colonise leaves but may also be observed on stems, branches or petioles (Unites States Department of Agriculture 2016).

- ***What is the optimum season for conducting detection testing?***

Summer is likely the optimum season for detection of sooty moulds due to warm temperatures and high humidity (Chomnunti et al. 2014). However, given the conspicuous symptoms of sooty moulds, growth at any time of year is likely to be detected by visual inspection.

5.28.8 References

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6. Pest risk assessments: Oomycetes

6.1 *Phytophthora mengei* (Phytophthora trunk canker)

Phytophthora mengei is a soil borne water mould (oomycete) that causes trunk cankers in avocado (*Persea americana*) plants. It can also infect the structural roots, lower limbs and fruit of the plant/tree.

6.1.1 Taxonomic description

Scientific name: *Phytophthora mengei* G.T. Brown, M.E. Gallegly & C.X. Hong 2009

Order/Family: Peronosporales/Peronosporaceae.

Other names include: *Phytophthora citricola* (prior to 2009, *P. mengei* was considered to be a subgroup of *P. citricola* that infected avocado).

Taxonomic notes: The pathogen was identified first as a *Pythiacystis* sp. A year later it was redescribed as *Phytophthora cactorum* (Lebert and Cohn) J. Schrot. *Phytophthora citricola* was separated from *P. cactorum* by Sawada in 1927. The avocado canker pathogen was subsequently identified as *Phytophthora citricola* (Sawada). In 1973, et al. recognised that the avocado canker pathogen was morphologically distinct from *P. citricola* (Hong et al. 2009).

Hong et al. (2009) recognised the avocado canker pathogen as a distinct species from *P. citricola* based on morphological and molecular differences and re-classified it as the novel species *Phytophthora mengei*. Literature published prior to 2009 that refers to *P. citricola* as a pathogen on avocado or other hosts is likely to correspond with *P. mengei*, as the literature suggests that *P. citricola* is not a pathogen of avocado. This creates uncertainty regarding the pathogen's distribution and host associations.

Phytophthora citricola was first identified in New Zealand in 1938 in hops. It has since been found on various hosts in New Zealand including avocado. Genetic analysis conducted by Scion and the Plant Health and Environment Laboratory (PHEL) showed that some isolates assigned as *P. citricola* were actually two recently described species within the *P. citricola* complex: *P. multivora* and *P. plurivora* (Romberg 2010). As there are no records of *P. mengei* in New Zealand, these records of *P. citricola* on avocado in New Zealand could refer to *P. multivora*, as it is a known pathogen of avocado (Farr and Rossman 2021).

There are 64 records of *Phytophthora* on avocado in PPIN (2021), 4 in STARS, 7 in LIMS (MPI 2021a), and 3 in Biota NZ (2022). The majority of these records are *P. cinnamomi*, followed by *P. cryptogea* and *P. megasperma*. The remaining are 3 records of *P. citricola* and 2 records of *Phytophthora* sp.. Among these 5 records, four records cannot be reassessed due to lack of cultures. The remaining record is genetically close to *P. drechsleri*, which is distinct from the *P. citricola* species complex. PHEL conducted DNA sequence analysis on published sequences of *P. mengei* from GenBank with DNA sequences from the PHEL database which contains 31k sequences that have been generated in the past 13 years. It contains many *P. citricola* sequences, but analysis shows that none of these are *P. mengei*. *Phytophthora mengei* sequences in GenBank are limited (~4) and are only from California, Mexico, and Guatemala. PHEL's analysis found no evidence to suggest that *P. mengei* is present in New Zealand and suggests that most records of *P. citricola* in New Zealand and other countries are unlikely to be *P. mengei* (W. Ho, pers. comm.).

6.1.2 Hazard identification

Phytophthora mengei is not known to be present in New Zealand:

- *Phytophthora mengei* is not listed in either Biota NZ (2022) or PPIN (2021).
- There is no entry for *Phytophthora mengei* in ONZPR (2021).

Phytophthora mengei has the potential to establish (and spread) in New Zealand because:

- avocado is grown commercially and domestically in northern parts of New Zealand. The climate in New Zealand is likely to be suitable for the reproduction and spread of *P. mengei. Phytophthora mengei* produces abundant chlamydospores, oospores and zoospores. Chlamydospores, oospores can survive in adverse environmental conditions for several years making it very difficult to eradicate. They are spread by watersplash, windblown rain, animals, insects, humans, equipment and vehicles (Marais et al. 2002; Marais 2004).

Phytophthora mengei has the potential to cause harm to New Zealand because:

- it causes trunk canker on avocado trees, resulting in reduced tree health, reduced productivity and eventual tree death. Fungicide applications can be used to prevent infection and reduce disease prevalence, but there is no way of eradicating the disease from an orchard (Marais 2004).
- Avocados are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$110 million and domestic sales earned NZ$56.5 million (Plant & Food Research 2020).

Phytophthora mengei may be associated with *P. americana* budwood because:

- *Persea americana* is a known host of *P. mengei* (Eskalen and Faber 2016).
- *Phytophthora mengei* infects woody tissue (Marais 2004).

Given the arguments and evidence above, *P. mengei* meets the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

6.1.3 Risk assessment

Biology and epidemiology of Phytophthora mengei

Host range

Phytophthora mengei is a pathogen of *Persea americana* (Eskalen and Faber 2016; Farr and Rossman 2021). There is one record of *Vigna unguiculata* (cowpea) as a host in Australia (Martin et al. 2014), but this is the only record of *Phytophthora mengei* infecting a host that is not in the *Persea* genus, as well as the only report of *P. mengei* in Australia, and should therefore be treated with caution.

Some of the newer clonal rootstocks which are resistant to Phytophthora root rot (caused by *Phytophthora cinnamomi*) such as Thomas, are susceptible to *P. citricola* (now *P. mengei*) (El-Hamalawi and Menge 1994; Marais 2004). Plants propagated as clones have more *Phytophthora* resistance than plants propagated from seed (Eskalen and Faber 2016).

Geographical distribution

Phytophthora mengei has a limited geographical distribution (Table 5-28)

Table 6-1: The geographical distribution of *Phytophthora mengei* (Farr and Rossman 2021).

Region	Country	State/Province (if recorded)	CMI[56] range
North America and the Caribbean	Guatemala		0.3–0.6
	Mexico		0.4–0.8
	USA	California	0.5–0.8
Oceania	Australia		0.4–1

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[56] Climate Match Index (CMI) with all of New Zealand (Phillips et al. 2018).

Phytophthora mengei is present in Mexico and the United States of America (California) (Table 5-28). There is some uncertainty associated with the records of in Australia and Guatemala. There is one record of *P. mengei* in Australia on *Vigna unguiculata* (cowpea) (Martin et al. 2014), but this is the only record of *P. mengei* infecting a non-*Persea* host, as well as the only report of *P. mengei* in Australia and should therefore be treated with caution (Table 5-28). Plant Health Australia considers *P. mengei* to be a high priority pest and states that it is not present in Australia and only present in USA and Mexico (Plant Health Australia 2021). *Phytophthora mengei* has also been reported in Guatemala, which is likely as it is present in Mexico (Table 5-28).

Symptoms

Phytophthora mengei causes cankers in the structural roots, base of the trunk and lower limbs of older host trees. Cankers grow slowly (over multiple years) and often go unnoticed until girdling has occurred and foliar dieback symptoms appear. Cankers are discoloured and can exude a red resinous, water-soluble exudate through cracks in the bark. This exudate dries to form a white crystalline deposit on bark. Cutting into a canker reveals a necrotic brown outer layer of wood instead of the normal white- or cream-coloured tissues and has a fruity odour when exposed to air. The cankers have distinct reddish-brown margins from which the fungus can readily be isolated (Marais 2004).

Foliar symptoms are similar to those of Phytophthora root rot (caused by *P. cinnamomi*). With Phytophthora trunk canker (caused by *P. mengei*), leaves retain their normal size and there is a gradual loss of leaves causing canopy thinning, whereas branch dieback (staghorning) is less typical. Unlike root rot, canker and collar rot affects the major tree roots, and the smaller feeder roots are usually still present. In mild to moderate canker stages the host tree foliage can appear healthy. Foliar dieback occurs when the canker progresses to a stage when it starts killing cambium and phloem tissues, effectively girdling the trunk (Marais 2004; Eskalen and Faber 2016).

The disease may infect low hanging fruit under some conditions, particularly in prolonged wet weather. Infected fruit have a distinct dark circular area that usually occurs at the lowest part of the fruit. Internally, the rot extends into the flesh, darkening it in the same pattern as the affected area on the surface (Marais 2004).

Reproduction and spread

Phytophthora mengei can propagate sexually or asexually, resulting in several types of structures that are specialised for survival, dispersal, or infection. Sexual reproduction requires both mating types and leads to the formation of thick-walled oospores that can survive for several years in harsh environments. In contrast, asexual reproduction involves the formation of sporangia and bi-flagellated wall-less zoospores (Hwu et al. 2017). The minimum temperature for colony growth is 6–10° C, the optimum about 25° C, and the maximum 30–33° C (Hong et al. 2009).

The development of lesions and reproductive and dispersive spores is favoured by high soil moisture levels, and dispersal is closely related to the movement of water. Sporangia develop on lesions and release mobile zoospores in moist or wet conditions. Zoospores are the main infective structures because they can swim through films of water on trees or in the soil, and can accumulate in puddles and ponds, making them easily dispersed by water splash (Martin et al. 2014).

Wounding is necessary for infection by *P. mengei* as spores cannot penetrate the bark or the structural barrier of lenticels (Shew and Benson 1981; Ouimette et al. 1988; El-Hamalawi and Menge 1994; Martin et al. 2014). Zoospores use chemotaxis to find wounds, and when they find a suitable infection site, they stops swimming, drop their flagella and develop a cell wall (encyst). Hyphae germinate from cysts, invading plant cells to obtain food. Once the pathogen infects the plant, it produces more chlamydospores, oospores, and/or sporangia, thus completing its life cycle (Parke 2016).

There are two types of thick-walled resting spores: vegetative chlamydospores and sexual oospores can survive adverse environmental conditions for several years and contribute to the pathogen's persistence in soils in the absence of hosts, making it very difficult to control (El-Hamalawi and Menge 1994; Bunny and Shearer 1995; Bradshaw et al. 2020).

The sugary exudate that is associated with cankers has been shown to be a source of inoculum as it contains zoospores, oospores and hyphal fragments. Canker/lesion exudate contaminates the soil around the base of the tree. Ants were shown to be attracted to the sugary exudate and could possibly transmit the pathogen. Snails were also identified as a possible vector as their faeces were shown to contain viable propagules that were not only resistant to, but stimulated to germinate by the digestive secretion of the snail (El-Hamalawi and Menge 1996).

One of the primary ways in which the disease is spread is through the trade of infected nursery stock. Nurseries that do not take precautions to disinfect water used for irrigation purposes, sterilize potting media and keep containers off the ground, are responsible for disseminating the disease throughout the industry (Marais 2004). The pathogen can also be spread via contaminated pruning tools, harvesting equipment and vehicles, on the shoes of pickers climbing into the trees, by storms and water movement, and by insects and animals (El-Hamalawi and Menge 1994; El-Hamalawi and Menge 1996; Marais 2004).

In avocado orchards, the removal of sucker shoots from the crown is common practice. This may be a major source of new infection and spread because contaminated equipment will injure and inoculate host trees with the pathogen (El-Hamalawi and Menge 1994). The use of irrigation systems is another source of disease, as cankers and lesions caused by *P. mengei* often occur on the side of trunks wetted by irrigation sprinklers. Spore spread and disease development are favoured by wet conditions (Marais 2004).

Time to symptom development

In one greenhouse trial, lesions developed on 5-1 0 month-old avocado plants in under 40 days from inoculation of susceptible avocado tissues in suitable environmental conditions (El-Hamalawi and Menge 1994). In another trial, lesions developed on 2-year old avocado plant stems within 4 weeks of inoculation (El-Hamalawi and Menge 1996). Foliar dieback symptoms only occur in late stages of canker development when the pathogen starts killing cambium and phloem tissues and disrupting the transportation of nutrients (Marais 2004).

There is no specific information available in the literature regarding whether an avocado host can be infected by *P. mengei* and remain asymptomatic. A range of *Phytophthora* species are reported to colonise and survive in symptomless or tolerant host plants, for example *P. rubi* and *P. idaei* (Schlenzig and Chard 2010), *P. cryptogea* (Lyubenova et al. 2015), *P. erythroseptica* (Peters et al. 2005), *P. cinnamomi* (Jacobs et al. 1997; Crone et al. 2013), *P. infestans* (Hussain et al. 2013) *P. inundata* ((Parkunan et al. 2010), *P. ramorum* and *P. kernoviae* (Denman et al. 2009; Thompson et al. 2021). Migliorini et al. (2015) tested potted ornamental plants in the EU using qPCR and detected *Phytophthora* in 87% of the tested pots and in 70% of the asymptomatic potted plants. Potted plants in soil carried several *Phytophthora* species without showing any external symptoms. Some *Phytophthora* sp. can remain viable within the xylem for a number of years (Brown and Brasier 2007).

6.1.4 Likelihood of entry

There is potential for *P. mengei* to be imported into post-entry quarantine on *P. americana* nursery stock because *P. mengei* can be present on leafless, rootless budwood cuttings and is reported in several countries that have a significant avocado industry:

- It is possible that budwood could be harvested from infected trees because cankers often go unnoticed for several years, but it is unlikely that infected budwood would be harvested because infections are usually localised in the trunk, collar and low hanging branches (Marais et al. 2002). If sanitary procedures are not followed during budwood harvesting then there may be opportunity for budwood to be infected because *Phytophthora mengei* infects wounds and can be transmitted by humans and contaminated equipment (Marais et al. 2002).
- *Phytophthora mengei* causes cankers and lesions on woody avocado stems, therefore it can be associated with leafless, rootless avocado budwood cuttings. However, such cankers and lesions are highly likely to be visually detected on budwood during preparation for export or on arrival in New Zealand.

- *Phytophthora mengei* is present in Mexico, the United States of America (California) and Guatemala. There is one unreliable record of *P. mengei* in Australia (Table 5-28).

If *P. mengei* is imported on avocado budwood, the residual biosecurity risks may not be managed by the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ because:

- There is no specific evidence for asymptomatic *P. mengei* infections or longer latent periods in avocado plants. El-Hamalawi and Menge (1994) showed that lesions on avocado stems caused by *P. mengei* infection developed within 40 days from inoculation, which would be enough time to develop, be detected by visual inspection and destroyed before they are released from PEQ. However, some *Phytophthora* species such as *P. cinnamomi* can infect hosts asymptomatically (Crone et al. 2013). *Phytophthora citricola* has been shown to infect the xylem of some hosts, although xylem infections did not extend further than 10cm from the source infection (a visible phloem lesion). Some *Phytophthora* sp. can remain viable within the xylem for a number of years (Brown and Brasier 2007). If *P. mengei* is capable if infecting the xylem, and if the budwood is harvested from within close proximity to a lesion, there is a chance that it could be imported in the xylem of budwood and not develop external symptoms for several years. The fact that *Phytophthora* pathogens can be present without visible symptoms, provides reason to suggest that *P. mengei* may not be managed by PEQ in some circumstances.

Given the arguments and evidence above, that is:

- *Phytophthora mengei* can infect wounds in woody tissue. It is likely that symptoms (lesions) will develop and be detected in Level 3A PEQ. Although there is no evidence of asymptomatic *P. mengei* infections or longer latent periods in avocado plants, other *Phytophthora* pathogens can be asymptomatic or infect the xylem of hosts. Such infections may not be visually detectable,

the likelihood of *P. mengei* entering New Zealand associated with *P. americana* budwood is considered to be low, with moderate uncertainty.

6.1.5 Likelihood of exposure

This assessment is made on the basis that *P. mengei* has entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *P. mengei*.

If avocado plants are infected with *P. mengei* at the time of their release from post entry quarantine they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments above, the likelihood of exposure of *P. mengei* in New Zealand from *P. americana* budwood is considered high, with low uncertainty.

6.1.6 Likelihood of establishment

This assessment is made on the basis that *P. mengei* has been successfully exposed to a suitable host plant in the New Zealand environment.

Suitable hosts for *P. mengei* are widespread in some areas of New Zealand. Commercial and domestic avocado production is concentrated in Northland and Bay of Plenty. Of the 3,795 hectares that are planted in commercial avocados, 1,647 are in Northland and 1,834 in the Bay of Plenty.

There is smaller scale commercial production in Auckland, Waikato, Gisborne, Hawke's Bay, Taranaki, Manawatu-Wairarapa, Wellington and Tasman/Nelson (Plant & Food Research 2019).

The required temperature range required for *P. mengei* colony growth is a minimum of 6-10℃, optimum of 25℃ and a maximum of 30-33° C (Hong et al. 2009). The mean annual temperature of Northland and Bay of Plenty is between 14-16℃. Typical summer daytime maximum air temperatures range from 22℃ to 26℃ in Northland (MPI 2021b). During summer the Bay of Plenty region experiences average daily air temperatures over 20℃ (MPI 2021b). This indicates that *P. mengei* could grow in New Zealand temperatures.

Northland and Bay of Plenty both receive abundant rainfall within the range of 1-2.2 metres median annual total rainfall (Chappell 2013b, 2013a), which would create favourable conditions for disease development and spread of *P. mengei*.

It is highly likely that there will be suitable environmental conditions for *P. mengei* to produce spores and infect avocado hosts year-round. Furthermore, *P. mengei* has been recorded in avocado in California, USA (Eskalen and Faber 2016) which has a CMI of ≥0.7, indicating a similar climate to New Zealand (Phillips et al. 2018). Marais (2004) reported that *P. citricola* (now *P. mengei*) is more of a problem on avocado in the cooler coastal production areas of California, which is more similar to the New Zealand climate than other areas of California.

Once *P. mengei* is established in New Zealand, the long-lived resting spores of *P. mengei* could continue to provide an ongoing source of inoculum for several years, making it very difficult to eradicate (El-Hamalawi and Menge 1994; El-Hamalawi and Menge 1996; Marais 2004).

Host availability is likely to be the main limit on the extent of spread of *P. mengei* in New Zealand, given that *P. americana* is the only reliably reported host (see 'Host range' section). *Phytophthora mengei* is likely to be able to survive and reproduce in any region of New Zealand where avocado plants can survive.

Given the arguments and evidence above,

- avocado is common in many regions of New Zealand and is the only known host of *P. mengei*.
- the climate, temperature and rainfall in New Zealand's avocado growing regions are favourable for *P. mengei* growth and reproduction.
- the high levels of rainfall in Northland and Bay of Plenty would facilitate disease development and spread.
- once *P. mengei* is established in New Zealand, the long-lived resting spores of *P. mengei* could continue to provide an ongoing source of inoculum for several years, making it very difficult to eradicate;

the likelihood of *P. mengei* establishing in New Zealand is considered high with low uncertainty.

6.1.7 Impacts in New Zealand

Phytophthora mengei causes cankers in the structural roots, base of the trunk and lower limbs of older host trees. Cankers grow slowly (over multiple years) and often go unnoticed until girdling has occurred and foliar dieback symptoms appear. *Phytophthora mengei* can propagate sexually or asexually, resulting in several types of structures that are specialised for survival, dispersal, or infection. The development of lesions and reproductive and dispersive spores is favoured by high soil moisture levels, and dispersal is closely related to the movement of water. Sporangia develop on lesions and release mobile zoospores in moist or wet conditions. Zoospores are the main infective structures because they can swim through films of water on trees or in the soil, and can accumulate in puddles and ponds, making them easily dispersed by water splash (Martin et al. 2014). Wounding is necessary for infection by *P. mengei* as spores cannot penetrate the bark or the structural barrier of lenticels (Shew and Benson 1981; Ouimette et al. 1988; El-Hamalawi and Menge 1994; Martin et al. 2014).

Economic impacts

Avocado (*P. americana)* is the only reliably documented host of *P. mengei.* However, *P. mengei* was only differentiated from *P. citricola* in 2009, so it is probable that many isolates that were called *P. citricola* have not been reidentified as *P. mengei.* Given this single host association, the economic impacts of *P. mengei* are considered to be limited to the avocado industry and communities in avocado production areas, with moderate uncertainty. Impacts on the avocado industry are likely to be moderate for the following reasons:

Phytophthora mengei was reported as the second most limiting pathogen to avocado production in California, behind *Phytophthora cinnamomi* (Marais et al. 2002). *Phytophthora mengei* may cause similar losses to the avocado industry as Avocado Sun Blotch viroid disease (Marais 2004). *Phytophthora mengei* is apparently present in 90% of avocado orchards in California, but the disease only occurs in approximately 5% of these orchards, compared to *P. cinnamomi* which appears to cause disease in 60 to 75% of orchards (Marais 2004). The difference between the two *Phytophthora* species in regard to infection rate and subsequent disease in orchards may be due to the fact that *P. mengei* requires wounds to infect the above ground parts of healthy trees, whereas *P. cinnamomi* is primarily a root pathogen and can infect roots in the absence of wounds (Marais 2004).

Chemical treatments that are used to manage *P. cinnamomi* and other *Phytophthora* pathogens already present in New Zealand, are also likely to be effective against *P. mengei*, so the additional costs for chemical treatment of *P. mengei* may not increase significantly. On the other hand, damage caused by *P. cinnamomi,* animals and tree pruning has been shown to facilitate *P. mengei* infection of avocado trees (Ouimette et al. 1988). Furthermore, *P. mengei* infects the trunk, collar and low hanging branches of avocado trees whereas *P. cinnamomi* is primarily a root pathogen, suggesting that new pest management practices may need to be implemented to manage *P. mengei* if it were to establish. Some of the newer clonal rootstocks which are resistant to Phytophthora root rot (caused by *P. cinnamomi*) such as Thomas, are susceptible to *P. citricola* (now *P. mengei*) (El-Hamalawi and Menge 1994; Marais 2004).

Phytophthora mengei can infect low hanging fruit (within 1 meter from the ground) in prolonged wet conditions, but this is of relatively minor importance in California, and is not likely to be a major factor in New Zealand.

The introduction of *P. mengei* could result in market access restrictions for exports of fresh avocados to some countries. Australia is currently the main export market for New Zealand avocados, importing approximately 80% of export grade fruit. Australia considers *P. mengei* to be a high priority (Plant Health Australia 2021). The remainder is exported to Japan, USA and South East Asian markets (New Zealand Avocado 2021).

In the 2018–2019 financial year, 31424 tonnes of avocados were produced in New Zealand. Export sales of fresh avocados in 2018–2019 earned NZ$110 million and domestic sales earned NZ$56.5 million. In addition, avocado oil earned NZ$10 million (combined domestic and export sales) (Plant & Food Research 2019).

Given the arguments and evidence above, that is:

- *Phytophthora mengei* was reported as the second most limiting pathogen to avocado production in California, behind *Phytophthora cinnamomi* (Marais et al. 2002). Chemical treatments that are used to manage *P. cinnamomi* and other *Phytophthora* pathogens already present in New Zealand may also manage *P. mengei*, which may mitigate some of the impacts associated with its establishment in New Zealand,
- The presence of *P. mengei* in New Zealand could result in market access restrictions for export since this pathogen is reported to infect fruit (Marais et al. 2002). Export sales of fresh avocados in 2018–2019 earned NZ$110m (Plant & Food Research 2019),

the economic impact of *P. mengei* to New Zealand is considered to be moderate, with low uncertainty.

Environmental impacts

There are five indigenous New Zealand plant species in the Lauraceae family (three endemic and two non-endemic) (Webb et al. 1988). However, no records were found of *P. mengei* infecting any species outside of the *Persea* genus, apart from in *Vigna unguiculata* (cowpea) (see 'Host range' section). Since the Lauraceae is a widespread family, including other important plant species and *P. mengei* is not reported from other genera in the family, the likelihood that it poses a risk to indigenous New Zealand plant species is considered to be low.

Given the arguments and evidence above and assuming that the pathogen is restricted to lauraceous hosts, the impact on the environment from the establishment of *P. mengei* in New Zealand is considered to be low, with moderate uncertainty.

Human Health impacts

No evidence for human health impacts from *P. mengei* was found:

- Searches using the terms 'Phytophthora mengei' or 'Phytophthora citricola' in Pubmed, Google Scholar, CAB abstracts and Google found no mention of adverse effects on humans and animals.

Given the arguments and evidence above, the human health impact of *P. mengei* to New Zealand is considered to be negligible with low uncertainty.

Sociocultural impacts

Avocados are widely consumed in New Zealand. In some parts of the country avocado trees are common in domestic gardens. Trunk canker may increase production costs for growers, but it is unlikely to cause scarcity in local markets.

Given the arguments and evidence above, the social/cultural impacts of *P. mengei* to New Zealand are considered to be low, with low uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, health and society is considered to be moderate, with moderate uncertainty.

6.1.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above and in table 1, that:

- the likelihood of entry is low
- assuming entry, the likelihood of exposure is high
- assuming successful exposure, the likelihood of establishment is high
- the overall impact on the New Zealand economy, environment, health and society is considered to be moderate

the overall level of assessed risk to New Zealand from *P. mengei* on imported *Persea americana* budwood (as in the commodity description) is considered to be moderate, with moderate uncertainty.

6.1.9 Management considerations

Phytophthora mengei causes cankers in the structural roots, base of the trunk and lower limbs of host trees (Marais et al. 2002). Cankers grow slowly (over multiple years) and often go unnoticed until girdling has occurred and foliar dieback symptoms appear (Zentmyer et al. 1974). Cankers are discoloured and can exude a red resinous, water-soluble exudate through cracks in the bark. This exudate dries to form a white crystalline deposit on bark. Cutting into a canker reveals a necrotic brown outer layer of wood instead of the normal white or cream coloured tissues and has a fruity odour when exposed to air. The cankers have distinct reddish-brown margins from which the fungus can readily be isolated (Marais et al. 2002).

Leaf loss and canopy thinning are typical symptoms of *P. mengei* infections. Leaves retain their normal size and the smaller feeder roots are usually still present. In mild to moderate canker stages the host tree foliage can appear healthy. Foliar dieback occurs when the canker progresses to a stage when it starts killing cambium and phloem tissues, effectively girdling the trunk. Affected trees show a gradual loss of vigour, decline of the top, and death in severe cases.

Environmental stress may predispose avocado trees disease caused by *P. mengei*. Under field conditions, stress of avocado plants could be attributed to factors such as water deficiency, salinity, excess fertilization, low temperatures, or root-rot disease caused by *P. cinnamomi* which affects about 60-75% of the acreage in southern California alone (Ouimette et al. 1988). The infection rate and canker development is much higher in stressed plants compared to non-stressed plants (El-Hamalawi and Menge 1994).

Similarly, increased levels of amino acids in the phloem tissue correlate positively with the rate of disease development (El-Hamalawi and Menge 1994). High nitrogen levels have been shown to correlate with increased lesion size, more lesions and host death in rhododendrons with *Phytophthora* dieback (Hoitink et al. 1986).

Disease development and spore spread is favoured by wet conditions. Cankers that are frequently exposed to water develop at an increased rate and intrude deeper into the phloem than cankers that are not frequently exposed (El-Hamalawi and Menge 1994). Cankers caused by *Phytophthora mengei* often occur on the side of trunks wetted by irrigation sprinklers. Furthermore, canker development and infection (production of sporangia, zoospores etc) is favoured by wet or moist conditions. The minimum temperature for colony growth is 6–10 ° C, the optimum about 25 ° C, and the maximum 30–33 ° C (Hong et al. 2009).

Phytophthora mengei is transmitted via contaminated pruning equipment. There is a chance for infection to occur during harvesting of budwood if the harvesting methods are not hygienic. Testing any fresh or historical wounds on the budwood may detect the presence of the pathogen before symptoms develop.

Confirming the identity of these Phytophthora requires culturing and/or molecular diagnostic tests. Current molecular detection tests for the genus *Phytophthora* are specific to several key species rather than the entire genus (Pegg et al. 2016).

It is very difficult to eradicate *Phytophthora* from an orchard once it is established in the soil. However, certain phosphonate fungistatics (phosphorous acid and phosphonate compounds) may increase the resilience of avocado trees to *Phytophthora* infection. They can be applied to the bark, to areas where cankers have been removed, injected into the trunk, or into the soil if the canker extends below ground (Eskalen and Faber 2016).

6.1.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Phytophthora mengei (formerly recognised as the 'avocado subgroup' within *P. citricola*) causes cankers in the structural roots, base of the trunk and lower limbs of older host trees. Cankers grow slowly (over multiple years) and often go unnoticed until girdling has occurred and foliar dieback symptoms appear. Cankers are discoloured and can exude a red resinous, water-soluble exudate through cracks in the bark. This exudate dries to form a white crystalline deposit on bark. Cutting into a canker reveals a necrotic brown outer layer of wood instead of the normal white- or cream-coloured tissues and has a fruity odour when exposed to air. The cankers have distinct reddish-brown margins from which the fungus can readily be isolated (Marais et al. 2002; Marais 2004).

Foliar symptoms are similar to those of *Phytophthora* root rot (caused by *P. cinnamomi*). With *Phytophthora* trunk canker (caused by *P. mengei*), leaves retain their normal size and there is a gradual loss of leaves causing canopy thinning, whereas branch dieback (staghorning) is less typical. Unlike *Phytophthora* root rot, *Phytophthora* trunk canker affects the major tree roots, and the smaller feeder roots are usually still present. In mild to moderate canker stages the host tree foliage can appear healthy. Foliar dieback occurs when the canker progresses to a stage when it starts killing cambium and phloem tissues, effectively girdling the trunk. Affected trees show a gradual loss of vigour, decline of the top and subsequent death.What are the known environmental conditions conducive to symptom expression?

Environmental stress may predispose avocado trees to disease. Under field conditions, stress of avocado plants could be attributed to factors such as water deficiency, salinity, excess fertilization and low temperatures (Ouimette et al. 1988). The infection rate and canker development are much higher in stressed plants compared to non-stressed plants (El-Hamalawi and Menge 1994).

Similarly, increased levels of amino acids in the phloem tissue correlate positively with the rate of disease development (El-Hamalawi and Menge 1994). High nitrogen levels have been shown to correlate with increased lesion size, more lesions and host death in rhododendrons with *Phytophthora* dieback, caused by multiple *Phytophthora* species (Hoitink et al. 1986).

Disease development and spore spread is favoured by wet soil conditions. Cankers that are frequently exposed to water develop at an increased rate and intrude deeper into the phloem than cankers that are not frequently exposed (El-Hamalawi and Menge 1994). Cankers caused by *Phytophthora mengei* often occur on the side of trunks wetted by irrigation sprinklers. Furthermore, canker development and infection (the production of sporangia, zoospores, oospores and chlamydospores) is favoured by wet or moist conditions. The minimum temperature for colony growth is 6–10 ° C, the optimum about 25 ° C, and the maximum 30–33 ° C (Hong et al. 2009).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Phytophthora mengei is a canker pathogen that infects wounds on woody tissue and is known to be transmitted by contaminated pruning equipment. There is a chance for infection to occur during harvesting of budwood if the harvesting methods are not hygienic. Testing any fresh or historical wounds on the budwood may detect the presence of the pathogen before symptoms develop.

- ***What is the optimum season for conducting detection testing?***

P. mengei infections last over multiple seasons and years. Canker/lesion development and infection is favoured by wet/moist conditions. The minimum temperature for colony growth is 6–10 ° C, the optimum about 25 ° C, and the maximum 30–33 ° C (Hong et al. 2009).

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6.2 *Phytophthora palmivora* (bud rot of palms)

Phytophthora palmivora is an oomycete (or water mould) that causes a range of rot, canker and dieback symptoms on various host tissues.

6.2.1 Taxonomic description

Scientific name: *Phytophthora palmivora* (E.J. Butler) E.J. Butler (1919)

Order/Family: Peronosporales/Peronosporaceae

Other names include: *Phytophthora arecae, Phytophthora cactorum* var. *arecae, Phytophthora faberi, Phytophthora hevae, Phytophthora omnivore* var. *arecae, Phytophthora palmivora* var. *heveae, Phytophthora palmivora var. theobromae, Phytophthora theobromae*, *Pythium palmivorum* (CABI 2018).

Taxonomic notes:

Phytophthora palmivora was originally identified as *P. omnivora*, the causal agent of black pod disease of cocoa (*Theobroma cacao)*, and as *Pythium palmivorum*, the cause of destructive diseases in palms. It was renamed *P. palmivora* by Butler in 1919 (Cooke et al. 2000; Cline et al. 2008).

6.2.2 Hazard identification

Phytophthora palmivora is not known to be present in New Zealand.

- *Phytophthora palmivora* is listed as absent from New Zealand in Biota NZ (2022). There are no records for *P. palmivora* in PPIN (2021).
- *Phytophthora palmivora* is a quarantine pest, regulated and unwanted (ONZPR 2021).

Phytophthora palmivora has the potential to establish (and spread) in New Zealand because:

- *Phytophthora palmivora* has a very wide host range, and known host plants including avocado (Farr and Rossman 2021), are widely grown in New Zealand, both commercially and in domestic gardens.
- *Phytophthora palmivora* causes disease in tropical and subtropical countries with high rainfall. Climate matching and climate modelling suggest that it is likely to have the ability to infect plants and spread in northern New Zealand, especially in the summer months between December and March (Figure 6-1). *Phytophthora palmivora* may be able to persist in the environment over winter months as resistant spores.
- However, disease expression and spread are very likely to be limited by climate, particularly in the south of New Zealand (Figure 6-1).

Phytophthora palmivora has the potential to cause harm to New Zealand because:

Phytophthora palmivora infects 130 to 1000 species of crop, ornamental, shade and hedge plants, including tomato, avocado, macadamia, potatoes and orchids (Farr and Rossman 2021).

- It causes a range of symptoms/diseases on a wide range of hosts in humid subtropical and tropical areas of the world (Drenth and Sendall 2004). It has been reported killing kiwifruit plants in Turkey (Çiftçi et al. 2015) and is relatively common as the causal agent of black rot of economically important orchids in Taiwan (Erwin and Ribeiro 1996).

Phytophthora palmivora is associated with *P. americana* budwood.

- *Persea americana* is a known host of *P. palmivora*.

- *Phytophthora* species normally attack the root system and stem base of the plant, but they are able to infect many different tissues (fine roots, bark, stems, leaves, shoots and fruits) on different host species (Tkaczyk et al. 2016)

Given the arguments and evidence above, *P. palmivora* meets the criteria to be a hazard for *P. americana* budwood imported to New Zealand.

6.2.3 Risk assessment

Biology and epidemiology of Phytophthora palmivora

Life cycle

Phytophthora are fungus-like eukaryotic microorganisms that all share similar life cycles (Pegg et al. 2016). They produce several types of structures adapted to survival in diverse environments and in different seasons. All species can rest for long periods (years) in the soil by producing thick-walled chlamydospores (asexual) and oospores (sexual) or as mycelial fragments. In the presence of water spores germinate to form sporangia on leaves and roots of host plants. In some *Phytophthora*, sporangia can detach and be blown or splashed in water to healthy plants. Sporangia release motile zoospores, which can swim through thin films of water on leaf surfaces and water-logged soil. Zoospores are short lived and susceptible to drying. In water-logged soil, zoospores are attracted to plant roots, and form short-lived resting cysts on them. Cysts germinate, forming hyphae, which then invade plant cells to infect the plant, producing more chlamydospores, oospores, and/or sporangia, thus completing the life cycle (Drenth and Guest 2004).

Phytophthora palmivora is heterothallic, and mating types are both found in many areas of the world (Zentmyer 1988).

Host range

It is not known for certain how many hosts *P. palmivora* can infect but the literature reports from 130 to 1000 different host species can be affected (Daniel and Guest 2008; Farr and Rossman 2021), including: *Adiantum raddianum* (maidenhair fern); *Anacardium occidentale* (cashew nut), *Ananas comosus* (pineapple), ***Annona cherimola* (cherimoya)**, *Antirrhinum majus,* ***Areca catechu* (betelnut palm)**, *Areca lutescens, Artocarpus altilis* (breadfruit), ***Borassus flabellifer* (toddy palm)**, *Capsicum annuum, Cattleya* sp., ***Carica papaya* (papaya)**, *Citrus limon, C. aurantiifolia, C. sinensis, C.* x *paradisi (grapefruit),* ***Cocos nucifera* (coconut palm)**, *Colocasia* sp., *Crotalaria* sp., *Cymbidium* sp., *Dendrobium* sp., *Dianthus caryophyllus, Dieffenbachia* spp., ***Durio zibethinus* (durian)**, *Elaeis guineensis* (African oil palm), *Ficus carica* (fig), *Fuchsia magellanica, Gossypium hirsutum* (cotton)*, Grevillea* sp.*, Hedera helix* (English ivy), ***Hevea brasiliensis* (rubber)**, *Hibiscus* sp., *Howea* sp., *Lavandula* sp., *Lycopersicon esculentum, Macadamia integrifolia, Magnolia grandiflora, Manihot esculenta* (cassava), *Mangifera indica* (mango), *Manilkara zapota* (sapodilla), *Myristica fragrans* (nutmeg), *Oncidium, Paphiopedilum* sp., *Paulowonia, Persea americana* (avocado), *Petunia violacea, Phaseolus* sp., *Philodendron* sp., *Piper nigrum* (black pepper), *Rhopalostylis baueri, Solanum tuberosum, Syzygium paniculatum,* ***Theobroma cacao* (cocoa)**, *Vanda spp.* (Farr and Rossman 2021).

Major hosts of *P. palmivora* are: betelnut palm, cherimoya, cocoa, coconut palm, durian, papaya, rubber and toddy palm (scientific names in bold above) (Farr and Rossman 2021).

Distribution

Phytophthora palmivora has a very wide geographical distribution (Table 6-2).

Table 6-2: The geographic distribution of *Phytophthora palmivora.*

Continent/Region	**Country/Area** (source CPC (2020), unless otherwise noted)	**CMI[57] range**
Africa	Angola, Cameroon, Central African Republic, Democratic Republic of the Congo, Republic of the Congo, Côte d'Ivoire, Egypt [**Note 1**], Equatorial Guinea, Gabon, Ghana, Liberia, Madagascar, Malawi, Mauritius, Morocco, Nigeria, Réunion, São Tomé and Príncipe, Senegal, Seychelles, Sierra Leone, Somalia, Tanzania, Togo, Uganda, Zimbabwe	0.3-0.8
Asia	Afghanistan, Brunei, Cambodia, China (Beijing, Fujian, Guangdong, Hainan, Jiangsu, Yunnan, Zhejiang), India, Indonesia, Iran, Japan (Kyushu, Shikoku), Jordan, Korea, Lebanon, Malaysia, Myanmar, Pakistan, Philippines, Singapore, Sri Lanka, Taiwan, Thailand, Turkey, Viet Nam	0.2-0.9
Europe	France, Greece, Italy, Norway, Poland, Spain (incl. Canary Islands)	0.7-0.9
North and Central America	Belize, Cayman Islands, Costa Rica, Cuba, Dominica, Dominican Republic, El Salvador, Grenada, Guatemala, Haiti, Honduras, Jamaica, Mexico, Nicaragua, Panama, Puerto Rico, Saint Kitts and Nevis, Saint Lucia, Saint Vincent and the Grenadines, Trinidad and Tobago, United States (Arizona, California, Florida, Hawaii, North Carolina, Tennessee, Virginia)	0.3-0.8
Oceania	American Samoa, Australia [**Note 2**], Cook Islands [**Note 3**], Fiji, French Polynesia, New Caledonia, Northern Mariana Islands, Papua New Guinea, Samoa, Solomon Islands [**Note 4**], Tonga, Vanuatu	0.4-0.7
South America	Argentina, Bolivia, Brazil (Alagoas, Bahia, Espírito Santo, Maranhão, Pará, Paraná, São Paulo), Colombia, Ecuador, Guyana, Peru, Suriname, Venezuela	0.3-0.9

Note 1. Regarded as a regulated non-quarantine pest by Egypt (EPPO 2020).
Note 2. *Phytophthora palmivora* is present in Australia, including Western Australia (Barber et al. 2013) where it was previously considered to be absent and was treated as a quarantine pest (Biosecurity Australia 2005).
Note 3. Reported from the Cook Islands (Dingley et al. 1981).
Note 4. Reported from Solomon Islands (Newhook and Jackson 1977).

Symptoms

Phytophthora palmivora is partially adapted to infect aerial portions of many of its hosts, in contrast to species such as *P. cinnamomi*, which is primarily a root and soil inhabiting pathogen. This allows it to infect and cause symptoms on various tissues on the same host (Erwin et al. 1995).

Phytophthora palmivora has been reported to cause a range of symptoms in *P. americana* including seedling blight, leaf infections and dieback of young stems (Zentmyer 1976), crown and root rot (Kurbetli et al. 2020) and cankers (Drenth and Guest 2004).

Leaf lesions are reddish-brown-black, enlarge rapidly along the larger veins and frequently cause contortion of the leaves. Dark, sunken, elongated lesions develop on young stems, and terminal buds are often killed. Conidia can develop in necrotic lesions on the leaf (Zentmyer 1976).

Symptoms disease occurs occasionally in the summer months, in periods of heavy rainfall and high humidity (Zentmyer 1976).

Asymptomatic infections

A range of *Phytophthora* species are reported to colonise and survive in symptomless or tolerant host plants, including *P. rubi* and *P. idaei* (Schlenzig and Chard 2010), *P. cryptogea* (Lyubenova et al. 2015), *P. erythroseptica* (Peters et al. 2005), *P. cinnamomi* (NCSU 2016), *P. infestans* (Hussain et al. 2013), *P. inundata* (Parkunan et al. 2010), *P. ramorum* and *P. kernoviae* (Denman et al. 2009). Migliorini et al. (2015) tested potted ornamental plants in the EU using qPCR, and detected

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[57] Climate Match Index (CMI) with all of New Zealand (Phillips et al. 2018).

Phytophthora in 87% of the tested pots and in 70% of the asymptomatic potted plants. Potted plants in soil carried several *Phytophthora* species without showing any external symptoms.

There is no information about the asymptomatic infection of avocado plants but *P. palmivora* was occasionally recovered from the roots of asymptomatic windmill palm (*Trachycarpus fortunei*) (Cacciola 2011), so asymptomatic infections are assumed to be possible.

Reproduction and transmission

Phytophthora species spread by the movement of infected soil, irrigation/rainwater and plant fragments, and by human activities involving the movement of these commodities and infected plants. Insects such as ants, beetles and flies are considered the main vectors of Black Pod disease of cocoa, caused by *P. palmivora* (Agrios 2008).

Low population levels of *P. palmivora* can persist in the soil. In citrus orchards, fallen fruit attracts *P. palmivora* from the soil, and sporulation is profuse under moist conditions. Sporangia are caducous[58] and can be dispersed at least 0.5m by rain splash (Martin et al. 2012). Fruits approaching maturity are infected by the sporangia and zoospores this way.

Although fresh wounds and stomata are considered key infection courts, *P. palmivora* is capable of direct penetration of leaf and fruit tissues (Drenth and Guest 2004). Secondary infections are then caused by sporangia spread through the tree by splashing water, windblown rain and, less commonly, invertebrates, such as snails and ants (Taylor and Griffin 1981; Graham and Menge 2000). According to Timmer et al. (2000), rain drops would be sufficient to propel small sporangium-filled droplets into the air where they could be carried some distance by wind, but sporangia are not dispersed by wind currents alone (as are sporangia of some *Phytophthora* species, e.g. *P. infestans*). Most infected fruit soon abscise but harvested fruit may not show symptoms until after they have been held in storage for a few days (Graham and Menge 2000).

Soil populations are maintained by repeated infections of the fibrous roots of host plants and *P. palmivora* can persist in unfavourable conditions for some time as chlamydospores (Graham and Menge 2000).

Phytophthora palmivora's ability to infect aerial portions of many of its hosts may theoretically allow spores to spread more easily via wind and windborne rain in contrast to *Phytophthora* species that are limited to the roots of hosts, such as *P. cinnamomi.* This allows it to infect and cause symptoms on various tissues on the same host, meaning that inoculum and susceptible host tissue are continuously available, and disease potential is always present (Erwin et al. 1995; Drenth and Guest 2004).

Climate

Phytophthora palmivora is a warm-climate species, historically restricted in distribution to tropical or subtropical regions. However, extrapolations from other hosts in other parts of the world and recent climate match assessments conducted by MPI (Appendix 6.2.12) suggest that this species is likely to establish in northern New Zealand, including avocado growing regions. *Phytophthora palmivora* has been reported on avocado in Florida, China, Honduras, Philippines, Thailand and Turkey (Farr and Rossman 2021). Spore spread is heavily reliant on water, and disease outbreaks often co-occur with rainy periods or waterlogged areas (Drenth and Guest 2004).

Optimal growth temperature

Phytophthora palmivora is a warm-climate species. Reported temperature optima for sporulation, infection and disease development and mycelial growth range from 24 to 30℃ as follows:

- the optimum for mycelial growth was reported as 30℃ by (Timmer et al. 2000), with little or no growth at 15 or 33℃ (Zitko et al. 1991),
- Timmer et al. (2000) found the optimum temperatures for infection and disease development on citrus fruit were 27 to 30℃, and that wetness of varying duration

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[58] They become dislodged readily and separated from the sporangiophore and therefore are readily dispersed.

depending on the pathosystem was required for infection to take place. Brown rot caused by *P. palmivora* on citrus fruit did not develop at 22℃ or below,

- the optimum temperature for sporulation in culture and on the fruit surface was 24℃, with sporangium production decreasing rapidly at higher or lower temperatures (Timmer et al. 2000). This optimum is considerably lower than the optimum for growth *in vitro* of 30℃, or for infection and disease development of 27 to 30℃ (Zitko et al. 1991; Timmer et al. 2000).

6.2.4 Likelihood of entry

There is potential for *P. palmivora* to be imported into post-entry quarantine on *P. americana* nursery stock because:

Phytophthora palmivora is known to infect and cause symptoms on various tissues (Zentmyer 1976), including stems, therefore, it could be associated with leafless, rootless *P. americana* budwood cuttings.

Phytophthora palmivora has a wide global distribution (Table 6-2), though it may not be reported from *P. americana* or as causing disease on *P. americana* in those countries:

- *Phytophthora palmivora* is present in Australia, Brazil, China, the Cook Islands, Egypt, Fiji, Japan, Korea, Mexico, New Caledonia, Samoa, Solomon Islands, Spain, Peru, the United States, Vanuatu, Viet Nam and other countries listed in Table 6-2.

If *P. palmivora* is imported on avocado budwood, the residual biosecurity risks may not be managed by the proposed growing period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’ in 3A PEQ because:

- There is no specific evidence for asymptomatic *P. palmivora* infections or longer latent periods in *P. americana*, however, *P. palmivora* was occasionally recovered from the roots of asymptomatic windmill palm (*Trachycarpus fortunei*) (Cacciola 2011), so asymptomatic infections are assumed to be possible.
- A range of *Phytophthora* species are are reported to colonise and survive in symptomless or tolerant host plants (Denman et al. 2009; Parkunan et al. 2010; Schlenzig and Chard 2010; Hussain et al. 2013; Lyubenova et al. 2015; NCSU 2016).
- Timmer et al. (2000) and Zitko et al. (1991) showed that mycelium grew above 15℃, but disease only developed above 22℃, highlighting a temperature range where infection could occur with suppressed disease symptoms. If symptoms are produced then they are highly likely to be visually detected on budwood during preparation for export, in phytosanitary inspections prior to export or on arrival in New Zealand.
- *Phytophthora palmivora* has been identified at the New Zealand border approximately 14 times, all either on nursery stock (*Dracaena, Epipremnum*) or on unknown pathways (MPI 2021). It has also been detected post border in 2018, in association with ornamental plants imported from Malaysia. The affected plants *Epipremnum*, *Philodendron* and *Ficus*) were destroyed in post-entry quarantine (MPI internal report).

Given the arguments and evidence above, that is:

- *Phytophthora palmivora* has a wide geographical range that overlaps with *P. americana.*
- *Phytophthora palmivora* can infect stems, indicating that it could be associated with budwood.
- Infections could be asymptomatic or not visually detectable if suitable environmental conditions are not met in PEQ.

the likelihood of *P. palmivora* entering New Zealand associated with *P. americana* budwood is considered to be low, with moderate uncertainty.

6.2.5 Likelihood of exposure

The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *P. palmivora*.

If avocado plants are infected with *P. palmivora* at the time of their release from Post Entry Quarantine they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments above, the likelihood of exposure of *P. palmivora* in New Zealand from *P. americana* budwood is considered high, with low uncertainty.

6.2.6 Likelihood of establishment

Assuming it has entered undetected and been successfully exposed to a suitable host plant, *P. palmivora* may establish in New Zealand because:

- The optimum temperature for sporangium production by *P. palmivora* (*in vitro*) is 24°C, and wet conditions are necessary for 18 to 72 hours (Timmer et al. 2000). Under these conditions, which may not be commonly met in New Zealand, sporulation is profuse.
- Sporangia can germinate and infect host tissues directly, or they can release large numbers of zoospores. Sporangia are spread by windblown rain through the air and are carried with water movement in soil. *Phytophthora palmivora* sporangia are caducous and can be dispersed at least 0.5 m by rain splash (Martin et al. 2012). Zoospores are released in water and are dispersed by irrigation or surface water. Zoospores are able to swim for several hours and can directly infect plant tissues (Lamour et al. 2003; Babadoost 2004) however a large amount of surface moisture is required for their movement (Roberts and Kucharek 2018).
- Phytophthora species grow and reproduce on both living and dead tissue in soil. Because they require water for spore production and infection, root and trunk rots are most frequent in sites with poor drainage and frequent waterlogging. Fruit infection can be initiated during wet conditions when spores are splashed from the soil onto fruit that are touching, or near, the ground. According to Timmer et al. (2000), raindrops would be sufficient to propel small sporangium-filled droplets into the air where they could be carried some distance by wind, but sporangia of *P. palmivora* are not dispersed by wind currents alone (unlike sporangia of some *Phytophthora* species, e.g. *P. infestans*). If wet weather persists, secondary infections are caused by spores produced on those fruit being splashed or blown onto fruit higher on the tree and, less commonly, invertebrates such as snails and ants (Taylor and Griffin 1981; Graham and Menge 2000; Mooney 2001).
- Other *Phytophthora* species have been transmitted from experimentally infected detached fruit on the soil surface to growing plants. *Phytophthora ramorum* was transmitted from infected *Pyracantha* fruit to the roots of *Viburnum* plants (Tooley et al. 2016). Tooley et al. (2016) commented that “Regulators will have to consider fruit transmission [of *P. ramorum*] as a possibility allowing for spread of the disease”.
- The spread of *P. palmivora* is unlikely to be limited by host availability. The pathogen has been reported from numerous species of crop, ornamental, shade and hedge plants, including many that are common in the New Zealand environment, e.g. *Capsicum annuum* (peppers), *Citrus* species, *Colocasia* species (taro), *Dianthus* species, *Ficus carica* (fig), *Fuchsia* species, *Grevillea* species, *Hedera helix* (English ivy), *Hibiscus* species, *Lycopersicon esculentum* (tomato), *Persea americana* (avocado), *Phaseolus* species (green and dried beans), *Philodendron* species, *Solanum tuberosum* (potato) and *Syzygium* species (Farr and Rossman 2021).

Phytophthora palmivora is a warm-climate species that is most prevalent in tropical and subtropical regions. New Zealand’s temperate climate is not optimal for this species, however:

- Major impacts were reported on kiwifruit in Elazığ Province, Turkey (38°29'01" N; 38°34'44" E; (Çiftçi et al. 2015). Most of this region has a 0.7 climate match index (CMI) with all of New

Zealand (Phillips et al. 2018), indicating that at least some parts of New Zealand are climatically suitable for establishment and disease expression.

- Climate match assessments conducted by MPI (Figure 6-1) indicate that this species is likely to be capable of infecting susceptible hosts and spreading in northern New Zealand, especially in the summer months of December to March. *Phytophthora palmivora* can also persist in the environment as dormant forms such as oospores[59] and chlamydospores, which are resistant to unfavourable conditions. Chlamydospores may form when soil moisture is limiting, conditions are cool or where the host roots are not actively growing and producing susceptible tissues for infection. Chlamydospores of some *Phytophthora* species can remain in the soil for several years (Kunta et al. 2020) and can serve as an inoculum source until the environmental conditions are optimal. However, it is uncertain how long chlamydospores of *P. palmivora* can survive and how long they would survive under conditions in New Zealand.
- Therefore, it is likely that *P. palmivora* would be able to infect plants and be able to spread in northern New Zealand, especially in the summer months of December to March. Disease expression and spread are very likely to be limited by climate, even in northern New Zealand, and the pathogen is unlikely to establish in southern regions.

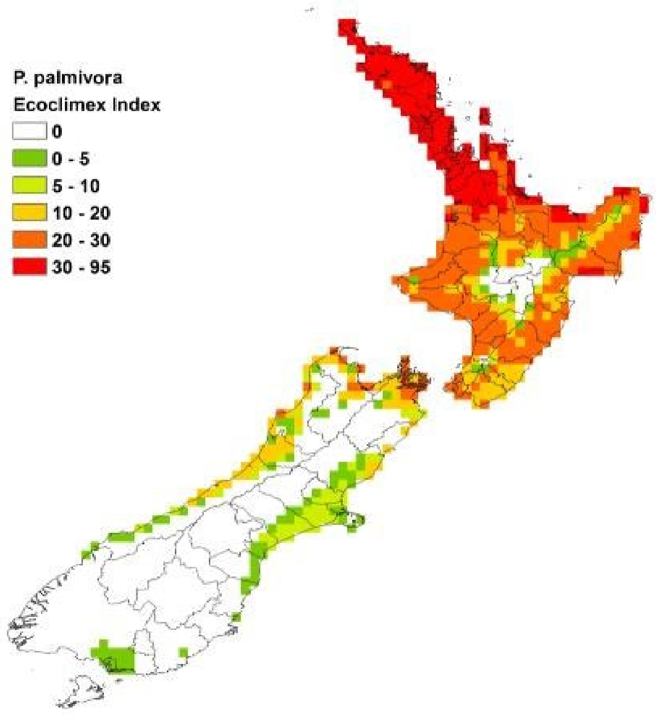

Figure 6-1: The potential distribution of *P. palmivora* in New Zealand, as predicted by CLIMEX modelling[1].

[1] The Eco-climate index shows the potential long-term establishment of the species and ranges from low (white) through increasing suitability (green through to red). The index *for P. palmivora* is around 40 (out of 100) in suitable areas in New Zealand. This index for tropical areas is above 90. This indicates that although New Zealand is less suitable compared to tropical and subtropical areas, the environment may still facilitate establishment.

Given the arguments and evidence above, that is:

- Warm (24°C) and prolonged wet conditions (18 to 72 hours) are optimal for sporangium production. Sporangia and zoospores are spread by windblown rain through the air and are carried with water movement in soil,

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[59] Oospores are formed when two different mating types are present.

- Phytophthora species require water for spore production and infection, root and trunk rots are most frequent in sites with poor drainage and frequent waterlogging,

the likelihood of *P. palmivora* establishing in New Zealand is considered high with low uncertainty.

6.2.7 Impacts in New Zealand

Phytophthora palmivora would be able to infect plants and be able to spread in northern New Zealand, especially in the summer months of December to March. Disease expression and spread are very likely to be limited by climate, even in northern New Zealand, and the pathogen is unlikely to establish in southern regions.

The spread of *P. palmivora* is unlikely to be limited by host availability because the pathogen has been reported from over 130 species of plant, some of which are common in the New Zealand environment.

Economic impacts

Phytophthora palmivora has an extremely wide host range and can cause severe losses to many tropical crops, for example, cocoa, durian, rubber, coconut, pineapple and oil palm. It has been reported attacking commercial crops overseas that are economically important in New Zealand, such as kiwifruit, citrus, apple, avocado and cut flowers (Farr and Rossman 2021). Losses resulting from *P. palmivora* are difficult to quantify because the pathogen causes a range of diseases and often occurs alongside other *Phytophthora* species. Furthermore, severity of disease is heavily influenced by climate conditions. Severe losses can occur after prolonged rains and high temperatures in late summer and early autumn (Graham and Menge 2000; Timmer et al. 2000)

- *Phytophthora palmivora* was reported to cause brown rot epidemics on citrus in Florida and California (CMI 0.5-0.8), that resulted in typical yield losses of up to 30% in some cases (Graham et al. 1998; Graham and Menge 2000). In Southeast Asia (CMI 0.3-0.6), *P. palmivora* is one of the most common species causing Phytophthora disease in citrus (Drenth and Sendall 2004).
- *Phytophthora palmivora* has been isolated from the roots and crowns of infected *Actinidia chinensis* var. *deliciosa* (cv. Hayward) plants in Turkey (CMI 0.7-0.9), causing crown and root rot symptoms on approximately 10% of plants in an experimental orchard (Çiftçi et al. 2015). Within two years, all the kiwifruit vines in the experimental orchard had died. Çiftçi et al. (2015) considered this to be a new host record and concluded that *P. palmivora* had the potential to be a major limiting factor in kiwifruit production, however, the host range and cultivar susceptibility within *Actinidia* is unknown. Furthermore, it is not known whether kiwifruit in New Zealand would be impacted since this appears to be the only report of infection of kiwifruit by *P. palmivora*.
- In Taiwan (CMI 0.4-0.8), *P. palmivora* is known to cause severe black rot in economically important orchids, including *Cattleya, Cymbidium, Dendrobium, Oncidium* and *Phalaenopsis* (Tsai et al. 2006). Many of these species are grown in New Zealand.
- Severe economic impacts of this pathogen are largely limited to tropical fruit crops in warm regions. In New Zealand, the severity of symptoms, spread and consequently the impact is likely to be limited by the climate (figure 1). Impacts in protected environments such as nurseries and glasshouses are likely to be more severe than in unprotected environments; for example, heavy losses in nurseries (up to 20% seedling mortality) are reported due to seedling blight caused by *Phytophthora* species including *P. palmivora* (Daniel and Guest 2008; Savita et al. 2012).
- An Australian risk assessment (Biosecurity Australia 2005) concluded that programmes to minimise the impact of *P. palmivora* on host plants are not likely to be more costly than existing management programs for other *Phytophthora* species; however this may depend on the crop and the country. For example, Zitko and Timmer (1994) report that *P. palmivora* is a more aggressive and competitive pathogen of citrus roots, stems and fruit tissues than *P. nicotianae*. Therefore, even if *P. nicotianae* (for example) is being managed, the establishment of *P. palmivora* may result in extra costs and damage.

The establishment of *P. palmivora* is unlikely to cause serious trade impacts.

- *Phytophthora palmivora* is widely distributed globally. However, some trading partners may impose trade restrictions in the event of an incursion, e.g.:
    - EPPO reports that several countries regard *P. palmivora* as a quarantine pest (Morocco, Bahrain and Chile), and Egypt regards it as a regulated non-quarantine pest.
    - In 2014, China closed the Californian fresh citrus fruit market (worth approximately US$140 million), based on detections of two other *Phytophthora* species (*P. syringae* and *P. hibernalis*, claimed to be absent from China) on Californian fruit. The Chinese market is open again to Californian growers, but strict export protocols have been implemented to manage the pathogens (Hao et al. 2018).

Given the arguments and evidence above, that is:

- *Phytophthora palmivora* has an extremely wide host range and can cause severe losses to many crops, but this usually limited to tropical crops and regions. Thus, impacts in protected environments such as nurseries and glasshouses are likely to be more severe than in unprotected environments in New Zealand,
- Hosts that are economically important to New Zealand include citrus, kiwifruit and cut flowers,
- Given the global distribution of *P. palmivora,* its establishment in New Zealand is unlikely to cause serious trade impacts,

the economic impact of *P. palmivora* to New Zealand is considered to be moderate with moderate uncertainty.

Environmental impacts

The establishment of *P. palmivora* may cause environmental impacts, but these are likely to be limited by climatic unsuitability:

- *Phytophthora palmivora* has been recorded on several species represented by genera in the native flora including *Hibiscus, Solanum, Pittosporum* and *Syzygium* species (Farr and Rossman 2021).
- It is most likely to establish in warmer, more humid regions of New Zealand. Although there are no native orchid species in the genera known to be affected by *P. palmivora*, Northland is the centre of diversity of native orchids in New Zealand, and this overlaps with the most likely area of establishment.
- *Phytophthora agathidicida* has been found in northern parts of New Zealand attacking kauri (*Agathis australis*), an iconic New Zealand tree species (Beever et al. 2007). Its biostatus (whether native or introduced) is not fully resolved, but it is reported to kill trees of all ages in natural forest remnants, amenity stands and silvicultural plantations (Bellgard et al. 2016). This indicates the potential for *Phytophthora* species to negatively impact on the natural environment and on socio-cultural values.

Given the evidence above, the impacts on the New Zealand environment from the establishment of *P. palmivora* in New Zealand is considered to be moderate with moderate uncertainty.

Health impacts

Phytophthora palmivora is unlikely to cause unwanted impacts on human health. No evidence of such impacts has been found.

Given the evidence above, the health impact of *P. palmivora* to New Zealand is considered to be negligible with low uncertainty.

Social/cultural impacts

Phytophthora palmivora is unlikely to cause significant social/cultural impacts. Amenity plants grown in sheltered conditions, such as tropical plant collections in botanic gardens, could be affected by this species. Host plants are common in domestic gardens but its impact and spread is likely to be limited by climate.

Given the evidence above, the social/cultural impacts of *P. palmivora* to New Zealand is considered to be low with moderate uncertainty.

6.2.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is low with moderate uncertainty,
- assuming entry, the likelihood of exposure is high with low uncertainty,
- assuming successful exposure, the likelihood of establishment is high with low uncertainty,
- the impacts on the New Zealand economy, environment, health and society are moderate with moderate uncertainty,

the overall level of assessed risk to New Zealand from *P. palmivora* on imported *P. americana* budwood is considered to be moderate, with moderate uncertainty.

6.2.9 Management considerations

Phytophthora palmivora is known to cause multiple diseases on the same host. On avocado, it causes seedling blight (Zentmyer 1976), root rot (Kurbetli et al. 2020), cankers (Tsao et al. 1994), leaf infections and dieback of young stems (Zentmyer 1976). One or more of these symptoms could be expressed on infected budwood if suitable conditions are met in PEQ.

Phytophthora palmivora is a high temperature organism. Reported optima for sporulation, infection, disease development and mycelial growth range from 24 to 30℃ (Zitko et al. 1991; Timmer et al. 2000).

Phytophthora palmivora was recovered from the roots of asymptomatic windmill palm (*Trachycarpus fortunei*) (Cacciola 2011) and is spread naturally through soil and water. *Phytophthora* species normally attack the root system and stem base of the plant, causing root rot (Parke 2016). Migliorini et al. (2015) tested potted ornamental plants in the EU using qPCR and detected *Phytophthora* in 87% of the tested pots and in 70% of the asymptomatic potted plants. Potted plants in soil carried several *Phytophthora* species without showing any external symptoms. Therefore, testing the roots and potting mix may provide the best opportunity to detect the pathogen in the case of asymptomatic infections.

The disease occurs occasionally in the summer months, in periods of heavy rainfall and high humidity (Zentmyer 1976). Therefore, testing in the summer may provide the best opportunity for detecting presence of the pathogen.

Confirming the identity of these Phytophthora requires culturing and/or molecular diagnostic tests. Current molecular detection tests for the genus *Phytophthora* are specific to several key species rather than the entire genus (Pegg et al. 2016).

It is very difficult to eradicate *Phytophthora* from an orchard once it is established in the soil. However, certain phosphonate fungistatics (phosphorous acid and phosphonate compounds) may increase the resilience of avocado trees to *Phytophthora* infection. They can be applied to the bark, to areas where cankers have been removed, injected into the trunk, or into the soil if the canker extends below ground (Eskalen and Faber 2016).

6.2.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as ‘shoot flush’) in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Phytophthora palmivora is known to cause multiple diseases on the same host. On avocado, it causes seedling blight, leaf infections and dieback of young stems (Zentmyer 1976), crown and root rot (Kurbetli et al. 2020) and cankers (Drenth and Guest 2004).

Leaf lesions are reddish-brown-black, enlarge rapidly along the larger veins and frequently cause contortion of the leaves. Dark, sunken, elongated lesions develop on young stems, and terminal buds are often killed. Conidia can develop in necrotic lesions on the leaf (Zentmyer 1976). One or more of these symptoms could be expressed on infected budwood if suitable conditions are met in PEQ.

- ***What are the known environmental conditions conducive to symptom expression?***

The optimal growing condition for *Phytophthora palmivora* is between 24-30℃. This range is optimal for sporulation, mycelial growth, infection and disease development:

- the optimum for mycelial growth was reported as 30℃ by Timmer et al. (2000), with little or no growth at 15 or 33℃ (Zitko et al. 1991),
- Timmer et al. (2000) found the optimum temperatures for infection and disease development on citrus fruit were 27 to 30℃, and that wetness of varying duration depending on the pathosystem was required for infection to take place. Brown rot caused by *P. palmivora* on citrus fruit did not develop at 22℃ or below,
- the optimum temperature for sporulation in culture and on the fruit surface was 24℃, with sporangium production decreasing rapidly at higher or lower temperatures (Timmer et al. 2000). This optimum for sporulation is considerably lower than the optimum for growth in vitro of 30℃, or for infection and disease development of 27 to 30℃ (Zitko et al. 1991; Timmer et al. 2000).

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Phytophthora palmivora was recovered from the roots of asymptomatic windmill palm (*Trachycarpus fortunei*) (Cacciola 2011) and is spread naturally through soil and water. *Phytophthora* species normally attack the root system and stem base of the plant, causing root rot (Parke 2016). Migliorini et al. (2015) tested potted ornamental plants in the European Union (EU) using qPCR and detected *Phytophthora* in 87% of the tested pots and in 70% of the asymptomatic potted plants. Potted plants in soil carried several *Phytophthora* species without showing any external symptoms. Therefore, testing the roots, potting mix or water may provide the best opportunity to detect the pathogen in the case of asymptomatic infections.

- ***What is the optimum season for conducting detection testing?***

Phytophthora palmivora is a warm climate organism. The associated diseases occur in the summer months, in periods of heavy rainfall and high humidity (Zentmyer 1976). Therefore, testing in the summer may provide the best opportunity for detecting presence of the pathogen.

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6.2.12 Appendix: Climate suitability of *Phytophthora palmivora*

Phytophthora palmivora is primarily known to be a warm-climate species reported from Malaysia, Hawaii, Brazil, Spain, Taiwan, Italy, Australia, Europe, Argentina, among other countries, causing crown rot, cankers and fruit rot in more than 200 host species where most of the tropical crops are impacted. Establishment of this species in the NZ climate needs to be assessed.

Advice

- Both models, CLIMEX and MaxEnt, predicted that North Island, especially the Northland area in New Zealand is generally suitable for the establishment of *P. palmivora* (Figure 6-1 and Figure 6-2). The weekly growth index was the highest from Dec to March (0.7) (Figure 6-3). This is due to suitable temperature (NZ summer) which allows the species to grow and establish. For the rest of the year, temperature seems to be the main limiting factor (Figure 6-3).

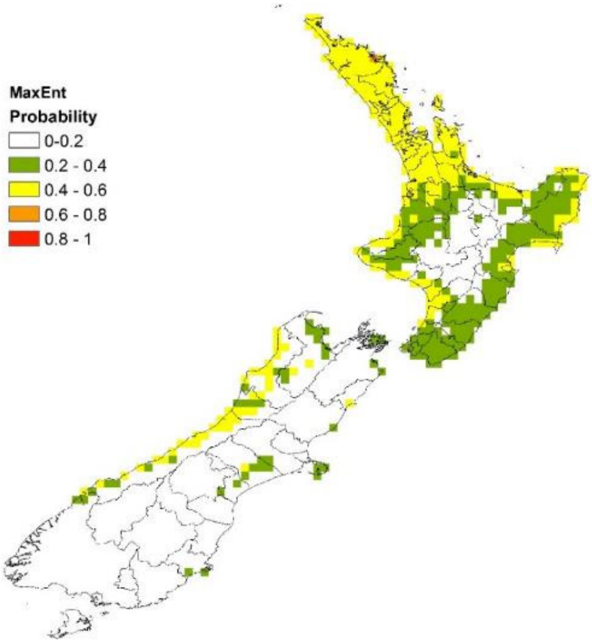

Figure 6-2: The potential distribution of P. palmivora in NZ predicted by MaxEnt model

- The Eco-climate index which shows the potential long-term establishment of the species is around 40 (out of 100) in suitable area in NZ. This index for tropical areas is above 90. This indicates although NZ is less suitable compared to tropical and subtropical area but the environment still may facilitate the pathogen establishment.

- This advice only considers the climate and current distribution of this pathogen. No other factors that may further facilitate or prohibit the species establishment have been considered.

- It should be noted that some worldwide reports are from greenhouses when the indoor environment is controlled, and the pathogen cannot grow in outdoor (such as Norway).

Table 6-3: Parameter values used in developing CLIMEX model for *P. palmivora*.

Index	Parameters	Values	Unit
DV$_0$	Lower temperature threshold	10	^{0}C
DV$_1$	lower optimum temperature	24	^{0}C
DV$_2$	upper optimum temperature	27	^{0}C
DV$_3$	Upper temperature threshold	35	^{0}C
SM$_0$	Lower soil moisture threshold	0.35	-
SM$_1$	Lower optimum soil moisture	0.7	-
SM$_2$	upper optimum soil moisture	1.5	-
SM$_3$	Upper soil moisture threshold	2.5	-
TTCS	Cold stress temperature threshold	8	^{0}C
THCS	Cold stress temperature rate	-	Week^{-1}
DTCS	Cold stress degree-day threshold	-	^{0}C
DHCS	Cold stress degree-day rate	0.008	Week^{-1}
TTHS	Heat Stress Temperature threshold	35	^{0}C
THHS	Heat Stress Temperature rate	0.0002	Week^{-1}
SDMS	Dry Stress Threshold	0.25	Week^{-1}
HDS	Dry Stress Rate	-0.01	Week^{-1}
DTCS	Cold-Dry Stress threshold (1-100)	1	-
MTCD	Cold-Dry Moisture threshold (0-5)	2.5	-
PCD	Cold-Dry Stress rate		
		0.01	Week^{-1}

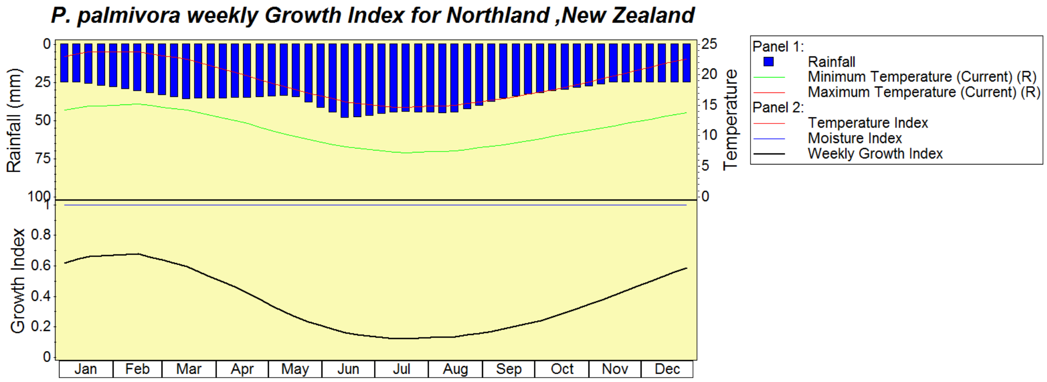

Figure 6-3: CLIMEX model output showing weekly growth index of *P. palmivora* in Northland, New Zealand

Current global distribution of the *P. palmivora* which includes 132 points were gleaned from a literature review (Figure 6-4). These presence data and long-term climate data were used to develop CLIMEX and MaxEnt models. For the CLIMEX model, the initial parameters were collected from literature (Table 6-3). An iterative adjustment of each parameter was done to reach a satisfactory agreement between the potential and the current know distribution of *P. palmivora* in these areas.

The current model predictions were made in a limited time period based on the available resource and the current distribution of the pathogen.

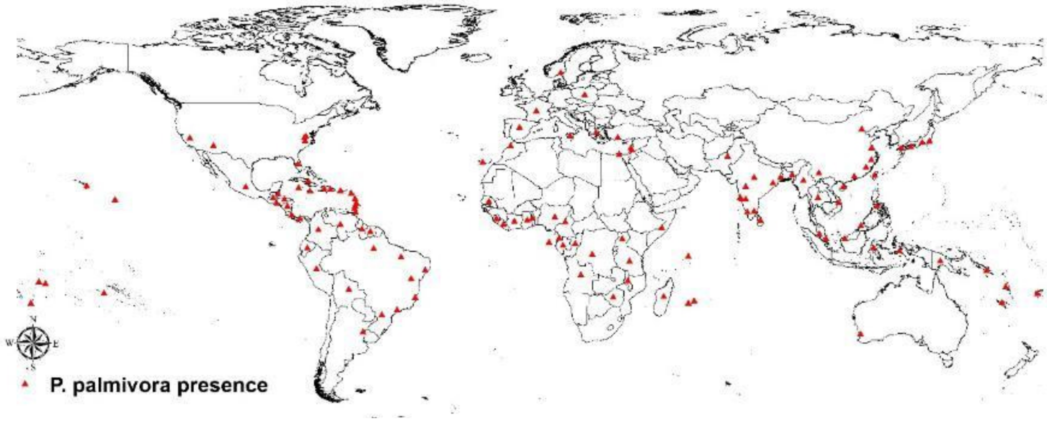

Figure 6-4: Current global distribution

7. Pest risk assessments: Viroids

7.1 Avocado sunblotch viroid (ASBVd)

Avocado sunblotch viroid is distributed in most avocado-producing areas globally and causes significant reductions in yield and fruit quality. All avocado cultivars have been reported as susceptible and infected asymptomatic trees play an important role in the epidemiology and spread of this disease.

7.1.1 Taxonomic description

Scientific name: *Avocado sunblotch viroid*

Order/Family: Avsunviroidae (Di Serio et al. 2018; ICTV 2021)

Other names include: Avocado sun blotch viroid, sun blotch of avocado, sunblotch, mancha de sol de los aguacate, viroide de la manche amarilla del aguacate, Sunblotch virus of avocado, Avocado sunblotch virus, kitmey shemesh, avokado sonvlek, ASBVd, avocado sun blotch (CABI 2021)

Taxonomic notes: *Avocado sunblotch viroid* is the single species in the genus Avsunviroid and the type member of the family Avsunviroidae. Sequence variation and high mutation rates are characteristic for viroids, and sequence variants of *Avocado sunblotch viroid* have been observed within a single tree, between trees with different symptoms, and from a single tree sampled over a period of years (Kuhn et al. 2017). Three types of sequence variants of *Avocado sunblotch viroid* have been described: B (from trees with bleached leaves), V (from trees with variegated leaves), and SC (from symptomless carrier trees) (Kuhn et al. 2017). Tissues with localized bleached symptoms usually contain higher titers of the variant B than non-symptomatic portions of the same leaf whereas a variegated leaf pattern is accompanied by the variant V (Markarian et al. 2004). The most commonly isolated variant is the asymptomatic variant SC (Markarian et al. 2004).

7.1.2 Hazard identification

Avocado sunblotch viroid is not known to be present in New Zealand. It had been listed as present in New Zealand (Pearson et al. 2006); however, following a surveillance survey carried out in 2009, it has been officially declared absent from New Zealand (Veerakone et al. 2015; PPIN 2021). *Avocado sunblotch viroid* is a regulated pest for New Zealand and has unwanted status (ONZPR 2021).

Avocado sunblotch viroid has the potential to establish (and spread) in New Zealand because:

- its natural host (*P. americana*) is available for its establishment (NZPCN 2021), and
- suitable climate is available for its establishment (Phillips et al. 2018).

Avocado sunblotch viroid has the potential to cause harm to New Zealand because:

- *Avocado sunblotch viroid* causes symptoms on *P. americana*, such as variegation on leaves, depressions and distortions on fruit, cracked bark, discoloured streaks on shoots, dwarfed, fruitless, and stunted trees and a diminished canopy (Kuhn et al. 2017; Saucedo Carabez et al. 2019), and
- even asymptomatic *P. americana* trees can have large yield reductions (Kuhn et al. 2017).

Avocado sunblotch viroid is associated with *P. americana* budwood because:

- *Persea americana* is a known host of *Avocado sunblotch viroid* (Saucedo Carabez et al. 2019), and
- *Avocado sunblotch viroid* infection is systemic (Markarian et al. 2004; Ding and Itaya 2007), it is graft-transmissible and diseased propagative material, such as budwood is a major route responsible for its spread (Saucedo Carabez et al. 2019).

Given the arguments and evidence above, *Avocado sunblotch viroid* meets the criteria to be a hazard on *P. americana* budwood (as per the commodity description) imported into New Zealand.

7.1.3 Risk assessment

Biology and epidemiology of Avocado sunblotch viroid

Host range

The only natural host of *Avocado sunblotch viroid* is *P. americana*. However, other members in the Lauraceae family have been inoculated experimentally, such as *Cinnamomum camphora* (present in New Zealand (NZPCN 2021)), *Cinnamomum zeylanicum*, *Ocotea bullata* and *Persea schiedeana* (Kuhn et al. 2017).

Symptoms

Avocado sunblotch viroid causes a variety of symptoms depending on avocado cultivar, environmental conditions, and the variants of the viroid that predominate in the host (Saucedo Carabez et al. 2019). The most recognizable symptom on fruits is sunken crevices of white, yellow, purple or reddish colour (Saucedo Carabez et al. 2019). The sunken areas on severely affected fruits sometimes become necrotic. Yellow or lighter-coloured streaks may occur on the younger branches and shoots of the tree and are usually most prominent in the new flush growth (Kuhn et al. 2017). Some leaves of infected trees develop distorted and variegated areas from the central vein that may progress and deform the entire leaf blade, however, leaves can remain asymptomatic (Kuhn et al. 2017). Bark symptoms are more common on mature trees, where rectangular checking or cracking ("crocodile bark" or "alligator skin") may be observed. Diseased trees may also be dwarfed with a diminished canopy, can be fruitless or remain stunted (Saucedo Carabez et al. 2019). The distribution of symptoms is usually irregular. Infected trees may develop only one or multiple symptoms or remain asymptomatic for their whole life (Saucedo Carabez et al. 2019). Diseased trees can become asymptomatic, and vice versa, symptoms can re-emerge following stress conditions such as hard pruning (Kuhn et al. 2017).

Transmission

Avocado sunblotch viroid is transmitted in seed and pollen, by grafting and by mechanical means (Kuhn et al. 2017). Transmission rates of 86%–100% occur in seed from asymptomatic carrier trees, however, transmission rates are much lower (0%–5.5%) in seed from symptomatic trees (Kuhn et al. 2017). Plants with seed-borne infection are often asymptomatic and infection may not become apparent until grafted with a scion of a susceptible plant. Pollen transmission of *Avocado sunblotch viroid* was demonstrated with a low transmission rate of 1.8–3.1% (Saucedo-Carabez et al. 2014). The principal route of spread of *Avocado sunblotch viroid* is grafting (Saucedo Carabez et al. 2019). The unintentional infection of grafted trees using contaminated tools and working surfaces, and the use of infected seedlings and budwood in avocado nurseries is likely to have spread *Avocado sunblotch viroid* widely throughout the avocado industries globally (Pegg et al. 2002; Kuhn et al. 2017). Natural spread through root grafts have also been reported as a mechanism of spread in the field (Ploetz 2003). Currently, *Avocado sunblotch viroid* has no known vector (Morey-León et al. 2018).

Time to symptom development and factors influencing symptom expression

The timeline of symptom development following infection by the *Avocado sunblotch viroid* is not well understood because the disease mechanism generating the symptoms of sunblotch disease in avocado is yet unknown (Kuhn et al. 2017). Furthermore, not all viroid-infected plants develop symptoms (Saucedo Carabez et al. 2019). Thus, diagnosis based on symptoms is not reliable and molecular diagnostic techniques are necessary to detect asymptomatic plants (Saucedo Carabez et al. 2019).

Factors influencing symptom expression is highly variable. Avocado sunblotch symptoms in "Collinson" indicator seedlings have been reported to develop significantly faster in hot (30/28°C, day/night) than in a cool (20/18°C, day/night) glasshouse after grafting (Da Graca and Van Vuuren

1981). Some plants develop symptoms after 2 months (Da Graca and Van Vuuren 1981), other remain symptomless (Kuhn et al. 2017). Traditionally, symptom development was observed over a period of at least 2 years using indexing (Kuhn et al. 2017).

7.1.4 Likelihood of entry

This assessment is made on the basis that *Avocado sunblotch viroid* is present in the countries where avocado budwood is being imported from. *Avocado sunblotch viroid* has been reported from all major avocado growing areas in the world, including Europe, Asia, Africa, Australia, and North, Central and South America (Saucedo Carabez et al. 2019; CABI 2021).

Avocado sunblotch viroid infection is systemic (Markarian et al. 2004; Ding and Itaya 2007), it is graft-transmissible and diseased propagative material, such as budwood is a major route responsible for its spread (Saucedo Carabez et al. 2019). Surface sterilisation would not affect the viroid. Therefore, *Avocado sunblotch viroid* can be associated with *P. americana* budwood.

Symptom expression and time to symptom development is highly variable. Symptom expression has been observed within 2 months to 2 years, however, asymptomatic variants of *Avocado sunblotch viroid* are also common (Da Graca and Van Vuuren 1981; Kuhn et al. 2017). In case of symptom expression during PEQ, the most likely observed symptoms would be yellow or lighter-coloured streaks on young branches and shoots usually exhibited in the new flush growth and occasionally white, yellow, or grey–green variegation of leaves. Some infected avocado plants remain asymptomatic for their entire life (Schnell et al. 2001; Saucedo Carabez et al. 2019). Therefore, *Avocado sunblotch viroid* can pass undetected during PEQ without molecular testing.

Given the arguments and evidence above, that is:

- *Avocado sunblotch viroid* symptom expression and time to symptom development is highly variable (Saucedo Carabez et al. 2019), and
- *Avocado sunblotch viroid* can replicate in avocado budwood without causing symptoms for the entire PEQ period (Schnell et al. 2001; Kuhn et al. 2017)

the likelihood of *Avocado sunblotch viroid* entering New Zealand associated with *P. americana* budwood is considered to be high with low uncertainty.

7.1.5 Likelihood of exposure

This assessment is made on the basis that *Avocado sunblotch viroid* has entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *Avocado sunblotch viroid* (Saucedo Carabez et al. 2019; NZPCN 2021).

If *P. americana* plants are infected with *Avocado sunblotch viroid* at the time of their release from PEQ they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, *Avocado sunblotch viroid* meets the criteria to be a hazard for New Zealand.

7.1.6 Likelihood of establishment

This assessment is made on the basis that *Avocado sunblotch viroid* has been successfully exposed to a suitable host plant in the New Zealand environment.

The natural host range of *Avocado sunblotch viroid* is reported to be restricted to *P. americana*. This host in the form of cultivated avocados is available for its establishment in the North Island (see section 2.3). If the host range includes other members of the family Lauraceae then the number of potential host species available for the establishment of these pathovars is greatly increased – there are five native and four introduced species (in addition to *P. americana*) from the Lauraceae family found in New Zealand (NZPCN 2021). Out of these present in New Zealand, only *Cinnamomum camphora* is confirmed as an experimental host of *Avocado sunblotch viroid* (Kuhn et al. 2017).

Avocado sunblotch viroid is reported from all major avocado growing areas in the world (Kuhn et al. 2017; Saucedo Carabez et al. 2019). Some avocado growing regions, such as California, the coastal areas in Israel, South Africa, East Australia, have a similar climate to the whole of New Zealand, as well as to the Auckland and Northland regions, as indicated by a climate match index of ≥0.7 (Phillips et al. 2018). Therefore, climate suitable for the establishment of *Avocado sunblotch viroid* is readily available. In addition, climate suitability is supported by the fact that *Avocado sunblotch viroid* was reported from New Zealand (Pearson et al. 2006) prior to its eradication (Veerakone et al. 2015).

Given the arguments and evidence above, that is:

- hosts (in the form of cultivated avocados) are available for the establishment of *Avocado sunblotch viroid* (NZPCN 2021), and
- suitable climate is available for the establishment of *Avocado sunblotch viroid* (Phillips et al. 2018), and
- the wetter conditions of New Zealand compared to the major avocado growing areas are likely to favour establishment of *Avocado sunblotch viroid*, and
- *Avocado sunblotch viroid* had been reported from New Zealand (Pearson et al. 2006), prior to its eradication (Veerakone et al. 2015)

the likelihood of *Avocado sunblotch viroid* establishing in New Zealand is considered high, with low uncertainty.

7.1.7 Impacts in New Zealand

The spread of *Avocado sunblotch viroid* infection is slow and more likely due to the reintroduction of infected asymptomatic materials than to natural processes. An average annual growth rate of 2.3% to 4.7% of disease incidence has been reported (Pegg et al. 2002). Thus, the greatest impact on New Zealand's avocado industry is assumed to occur years after first introduction.

Economic impacts

The economic impacts of *Avocado sunblotch viroid* are likely limited to the avocado industry because *P. americana* is the only known natural host. The most relevant economic impact to New Zealand of sunblotch disease would likely be the effect on avocado yield (Kuhn et al. 2017). Likewise, the fruit quality and the tree growth are affected (Kuhn et al. 2017). All avocado cultivars have been reported as susceptible to this disease. For example, yield losses of 14% in symptomatic "Fuerte" trees and 80% in asymptomatic "Edranol" have been reported (Kuhn et al. 2017). High yield losses of 95% were observed in both asymptomatic "Caliente" and "Reed" trees (Kuhn et al. 2017). Yield loss in asymptomatic "Hass" trees is in the range of 15–30%, while symptomatic "Hass" trees have yield losses of 50–80% (Saucedo-Carabez et al. 2014; Kuhn et al. 2017). In addition, about 50% of the fruit produced by a symptomatic "Hass" trees may be scarred and therefore downgraded in quality (Saucedo-Carabez et al., 2014).

The costs of managing *Avocado sunblotch viroid* is high because infected trees need to be removed as well as all trees within 15 m and there is no cure for the disease (Kuhn et al. 2017). If there was an outbreak of *Avocado sunblotch viroid* in one of the major avocado-producing regions in New Zealand, all trees would need to be tested including asymptomatic avocado trees. Disinfestation of pruning tools and harvesting and grafting material is crucial to prevent the undetected spread of *Avocado sunblotch viroid*. Eradication could be possible by testing and wide-scale removal of infected avocado trees in the area, causing significant economic losses for a large portion of the industry. If new trees were planted it would take approximately 3 years for them to produce their first commercial crop and up to 10 years for them to become as productive as older trees which were removed (Tupu.nz 2021).

The indirect impact of an outbreak of *Avocado sunblotch viroid* on trade is considered negligible for New Zealand because the viroid is reported from all major trading partners and avocado growing areas around the world, including Australia which is New Zealand's biggest export market for avocado (Saucedo Carabez et al. 2019; CABI 2021). Furthermore, trade restrictions seem unlikely as asymptomatic fruits can still be exported since they are for human consumption rather than for propagation (Kuhn et al. 2017).

Given the arguments and evidence above, that is:

- avocados are the only natural hosts of *Avocado sunblotch viroid* commercially produced in New Zealand, and
- infection of avocado trees with *Avocado sunblotch viroid* could cause large yield losses and significant downgrading in quality of the fruit, and
- control of an outbreak could involve wide scale destruction of avocado trees (e.g., removal of all infected trees), and
- there are likely to be negligible trade restrictions, and
- a slow spread of disease incidence

the economic impact of *Avocado sunblotch viroid* to New Zealand is considered to be moderate, with low uncertainty.

Environmental impacts

The natural host range of *Avocado sunblotch viroid* is restricted to *P. americana* (Saucedo Carabez et al. 2019). There are no reports of natural infection of other plant species (google scholar search terms ["wild plants" AND "Avocado sunblotch viroid"] and ["reservoir plants" AND "Avocado sunblotch viroid"] and ["Avocado sunblotch viroid" AND "natural environments"]).

Given the arguments and evidence above, the impact on the environment from the establishment of *Avocado sunblotch viroid* in New Zealand is considered to be negligible, with low uncertainty.

Human health impacts

There are no known human health impacts associated with *Avocado sunblotch viroid* (google scholar search terms ["human health" AND "Avocado sunblotch viroid"] and ["toxin" AND "Avocado sunblotch viroid"]).

Given the arguments and evidence above, the human health impact of *Avocado sunblotch viroid* to New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

A response to an outbreak of *Avocado sunblotch viroid* may involve the removal of infected or exposed avocado trees growing on residential properties. This is likely to cause distress to affected residents, however, this impact is likely to be localised and short-lived. Moreover, a response would rather target commercial growers and nurseries as the main grafters.

Given the arguments and evidence above, the sociocultural impact of *Avocado sunblotch viroid* to New Zealand is considered to be negligible, with high uncertainty.

Overall impact to New Zealand

The overall impact of *Avocado sunblotch viroid* on the New Zealand economy, environment, human health and society is considered to be moderate, with low uncertainty.

7.1.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is high
- assuming entry, the likelihood of exposure is high
- assuming successful exposure, the likelihood of establishment is high
- the overall impact on the New Zealand economy, environment, human health and society is considered to be moderate

the overall level of assessed risk to New Zealand from *Avocado sunblotch viroid* on imported *P. americana* budwood (as in the commodity description) is considered to be moderate, with low uncertainty.

7.1.9 Management considerations

In case of symptom expression during PEQ, the most likely observed symptoms would be yellow or lighter-coloured streaks on young branches and shoots usually exhibited in the new flush growth and occasionally white, yellow, or grey–green variegation of leaves (Kuhn et al. 2017; Saucedo Carabez et al. 2019). However, some infected avocado plants remain asymptomatic for their entire life and asymptomatic variants are commonly isolated (Schnell et al. 2001; Saucedo Carabez et al. 2019). Therefore, *Avocado sunblotch viroid* can pass undetected during PEQ without molecular testing. Usually the pathogen is detected using RT-qPCR, no matter where on the tree the leaf samples are taken and in what month of the year the tree is sampled (Geering 2011).

7.1.10 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Symptom expression of *Avocado sunblotch viroid* is highly variable because changes in single nucleotides in the viroid genome can result in altered symptoms in avocado trees (Schnell et al. 2001). In case of symptom expression during PEQ, the most likely observed symptoms would be yellow or lighter-coloured streaks on young branches and shoots usually exhibited in the new flush growth and occasionally white, yellow, or grey–green variegation of leaves (Kuhn et al. 2017; Saucedo Carabez et al. 2019). However, some infected avocado plants remain asymptomatic for their entire life and asymptomatic variants are commonly isolated (Schnell et al. 2001; Saucedo Carabez et al. 2019). Therefore, *Avocado sunblotch viroid* can pass undetected during PEQ without molecular testing.

- ***What are the known environmental conditions conducive to symptom expression?***

The factors influencing symptom expression after infection with *Avocado sunblotch viroid* are not well understood. Using indexing, symptom expression was reported to increase during higher temperatures, stress and heavy pruning. Traditionally, symptom development was observed over a period of at least 2 years using indexing and a range of indicator seedlings (Kuhn et al. 2017). However, the possibility of remaining asymptomatic even during periods of stress and high temperatures cannot be ruled out (Schnell et al. 2001; Saucedo Carabez et al. 2019).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Flower buds are preferable over leaves for diagnosis, as the viroid titer is suspected to be higher and carbohydrate contamination of the RNA extract less of a problem (Kuhn et al. 2017). The preferred method sensitive enough for detecting *Avocado sunblotch viroid* in leaves and young leaf flushes is real-time reverse transcription PCR (RT-qPCR) (Morey-León et al. 2018).

- ***What is the optimum season for conducting detection testing?***

The concentration of *Avocado sunblotch viroid* can fluctuate during the season. However, usually the pathogen is detected using RT-qPCR, no matter where on the tree the leaf samples are taken and in what month of the year the tree is sampled (Geering 2011).

7.1.11 References

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7.2 Potato spindle tuber viroid

Potato spindle tuber viroid (PSTVd) is a plant pathogen which consists of a small, unencapsidated (no coat protein), circular, single-stranded RNA molecule 356-361 nucleotides in length. It causes disease primarily in solanaceous plants, including potato, tomato and capsicum. However, the reported natural host range is large and includes avocado.

7.2.1 Taxonomic description

Scientific name: *Potato spindle tuber viroid*

Order/Family: Lefavirales / Pospiviroidae (ICTV 2021)

Other names include: PSTVd, Potato gothic virus, Potato spindle tuber pospiviroid, Potato spindle tuber virus (PSTV, prior to 1971), Tomato bunchy top viroid (EPPO 2020).

Taxonomic notes: *Potato spindle tuber viroid* was initially isolated from diseased potato plants and was the first viroid to be identified (Diener, 1971 in Diener 2003). It is the type species of both the genus *Pospiviroid* and family Pospiviroidae (Di Serio et al. 2021). Strains used to be classified as mild, intermediate or severe based on symptoms observed in sensitive tomato cultivars but these designations do not necessarily reflect the severity of symptoms seen in natural hosts under field conditions (Owens 2007). *Potato spindle tuber viroid* is known for a constant change in sequence variation throughout infection (so-called quasispecies, Adkar-Purushothama et al. 2020). Because of its high mutation rate, PSTVd is highly adaptable (i.e. new hosts, environment) and highly variable (in terms of symptoms and transmission). An arbitrary level of less than 90% sequence identity and distinct biological properties are the main criteria for separating viroid species within a genus (Adkar-Purushothama et al. 2020).

7.2.2 Hazard identification

Potato spindle tuber viroid is not known to be present in New Zealand:

- *Potato spindle tuber viroid* has occurred transiently in New Zealand, mainly in commercial glasshouse tomatoes and capsicums (Lebas et al. 2005). The viroid has been eradicated and is considered to not be established in NZ (Veerakone et al. 2015; PPIN 2021), and
- it is a regulated pest and an unwanted organism (ONZPR 2021).

Potato spindle tuber viroid has the potential to establish (and spread) in New Zealand because:

- Hosts in the form of commercial crops (e.g. potatoes, avocadoes and tomatoes), common weeds (e.g. black nightshade, cape gooseberry, and wavy-leaved fleabane) and common ornamentals (e.g. dahlias, petunias and angel’s trumpets) are available for its establishment (EPPO 2021; NZOR 2021; NZPCN 2021), and
- *Potato spindle tuber viroid* has been recorded from all continents, from the tropics as far north as Russia (EPPO 2021). Thus, suitable climate is available for the establishment of the viroid (Phillips et al. 2018) and temperatures are high enough to support its transmission and spread (Verhoeven et al. 2010a; NIWA 2021).

Potato spindle tuber viroid has the potential to cause harm to New Zealand because:

- *Potato spindle tuber viroid* can cause direct impacts on the potato, tomato and capsicum industry through large yield losses, and
- *Potato spindle tuber viroid* can cause indirect impacts through export or trade restrictions imposed by other countries, and
- control of an outbreak would involve wide scale testing of growing areas and destruction of infected plants and trees.

Potato spindle tuber viroid can be associated with *Persea americana* budwood because:

- *Persea americana* is a known host of PSTVd (Querci et al. 1995; Barrera and Rojas 2007; Nieto-Taype et al. 2014);
- *Potato spindle tuber viroid* infection is systemic and graft-transmissible (Zhu et al. 2001; Di Serio et al. 2020).

Given the arguments and evidence above, *Potato spindle tuber viroid* meets the criteria to be a hazard on *Persea americana* budwood (as in the commodity description) imported to New Zealand.

7.2.3 Risk assessment

Biology and epidemiology of Potato spindle tuber viroid

Host range

Potatoes and other tuber-bearing *Solanum* species are considered to be the main hosts of *Potato spindle tuber viroid* (EPPO 2021). In addition, PSTVd has been found naturally infecting a range of solanaceous crop species such as tomato (*S. lycopersicum*) (Puchta et al. 1990; Lebas et al. 2005) and capsicum (*Capsicum annuum*) (Lebas et al. 2005), eggplant (*S. melongena*), pepino (*S. muricatum*), as well as solanaceous ornamentals like potato vine (*S. jasminoides*) (Di Serio 2007; Verhoeven et al. 2010b) and angel's trumpets (*Brugmansia* spp.) (Verhoeven et al. 2010b). Various solanaceous and non-solanaceous plants have also been identified as reservoirs of PSTVd (Mackie et al. 2016). EPPO (2021) lists 34 known hosts (from six families) and a further 32 experimental hosts (from 9 families).

Potato spindle tuber viroid was first reported from avocado in 1995 after it was detected in 72% of symptomatic and 21% of asymptomatic avocado trees surveyed in Peru (Querci et al. 1995). Although eradication of PSTVd in avocado breeding stock in Peru has been attempted (Barrera and Rojas 2007) it is still considered an avocado pest in that country (Nieto-Taype et al. 2014). *Potato spindle tuber viroid* has been reported from the following avocado varieties:

- Hass (Barrera and Rojas 2007)
- Naval (Barrera and Rojas 2007)
- Bacon (Barrera and Rojas 2007)
- Fuerte (Querci et al. 1995)
- Super Fuerte (Querci et al. 1995)
- Topa Topa (Querci et al. 1995)
- Zutano (Querci et al. 1995)
- Duke (Querci et al. 1995)
- Campong (Querci et al. 1995)
- P-11 (Querci et al. 1995)
- Mexicano (Querci et al. 1995).
- Collinred (Querci et al. 1995; Barrera and Rojas 2007)
- Villacampa (Querci et al. 1995; Barrera and Rojas 2007).

Mild, intermediate and severe strains of PSTVd have all been reported from avocados (Querci et al. 1995).

Symptoms

Potato spindle tuber viroid infection in avocados is associated with a range of symptoms including:

- A vertical branching pattern with weak, slender branches (Querci et al. 1995; Barrera and Rojas 2007),
- slender horizontal branching (Querci et al. 1995),
- leaf size reduction, chlorosis and defoliation (Querci et al. 1995),
- lack of fruit setting (Barrera and Rojas 2007) and a decrease in fruit size and number (Querci et al. 1995),
- bunchiness or proliferation of inflorescences (Querci et al. 1995; Barrera and Rojas 2007),
- stunting (Barrera and Rojas 2007),
- decline in the organoleptic properties of the fruit (Nieto-Taype et al. 2014), and

- death of branches, desiccation and tree death (Querci et al. 1995).

It is difficult to definitively ascribe all of these symptoms to infection with PSTVd because some symptomatic trees were also infected with *Avocado sunblotch viroid* (ASBVd) (Querci et al. 1995; Barrera and Rojas 2007). One report ascribes leaf yellowing and fruit grooving to ASBVd infection, vertical branching and prolific inflorescences to PSTVd, and stunting and lack of fruit set to infection by both viroids (Barrera and Rojas 2007). However, the data and analysis underpinning this conclusion are not presented in Barrera and Rojas (2007). In addition, symptoms were not associated with infection by either viroid in 25% of trees surveyed in Peru (Querci et al. 1995). As it is the case for ASBVd, PSTVd likely causes a variety of symptoms depending on avocado cultivar, environmental conditions, and the variants of the viroid that predominate in the host but can also remain asymptomatic (Hadidi et al. 2017; Saucedo Carabez et al. 2019; Adkar-Purushothama et al. 2020).

Life cycle and transmission

Viroids replicate autonomously and usually infect their hosts systemically (Di Serio et al. 2020). *Potato spindle tuber viroid* is imported into cell nuclei prior to replication, exported out of the nucleus after replication, and then travels from cell to cell via plasmodesmata (Ding et al. 1997). In tomatoes (Palukaitis 1987; Zhu et al. 2001) and the experimental host *Nicotiana benthamiana* (Zhu et al. 2001) PSTVd has been shown to move systemically in phloem. It is generally assumed that this is the case for all viroids (Di Serio et al. 2020) and it is likely that PSTVd moves in the phloem of all host plants, including avocado.

Potato spindle tuber viroid is efficiently transmitted by vegetative propagation and this mode of transmission is a major cause of spread of PSTVd in potatoes and vegetatively-propagated ornamentals (EPPO 2021). Mechanical transmission has also been demonstrated experimentally in a range of species including from potatoes to potatoes, and from solanaceous ornamentals (angel's trumpets and potato vine) to potatoes and tomatoes. *Potato spindle tuber viroid* has been successfully transmitted by applying infected sap to abraded, bruised or cut plant parts, cutting plants with contaminated tools, or rubbing leaves with contaminated fingertips (Bonde and Merriam 1951; Verhoeven et al. 2010a).

High rates of field transmission have been shown in potatoes due to tractor tyres being contaminated by driving over infected plants (Merriam and Bonde 1954 in Verhoeven 2020) and by using contaminated cultivating and hilling equipment (Manzer and Merriam 1961 in Verhoeven 2020). The transmission rate is dependent on which species and varieties are used as the inoculum source and which are inoculated (Bonde and Merriam 1951; Verhoeven et al. 2010a). The temperature at which inoculation takes place also affects transmission rates, with the highest rates occurring at 20–25℃(Verhoeven et al. 2010a). At 15℃, transmission failed in potato and was rare in tomatoes (Verhoeven et al. 2010a).

The rod-like secondary structure of PSTVd makes it resistant to nuclease degradation (Owens 2007). The viroid’s stability means that it can remain infective in extracted tomato sap on common surfaces such as cotton, wood, rubber tire, leather, metal, plastic, and string for up to 24 hours, although it remains infective on human skin for only 30 minutes (Mackie et al. 2015). In addition, PSTVd remains infectious in water at 20 ± 4℃ for up to seven weeks (Mehle et al. 2013).

Many pospiviroids, including PSTVd, are predominately located in the nuclei of infected cells and are seed-transmitted (CABI 2021). *Potato spindle tuber viroid* was detected in true potato seeds that had been stored for 21 years (Singh et al. 1991). Contaminated seed is an important source of spread of PSTVd but efficiency of transmission of PSTVd can be highly variable. The rate of seed-transmission of pospiviroids is dependent on the viroid strain, host plant species, cultivar and distribution of viroids in seed parts (Matsushita et al. 2018). The rate of PSTVd seed-transmission was found to be 0–90.2 % in tomato, 0.3 % in *Capsicum annuum* var. *grossum*, 0.5 % in *C. annuum* var. *angulosum*, 1.2 % in *Glebionis coronaria*, and 81 % in *Petunia* × *hybrida* (Matsushita et al. 2018). No seed-transmission was observed for *C. annuum* cv. Yolo Wonder (Verhoeven et al. 2020).

Pollen is another potential source of infection. Transmission of PSTVd from infected plants to pollen has been demonstrated experimentally in potatoes. Pollination of potato plants with infected pollen results in infection of apical leaves, inflorescences, sepals on fruit, fruit skin, fruit pulp, tubers, and true seed (Singh et al. 1992).

Transmission of PSTVd by the aphid *Myzus persicae* has been demonstrated in potatoes and cape gooseberry (*Physalis floridana*) (Syller et al. 1997). However, transmission likely only occurs if plants are co-infected with *Potato leafroll virus* (Querci et al. 1997; Singh and Kurz 1997; Syller et al. 1997). *Potato leafroll virus* and *M. persicae* are both reported from New Zealand (Veerakone et al. 2015; NZOR 2021). Grasshoppers, beetles (Hančinský et al. 2020) and oomycetes (Afanasenko et al. 2021) are also suspected to transmit PSTVd to host plants.

It is likely that all of the described transmission modes of PSTVd apply to avocado.

Factors influencing symptom expression and rate of asymptomatic infection

Potato spindle tuber viroid symptom expression is highly variable and influenced by the viroid strain, environmental conditions and host species and cultivar (Mackie et al. 2019).

In potato warm and dry climatic conditions usually increase symptoms of PSTVd (Schmitz and Steger 2007) and are more obvious at 25 ˚C than at 15 ˚C (Goss and Peltier 1925). Similarly, in tomato plants more obvious symptoms may develop at high temperatures (24-39˚C) at higher light intensity, but tomatoes infected with the mild strain of PSTVd may never develop symptoms despite high viroid concentrations (Harris and Browning 1980; Grasmick and Slack 1985). In peppers, symptoms in response to PSTVd infection are usually mild and depend on variety (Lebas et al. 2005). Infections of solanaceous ornamentals and weeds are usually mild or symptomless (Matsushita and Tsuda 2015; Mackie et al. 2016).

Whether the relationship between temperature, light intensity and strain type holds true for symptom expression in avocados is not known. However, asymptomatic infection of avocados by PSTVd is common (Hadidi et al. 2017). A survey in Peru showed that approximately 21% of asymptomatic trees were infected with PSTVd and at some sites 93-100% of all asymptomatic trees sampled were PSTVd positive (Querci et al. 1995). It was hypothesised that PSTVd infections in avocado are often latent unless the tree is co-infected with ASBVd (Querci et al. 1995). However, vertical branching and prolific inflorescences symptoms were also ascribed to single infection with PSTVd (Querci et al. 1995; Barrera and Rojas 2007).

7.2.4 Likelihood of entry

This assessment is made on the basis that *Potato spindle tuber viroid* is present in the countries where avocado budwood is being imported from. *Potato spindle tuber viroid* infects its hosts systemically (Zhu et al. 2001; Di Serio et al. 2020). Therefore, it may be associated with avocado budwood. Rates of up to 100% asymptomatic infection of avocado trees have been reported (Querci et al. 1995). Because infection can be asymptomatic it is possible that budwood may be harvested from infected trees, and infection would not be evident in quarantine inspections (in exporting country and in NZ).

Given the arguments and evidence above, that is:

- *Potato spindle tuber viroid* infects its hosts systemically (Zhu et al. 2001; Di Serio et al. 2020) and can therefore be associated with avocado budwood, and
- it is likely to display no symptoms during the PEQ period (Querci et al. 1995)

the likelihood of PSTVd entering New Zealand associated with *Persea americana* budwood is considered to be high, with low uncertainty.

7.2.5 Likelihood of exposure

This assessment is made on the basis that *Potato spindle tuber viroid* has entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand.

If avocado plants are infected with PSTVd at the time of their release from Post Entry Quarantine they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of PSTVd in New Zealand from *Persea americana* budwood is considered high, with low uncertainty.

7.2.6 Likelihood of establishment

This assessment is made on the basis that *Potato spindle tuber viroid* has been successfully exposed to a suitable host plant in the New Zealand environment. *Potato spindle tuber viroid* has a very broad host range (EPPO 2021) and many of its hosts are either grown commercially in New Zealand (i.e. potatoes, avocadoes, eggplant and tomatoes), are common ornamentals (e.g. *Dahlia* spp., *Petunia* spp. and *Brugmansia* spp.), or are common weeds (e.g. black nightshade, *Solanum nigrum*; cape gooseberry, *Physalis peruviana*; and wavy-leaved fleabane, *Erigeron bonariensis*) (NZOR 2021; NZPCN 2021). Therefore, hosts for the establishment of PSTVd are readily available.

Climate is likely to influence the distribution of PSTVd mainly through influencing the distribution of its hosts. Under laboratory conditions, temperature can influence the transmission rate of PSTVd with a higher rate at 20-25°C than 15°C (Verhoeven et al. 2010a). In the main avocado growing regions of New Zealand daily maximum temperatures at or above 20°C are common in the summer months, and above 15°C for the majority of the year (NIWA 2021). Therefore, temperatures to support transmission and establishment of PSTVd are readily available.

Potato spindle tuber viroid has occurred transiently in New Zealand in the past. It had been detected on a few occasions in New Zealand in tomato, tobacco and capsicum glasshouse plants and in Cape gooseberry, but was eradicated each time (Lebas et al. 2005; Ward et al. 2010). Successful eradication of the viroid was likely aided by the fact that it was detected early and in an environment that was relatively easy to control (i.e., inside a glasshouse and seeds). Due to the many reported reservoir plants, eradication seems unlikely in case undetected spread occurs in an outdoor environment.

Given the arguments and evidence above, that is:

- Hosts and reservoir plants for the establishment of PSTVd are readily available (NZOR 2021; NZPCN 2021), and
- climate suitable for the establishment of PSTVd is readily available (Phillips et al. 2018; NIWA 2021)

the likelihood of PSTVd establishing in New Zealand is considered high, with low uncertainty.

7.2.7 Impacts in New Zealand

There is no effective chemical control known for PSTVd. The most effective means of control is using certified viroid-free planting material, planting resistant cultivars and maintaining sanitary cultural practices. Control of an outbreak is generally achieved by removal and destruction of infected plants, sanitation of the production location, temporarily banning cultivation of host plants, and thorough cleaning and disinfection of tools, machinery and other equipment (Verhoeven 2020).

If PSTVd were introduced to an avocado producing region of New Zealand, it may remain undetected for a prolonged period given the high rates of asymptomatic infection reported (Querci et al. 1995). This allows time for the viroid to spread to other crops and reservoir plants. Successful mechanical transmission from solanaceous ornamentals (*Brugmansia* spp. and *S. jasminoides*) to tomatoes has been demonstrated (Verhoeven et al. 2010a). In addition, phylogenetic analyses have indicated that volunteer crop plants, weeds and native plants are likely to be the source of recurring outbreaks of PSTVd in field-grown tomatoes, capsicums and chillies in a remote agricultural area in Australia (Mackie et al. 2016). Intercropping of potatoes with avocadoes in Peru is suspected to be the source of PSTVd in avocados (Querci et al. 1995). Therefore, spread of PSTVd from avocados to other species in the field is plausible.

Economic impacts

Hosts of PSTVd, such as avocados, potatoes, tomatoes and capsicums are high value crops for New Zealand. They had a combined domestic value of $163.5 million and an export value of $259 million in 2020 (Plant & Food Research 2020).

Direct impacts would likely occur on the tomato, potato and capsicum industry (Lebas et al. 2005; Mackie et al. 2019). Yield losses vary with host cultivar, PSTVd variant and season, and are particularly strong under warm conditions (Harris and Browning 1980). The general pattern is of a progressive decrease in yield as infection increases. In eastern Canada, field trials of potatoes (with 100% infection rates) have shown that even the mild strain can cause up to 24% yield reductions whereas the severe strain has been reported to cause reductions of up to 64-90%% (Singh et al. 1971; Mackie et al. 2016; Mackie et al. 2019). With infection rates of 3.8% in the field, and an observed ratio of 11:1 of the mild to severe strains, yield loss in the field was calculated to be approximately 1% (Singh et al. 1971). Higher infection rates (6.5%) among field-sampled commercial seed potatoes have been reported in China (Qiu et al. 2016), although yield losses were not reported during this survey. A previous survey in northern China reported yield losses of 20-30% (Cui et al. 1992), but information about infection rates and the strains involved is unavailable. Depending on cultivar and strain, in a small-scale pot experiment, PSTVd was reported to decrease the yield of tomato fruit by up to 47% (Mackie et al. 2019). FreshFacts (2019) estimated New Zealand's tomato exports at around 15 million NZD, capsicum exports at around 20 million NZD and potato exports at around 130 million NZD. Generally, the magnitude of the PSTVd-induced yield losses overseas has been found considerable, regardless of cultivar or crop: 50 to 90% for potato tubers and 52 to 89% for tomato fruit (Mackie et al. 2019).

It is likely establishment of PSTVd would have additional implications for New Zealand's horticultural industry through export or trade restrictions imposed by other countries. Given the presence of PSTVd in the EU, Australia and China (EPPO 2021), the biggest potential loss of market access would likely be to the USA and South Asia.

The costs from eradication attempts or a response to an outbreak of PSTVd are considered high because they would include widespread testing and the destruction of infected plants (including avocado trees), sanitation of the production location, temporarily banning cultivation of host plants, and thorough cleaning and disinfection of tools, machinery and other equipment (Verhoeven 2020). Moreover, host crops of PSTVd should not be grown in the infested fields for 3 years (Verhoeven 2020).

Given the arguments and evidence above, that is:

- direct impacts would likely occur on the tomato, potato and capsicum industry in New Zealand through yield losses, and
- indirect impacts would occur from export or trade restrictions imposed by other countries, and
- control of an outbreak would involve wide scale testing of growing areas and destruction of infected plants and trees

the economic impact of *Potato spindle tuber viroid* to New Zealand is considered to be moderate, with low uncertainty.

Environmental impacts

The natural host range of PSTVd is large and increasing (Matsushita and Tsuda 2015). In Australia PSTVd was detected infecting native plants and weeds after floodings (Mackie et al. 2016). Native

New Zealand plant species from the nightshade family, such as Poroporo (*Solanum aviculare* var. *aviculare*, *Solanum aviculare* var. *latifolium*, *Solanum laciniatum*) (NZPCN 2021) could be prone to PSTVd infection. Although no evidence of Poroporo as a host of PSTVd could be found, this relationship seems plausible as PSTVd naturally infect solanaceous plants (Hančinský et al. 2020). However, PSTVd infections in wild reservoir plants are usually asymptomatic and few are mildly symptomatic (Mackie et al. 2016), so PSTVd would likely have minimal impact on New Zealand native species. Overseas impacts from PSTVd infection are, so far, only known for potato, tomato and capsicum (Matsushita and Tsuda 2015; Verhoeven 2020) and no evidence could be found that PSTVd severely impacts natural ecosystems.

Given the arguments and evidence above, the impact on the environment from the establishment of *Potato spindle tuber viroid* in New Zealand is considered to be negligible, with moderate uncertainty.

Human health impacts

There are no known human health impacts associated with *Potato spindle tuber viroid* (google scholar search terms ["human health" AND "Potato spindle tuber viroid"] and ["toxin" AND "Potato spindle tuber viroid"]).

Given the arguments and evidence above, the human health impact of *Potato spindle tuber viroid* to New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

A response to an outbreak of *Potato spindle tuber viroid* may involve the removal of infected or exposed avocado trees, tomato and capsicum plants, ornamental and reservoir hosts growing on residential properties. This is likely to cause distress to affected residents, however, this impact is likely to be localised.

Given the arguments and evidence above, the sociocultural impact of *Potato spindle tuber viroid* to New Zealand is considered to be very low, with low uncertainty.

Overall impact to New Zealand

The overall impact of *Potato spindle tuber viroid* on the New Zealand economy, environment, human health and society is considered to be moderate, with low uncertainty.

7.2.8 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is high
- assuming entry, the likelihood of exposure is high
- assuming successful exposure, the likelihood of establishment is high
- the overall impact on the New Zealand economy, environment, human health and society is considered to be moderate

the overall level of assessed risk to New Zealand from *Potato spindle tuber viroid* on imported *P. americana* budwood (as in the commodity description) is considered to be moderate, with low uncertainty.

7.2.9 Management considerations

Potato spindle tuber viroid was detected in asymptomatic avocado trees surveyed in Peru (Querci et al. 1995). Therefore, PSTVd can be asymptomatic in avocado budwood and pass undetected during PEQ without molecular testing. The preferred methods sensitive enough for detecting PSTVd are real-time reverse transcription PCR (RT-qPCR) or real-time RT-PCR (Botermans et al. 2020). Detailed management options to reduce the risk of entry, spread and consequences of PSTVd were identified and evaluated by EFSA (2011).

Potato spindle tuber viroid was detected in 72% of symptomatic and 21% of asymptomatic avocado trees surveyed in Peru (Querci et al. 1995). Therefore, PSTVd can be asymptomatic and can pass undetected during PEQ without molecular testing. In case of symptom expression, PSTVd likely causes a variety of symptoms depending on avocado cultivar, environmental conditions, and the variants of the viroid that predominate in the host (Saucedo Carabez et al. 2019; Adkar-Purushothama et al. 2020). Vertical branching and prolific inflorescences in avocado were attributed to PSTVd infection, and stunting and lack of fruit set to co-infection with *Avocado sunblotch viroid* (ASBVd) (Barrera and Rojas 2007).

As it is the case for many viroids, PSTVd likely causes a variety of symptoms depending on avocado cultivar, environmental conditions, and the variants of the viroid that predominate in the host (Saucedo Carabez et al. 2019; Adkar-Purushothama et al. 2020). In hosts such as potato and tomato, high temperatures (>24˚C) and high light intensity were reported to favour symptom expression of PSTVd (Harris and Browning 1980; Grasmick and Slack 1985). However, it is unknown if this relationship holds true for symptom expression in avocado. Likely symptom expression also depends on the avocado variety and the viroid strain. Moreover, asymptomatic infection of avocados by PSTVd is common. A survey in Peru showed that approximately 21% of asymptomatic trees were infected with PSTVd and at some sites 93–100% of all asymptomatic avocado trees sampled were PSTVd positive (Querci et al. 1995).

The preferred methods sensitive enough for detecting PSTVd is real-time reverse transcription PCR (RT-qPCR) or real-time RT-PCR (Botermans et al. 2020). Usually flowers, leaves and young leaf flushes are used for viroid testing in avocado (Morey-León et al. 2018). Methods have been optimised for testing for both ASBVd and PSTVd in avocado (Nieto-Taype et al. 2014; Mathews et al. 2022). In young trees usually one leaf from each branch that traces back to the trunk should be collected (Mathews et al. 2022).

The concentration of viroids can fluctuate during the season and within an infected plant. However, usually systemic infection of a viroid is detected using a sensitive method such as RT-qPCR no matter where on the tree the leaf samples are taken and in what month of the year the tree is sampled (Geering 2011).

7.2.10 References

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Appendix 1. Summary of taxa excluded at PRA

The following organisms were excluded at the PRA stage. Fifteen species (including two diseases of unknown aetiology) were selected for PRA but upon investigation were excluded and required further rationale that was not suitable for a table (i.e. excluded hazard table in Appendix 2). *Diplodia pseudoseriata* was completed as a full PRA but while this IRA was being prepared *Diplodia pseudoseriata* became known to be present in New Zealand and was thus, included in this appendix.

App 1.1 Avocado Black Streak Disease

Taxonomic notes

Disease of unknown etiology

NZ status

- not known to be present in New Zealand (NZOR, accessed 20/12/2021).
- regulated (ONZPR, accessed 20/12/2021)

Summary of reason for exclusion

Avocado Black Streak Disease can be excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- To date there is no evidence that this disease is caused by biotic stress.
- There is no evidence of graft transmission or by vectors
- Current literature suggests that this disease is a result of abiotic stress and that symptoms can be overturned by good irrigation and fertilization schemes

Supporting information

Avocado black streak (ABS) was first reported in 1934 (Carrillo and Eskalen 2017) . This disease has been predominantly reported in avocado producing regions in California with isolated reports from Florida in the 1990s and outside the United States, in the Canary Islands and in Israel in the 1980s (Marais 2004; Carrillo and Eskalen 2017). However, in the case of Florida and in the Canary Islands, it is known that budwood from symptomatic trees were brought from California (Marais 2004; Carrillo and Eskalen 2017).

Trunk and branch cankers are the most obvious symptoms and are used as the “indicator” symptom of ABS (Ohr and Murphy 1987) . However, using cankers as an indicator symptom has been a subject of debate. The canker has being described as visible when there is accumulation of dry sugar exudate (Marais 2004). When the canker first appears, the resulting lesion under the outer bark is reddish brown and is usually limited to the phloem but can extend past the vascular cambium into the xylem tissue (Carrillo and Eskalen 2017). Cankers often first appear on the lower trunk and underside of lower branches and then later appear higher in the tree (Eskalen and Faber 2016). Initially, the canker formation was attributed to a potential viral etiology, but survey results from Carrillo and Eskalen (2017) suggest that canker formation is likely due to a complex of fungi. One of the hypotheses is that environmental stress triggers symptom development of ABS and this leads to the formation of small open wounds which serve as entry points for secondary, opportunistic fungi (Carrillo and Eskalen 2017). Given the information above, it is uncertain if cankers should be considered as the indicator symptom for this disease.

Other symptoms reported are chlorosis, early bloom, branch dieback, leaf blotching, zinc deficiency, wilting of foliage and rapid death of new growth (Carrillo and Eskalen 2017). This disease usually results in tree death, but if the tree recovers fruit production is impacted (Ohr and Murphy 1987). Affected trees under normal progression of the disease can gradually decline and eventually die, however there are reports of rapid tree collapse. The symptoms associated with ABS are varied and can be attributed to other causes such as abiotic stress and local farm management practices (Ohr and Murphy 1987).

Studies performed by Ohr et al. (1976) indicate that this disease occurs mainly in Guatemalan avocado varieties, including the market dominant “Hass” variety. The disease is known to affect trees of all ages, since symptoms have been observed on trees ranging from 1 year to over 35 years old (Eskalen and Faber 2016). The results of a survey undertaken by Ohr and Murphy (1987) during three consecutive years in two selected orchards in the Ventura and San Diego County, California revealed a 13.2% and 20.1% increase respectively, in the number of diseased trees (Table 7-1).

Table 7-1 Results of a survey performed during three consecutive years (1975 -1977) in two orchards, one in Ventura Country containing 1,731 (10-year-old) Hass trees on mixed rootstocks and the other in San Diego County containing 374 (22-year-old) trees on Topa-Topa rootstocks (Ohr and Murphy 1987).

Year	Location	Percentage (%) infected trees	Number of trees infected
1975	Ventura County	6.7	116
1976		15.1	261
1977		19.9	344
1975	San Diego County	17.1	64
1976		30.2	113
1977		37.2	139

The causal agent/s of avocado black streak is/are unknown. Possible causes that have been investigated include:

Fungi: symptomatic and asymptomatic mature branch samples were obtained from three to five trees within each orchard. The most consistently isolated organisms included *Lasiodiplodia* spp., *Neofusicoccum luteum*, *Diplodia mutila*, and *Phaeoacremonium* spp. However, pathogenicity tests revealed that the lesions and necrosis observed on the mature avocado branches were not consistent with symptoms of ABS (Carrillo and Eskalen 2017). One of the hypotheses is that the isolated organisms are secondary fungi that causes damage upon introduction into the host from open wounds formed during ABS disease development (Carrillo and Eskalen 2017).

Bacteria: antibiotic treated trees still produced symptoms of ABS therefore, infection by bacteria was ruled out (Ohr and Murphy 1987).

Virus and viroids: not detected with electron microscope and no double stranded ribonucleic acid (dsRNA) were detected using polyacrylamide gels. Three separate dsRNA were detected that might indicate a combination of three viruses, but the presence of the dsRNA patterns is not consistent amongst avocado varieties. Field surveys revealed that most avocado varieties have at least one of the patterns and that some have all three (Ohr and Murphy 1987). Further studies to determine dsRNA-ABS correlation were performed by Ohr (unpublished work), but because of the few samples that they were able to analyse (343 trees) it was not possible to support a correlation between dsRNA and ABS.

Graft transmission of dsRNA patterns: Jordan et al. (1983) reported two of the three dsRNA patterns are able to be transmitted from the rootstock to the scion in avocado. The transmission of a condition called “pitting” suggested that some agent could be transmitted from scion to rootstock. However, Marais (2004) considers that graft transmission of dsRNA patterns could not be proven in the Jordan et al. (1983) study because it was not designed to test this due to the difficulties associated with analysing rootstock material. Observations of spread in the field in the past twenty years have not yet been substantiated by graft transmission (Marais 2004).

Nematodes: *Xiphinema americanum* nematodes were collected from ABS and healthy trees, but no correlation was found with ABS (Ohr et al. 1976).

Nutrient deficiency or toxicity: no consistent results (Ohr et al. 1976).

Currently this disease is considered of minor economic importance (Carrillo and Eskalen 2017) but there is still uncertainty regarding the cause of ABS. In recent years, authors have hypothesized that it is possible that this disease is related to moisture stress. After periods of abiotic stress (especially conditions of high salinity and insufficient water), trees died faster, but with improved agricultural management practices (e.g., irrigation and fertilization schemes), trees recovered and symptoms disappeared (Eskalen and Faber 2016; Carrillo and Eskalen 2017).

Efforts to use molecular techniques to trace fungi, bacteria, viruses, and mycoplasma organisms in infected samples and to investigate if there is any correlation with ABS are underway (Carrillo and Eskalen 2017).

Given the evidence above, it is concluded that:

- This disease is likely to be a result of abiotic stress since recent literature report that with improved agricultural management practices (irrigation and fertilization schemes) the trees recovered, and symptoms disappear.
- Symptoms observed on avocado branches (i.e., cankers) are likely to be a result of opportunistic fungi that entered into the host from open cracks caused by abiotic stress. These opportunistic fungi are being assessed in separate pest risk assessments in the current avocado budwood IRA. Therefore, it is considered that the current measures are likely to mitigate the entry of these fungi into New Zealand.
  There is no evidence that supports that this disease is a result of biotic stress.

Due to the unresolved causal agent of ABS, the high uncertainty about its transmission and the recent observations that the symptoms of this disease can be overturned by good irrigation and fertilization practices, it is not justified to consider ABS, at this point, as a hazard requiring further assessment in the avocado budwood IRA. The Emerging Risk System was checked on December 20th, 2021 and no alert has been registered under the Causative Agent Name: ‘Avocado Black Streak Disease’ or ‘Blackstreak’. A monitoring system its being set up for active monitoring of this disease and if further information is available this disease could be reassessed.

Literature search

Searches of Google Scholar and Google were conducted using the search terms “Avocado Black Streak Disease” and “Black Streak in Avocado”. Six pages with a total of 60 results were screened for relevant information and the search stopped after that because unrelated or repeated information started to appear.

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Ohr, H D; Platt, R G; Zentmyer, G (1976) Black streak disease of avocados. *California Avocado Society Yearbook* 60: 177 — 179.

App 1.2 *Botryobasidium perseae*

Scientific name: *Botryobasidium perseae* (R.F. Castañeda) G. Langer (2021)

Order/Family: Cantharellales/Botryobasidiaceae

Other names include: *Haplotrichum perseae* R. F. Castañeda 1996

NZ status

- Not listed in Biota NZ (2022), PPIN (2021) or ONZPR (2021).

Reason for exclusion

Botryobasidium perseae (syn. *Haplotrichum perseae*) can be deemed not to be a hazard and excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- Castañeda (1996) reported the fungus on dead branches of a living avocado tree in Cuba, which is the only record of it associating with *Persea americana* (avocado),
- there is insufficient evidence that the fungus is associated with living *Persea americana* tissue, and insufficient information available on the fungus to carry out a PRA,
- other species in the Botryobasidiaceae family are wood-rotting saprotrophs and none are of economic importance (Cannon and Kirk 2007).

Literature Search

Searches of Google Scholar, CAB abstracts, CPC, USDA fungal database and Google were conducted using the search terms '*Haplotrichum perseae*' and '*Botryobasidium perseae*'.

References

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PPIN (2021) Plant Pest Information Network, Version 5.03.01. Ministry for Primary Industries Internal Database. Accessed June 2021

App 1.3 *Ceratocystis fimbriata* (Ceratocystis blight)

Scientific name: *Ceratocystis fimbriata* Ellis & Halsted (1890)

Taxonomic position: Microascales/Ceratocystidaceae

Synonyms: *Ceratostomella fimbriata* (Ellis & Halst.) J. A. Eilliot (1923), *Endoconidiophora fimbriata* (Ellis & Halst.) R. W. Davidson (1935), *Rostrella coffeae* Zimm. (1900), *Sphaeronaema fimbriatum* (Ellis & Halst.) Sacc. (1892), *Ophiostoma fimbriatum* (Ellis & Halst.) Nannf. (1934)

Taxonomic notes: *Ceratocystis* has been placed into four geographical clades, the Latin American clade (LAC), the North American clade (NAC), the African clade (AFC) and the Asian-Australian clade (AAC) (Holland et al. 2019). *Ceratocystis fimbriata* is in the LAC (Holland et al. 2019) and causes black rot of sweet potato and wilt or cankers on coffee, *Eucalyptus* spp., rubber trees and mango (Holland et al. 2019).

NZ status

- Recorded in New Zealand; however, it seems the strain infecting kumara *(Ipomoea batatas)* is the only strain present (Biota NZ 2022).
- *Ceratocystis fimbriata* (*Ipomoea* strain) is non-regulated (ONZPR 2021)
- *Ceratocystis fimbriata* (strains not in New Zealand) is regulated (ONZPR 2021)

Summary of reason for exclusion

Ceratocystis fimbriata can be deemed not to be a hazard and excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- there is no evidence that *Persea americana* is a host of *C. fimbriata*,
- one record in Portuguese literature states symptoms on dead and dying avocado trees in Brazil showed symptoms similar to those observed by *C. fimbriata* (Mariano and Menezes 1989).
    - this study (Mariano and Menezes 1989) isolated a fungus which had characteristic macroconidia and endoconidia, but the identity of the fungus was not confirmed (by either morphological or sequence confirmation) (Mariano and Menezes 1989).
    - since this study there have been no further records of this host-pathogen association.
    - this study is the only one underpinning the inclusion of avocado as host of *C. fimbriata* in USDA.
- a further literature search produced no records of *C. fimbriata* causing disease in *P. americana*
    - a single record from Portuguese literature states the possibility of *P. americana* being resistant to *C. fimbriata*, but this has also not been confirmed (Fernández et al. 1987).
    - No further studies have been conducted showing this resistance.

Supporting information

Searches of Google Scholar, CAB abstracts, CPC, USDA fungal database and Google were conducted using the search terms '*Ceratocystis fimbriata*' (and synonyms) and '*Persea americana*' (and 'avocado'). The record from USDA refers to Mariano and Menezes (1989) and neither CABI (2021) or EPPO (2021) list *Persea americana* as a host of *C. fimbriata*. In addition, searching the same search terms in Web of Science produced no results.

References

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ONZPR (2021) Official New Zealand Pest Register. MPI public database. https://pierpestregister.mpi.govt.nz/ Accessed 17 March 2021

App 1.4 *Diaporthe pascoei*

Scientific name: *Diaporthe pascoei* R.G. Shivas, Jacq. Edwards & Y.P. Tan (2013)

Order/Family: Diaporthales/Diaporthaceae

Synonyms: None

Known distribution: Australia and Malaysia

NZ status (Lim et al. 2019)

- Not reported to be present in New Zealand (not listed in Biota NZ (2022), accessed 1/11/2021, or PPIN, accessed 21/12/2021)
- Not listed in ONZPR (2021) (accessed 1/11/2021)

Summary reason for exclusion

Diaporthe pascoei can be deemed not to be a hazard and excluded from the avocado Germplasm Import Risk Analysis (IRA) because:

- There is insufficient information available on the fungus to carry out a PRA, and insufficient evidence that the fungus is associated with fruits with stem-end rot disease in *Persea americana*.
- There is only a single report of this species in association with *Perseae americana* fruit
- Pathogenicity test was not done to confirm if the pathogen caused disease symptoms observed on avocado fruit.

Supporting Information

Tan et al. recently described *D. pascoei* and other five new species of *Diaporthe*, on the basis of morphological characteristics and molecular analyses (Tan et al. 2013). *Diaporthe pascoei* was isolated from decayed patches on *P. americana* (avocado) fruit with stem-end rot disease symptoms on one occasion in Australia (Tan et al. 2013). Pathogenicity tests was not carried out to confirmed if *D. pascoei* was responsible for causing disease symptoms on avocado fruit because it did not grow on culture media (Tan et al. 2013). As such, there is no clear information linking this fungus to the symptoms noted, which could be due to other pathogens present at the same time.

Diaporthe pascoei was recently associated with mangos. It was isolated along with other pathogens known to cause stem-end rot disease (*Diaporthe* spp.). Pathogenicity test confirmed *D. pascoei* to cause disease symptoms (stem-end rot) in mango in Malaysia (Lim et al. 2019).

Searches using the search term ‘*Diaporthe pascoei* on avocado’ from Google, Google scholar, CABI and the USDA fungal database found no additional records (other than the single reference above) of *D. pascoei* association with avocado.

References

Biota NZ (2022) Biota of New Zealand: names and classification of bacteria, fungi, land invertebrates and plants. https://biotanz.landcareresearch.co.nz/ Accessed February 2022

Lim, L; Mohd, M H; Zakaria, L (2019) Identification and pathogenicity of *Diaporthe* species associated with stem-end rot of mango (*Mangifera indica* L.). *European Journal of Plant Pathology* 155(2): 687-696.

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App 1.5 Duke 6 stem pitting

Taxonomic notes: disease of unknown aetiology

NZ status

- not known to be present in New Zealand (not listed in NZOR (2021)or PPIN (2021), accessed 6/01/2022)
- not listed in ONZPR (2021) (accessed 6/01/2022)

Summary of reason for exclusion

Duke 6 stem pitting can be excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- the disease is restricted to the variety Duke 6 which is no longer planted commercially anywhere in the world,
- it was identified in South Africa and since then it has not been reported elsewhere,
- no other variety has been reported to have similar symptoms,
- the most recent literature catalogues this disease as a minor problem which rarely impacts plant health and does not reduce yield,
- it appears to be triggered by a source of extreme drought stress,
- there is no indication in the literature of ongoing research to determine the source of the observed symptoms

Supporting information

Duke 6 stem pitting was first reported in the 1980s when the avocado cultivar 'Duke 6' was planted on a commercial scale at Westfalia Estate in South Africa (Roe et al. 1995). The Californian cultivar Duke 6 was known to be a root rot tolerant rootstock and therefore was amongst the cultivars imported to South Africa for evaluation under South African conditions. After 2 to 3 years the trees began dying due to an unidentified disease associated with the formation of depressions or indentations in the stem (stem pitting) (Kremer-Köhne and Köhne 2007).

The first symptom observed was wilting of the leaves which subsequently abscised, leaving the tree almost bare of leaves. This resulted in severe sunburn of bark, twigs and branches. In addition, a very clear pitting, confined to the rootstock, could be observed in the wood when the bark was stripped across the grafting point and protuberances were observed on the bark removed (Moll et al. 1987).

In one orchard in South Africa containing 4,000 avocado trees, of which 2,000 trees where Hass scions grafted on clonal Duke 6 and 2,000 trees where Hass scions grafted on Guatemalan seedling (cv Edranol), the trees that were grafted on Duke 6 started defoliating and declined rapidly after three years of vigorous growth. Later on, this disease spread to further 1,200 trees grafted on Edranol rootstock (Moll et al. 1987). This raised questions about the ability of this disease to spread rapidly. In addition, it was observed that when Duke 6 was budded to other rooststocks (i.e. Duke 7 and G6) a distinct leaf symptom developed within six months and the plants were relatively stunted. The observations above prompted a series of research, to determine the source of the observed symptoms (Moll et al. 1987).

Fungi: Fifteen root and soil samples were taken from diseased avocado trees and fungi isolated on P_{10}VP and PDA agar were incubated at 25℃ in a dark incubator for 10 days. One of the samples tested positive for *Phytophthora cinnamomi* whereas *Fusarium* was isolated from several samples. There were no fungi consistently associated with the diseased samples (Moll et al. 1987). No conclusive results to date regarding this disease being ascribed to fungi.

Bacteria: Fifteen root and soil samples were taken from diseased avocado trees and isolates were made into nutrient agar and incubated at 22 ℃ and compared under a light microscope after being Gram stained. Numerous bacteria were isolated but there was no bacteria consistently associated with the diseased samples (Moll et al. 1987). No conclusive results to date regarding this disease have being ascribed to bacteria.

Nematodes: Five root (10 g per sample) and five soil samples (250 g per sample) were extracted from diseased and healthy trees as well as samples from an adjacent field that had not been disturbed by agricultural practices. No nematode was found distributed consistently throughout the samples and when nematodes were encountered, the numbers were low (Moll et al. 1987).

Insect vectors: No arthropod was found with sufficient consistency to support the hypothesis of spread of the disease by an insect vector (Moll et al. 1987).

Nutrition: Samples of wood were obtained from primary roots, the trunk above and below the grafting point and primary branches from diseased and healthy trees. Zinc levels were higher in diseased trees as opposed to nitrogen, phosphates, potassium, and magnesium levels, which were higher in healthy trees. Levels of all the other nutrient elements (copper manganese, iron and boron) were the same in healthy and diseased trees (Moll et al. 1987). According to Moll et al. (1987), no nutrient was either sufficiently deficient or excessive to cause the symptoms observed

Viruses: Leaf samples from diseased trees from different rootstocks were analysed using the Bar-Joseph et al. (1983) extraction technique and poly-acrylamide gel electrophoresis. Avocado samples known to be infected with sunblotch viroid and *Nicotiana glauca* infected with cucumber mosaic virus were used as positive controls. The cultivar Duke 6 failed to yield any viruses (Moll et al. 1987).

Despite the fact that no foreign RNA could be detected in the cultivar Duke 6, Moll et al. (1987) suggested that the stem-pitting symptoms, the differential host symptom expression, and the observed transmissibility in the field all indicated a viral pathogen. According to da Graca and Trench (1985), not detecting dsRNA in Duke 6 does not eliminate the possibility that the causal virus agent might be present. It could be that the levels are too low to detect, or the sample material might need to be increased above the levels already tested, or perhaps the causal agent is a DNA virus.

The observations in South Africa prompted research in Israel on similar disease problems in local avocado plants. After screening several top grafted trees with different varieties, they concluded that these phenomena were not caused by a particular disease (Bar-Joseph et al. 1987).

It has been suggested that Duke 6 stem pitting symptom is similar to the avocado black streak disease (another disease of unknown aetiology) and that it could be possibly that the disease reported in California, is the same as the one reported in South Africa. However this has been rebuked by researchers in California by saying that the pitting symptom is not observed throughout all the trees showing black streak cankers and that Duke 6 is not a variety of common use as there are more resistant cultivars developed since the first release of Duke 6 in the 1980s (Moll et al. 1987).

Currently, this disease is catalogued as a minor problem which rarely impacts plant health and does not reduce yield (Dann et al. 2013). It appears that this disease is exacerbated by severe stress conditions, like prolonged drought periods (i.e. severe drought in 1984 in South Africa) (Moll et al. 1987).

The cultivar Duke 6 is no longer commercially planted in South Africa or other countries. As soon as the unidentified disease was detected in the 1980's, all the Duke 6 plant material (including mother plants) were destroyed in an attempt to eradicate the disease and therefore this rootstock no longer exists in South African orchards (Kremer-Köhne and Köhne 2007).

Due to the unresolved causal agent of Duke 6 Stem Pitting, the high uncertainty about its transmission, its restricted distribution and that no recent information regarding new avocado varieties showing similar symptoms, it is not justified to consider this 'disease' as a hazard. The Emerging Risk System was checked on January 6, 2022 and no alert has been registered under the Causative Agent Name: 'Duke 6 stem pitting". A monitoring system has been set up for active monitoring of this disease and if further information is available this disease could be reassessed.

Literature search

Searches of Google Scholar and Google were conducted using the search terms "Duke 6 Stem pitting" and "Duke 6 Stem pitting" AND "avocado". Three pages with a total of 30 results were screened for relevant information and the search stopped after that because unrelated (e.g., 'Duke university' or 'Cherry Stem Pitting') or repeated information started to appear.

References

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Bar-Joseph, M; Rosner, A; Moscovitz, M; Hull, R (1983) A simple procedure for the extraction of double-stranded RNA from virus-infected plants. *Journal of Virological Methods* 6(1): 1-8.

da Graca, J; Trench, T (1985) Evidence of dsRNA in South African avocados. In *South African Avocados Growers' Association* (Vol. 8, pp. 72-74).

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App 1.6 *Guignardia foeniculacea*

Scientific name: *Guignardia foeniculacea* (Mont.) Arx & E. Müll. (1954)

Taxonomic position (order/family): Botryosphaeriales /Phyllostictacea

Synonyms: *Sphaeria foeniculacea* Mont. (1849), *Diaporthe foeniculacea* (Mont.) Niessl. (1879)

NZ status

- Absent from New Zealand (Biota NZ 2022) (accessed Sept 2021).
- Not listed in ONZPR (2021) (accessed Sept 2021).

Summary of reason for exclusion

Guignardia foeniculacea (syn. *Diaporthe foeniculacea*) can be deemed not to be a hazard and excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- the pathogen reported by Guarnaccia et al. (2016) on avocado in Italy is most likely *Diaporthe foeniculina* (already present in New Zealand) and,
- there is insufficient evidence that *Guignardia foeniculacea* is associated with avocado

Supporting information

Guarnaccia et al. (2016) reported *Diaporthe foeniculacea* as a mildly virulent pathogen on avocado branches in Italy. Guarnaccia et al. (2016) refer to *Diaporthe foeniculacea* and *D. foeniculina* as synonyms but this synonymy is not supported by Manaaki Whenua Landcare Research (P. Johnston, pers. comm.) or by the Biota New Zealand database (accessed Sept 2021) (Biota NZ 2022), or based on information in a study by Udayanga et al. (2014). Furthermore, taxonomic trees based on molecular sequence alignments show that the CBS isolate (CBS-111553) referred to by Guarnaccia et al. (2016) as *D. foeniculacea* actually corresponds with *D. foeniculina* in the CBS-KNAW culture collection and is closely related to an isolate from blackcurrants (*Ribes*) in New Zealand (Udayanga et al. 2014). This indicates that the pathogen reported by Guarnaccia et al. (2016) on avocado in Italy is *Diaporthe foeniculina*, which is already present in New Zealand (Biota NZ 2022). This is the only study that has reported *Diaporthe foeniculacea* (syn. *Guignardia foeniculacea*) as a pathogen of avocado.

Literature Search

Google and Google Scholar were used to search for information on *Guignardia foeniculacea* and its synonyms (*Sphaeria foeniculacea, Diaporthe foeniculacea*). *Diaporthe foeniculina* and its synonyms (*Phoma foeniculina, Phomopsis theicola* and *Diaporthe neotheicola*) were also searched for in Google and Google Scholar. The USDA fungal database (Farr and Rossman 2021) and CBS-KNAW culture collection database were also used to search for species and isolates.

References

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App 1.7 *Lasiodiplodia mahajangana*

Scientific name: *Lasiodiplodia mahajangana* Begoude, Jol. Roux & Slippers

Order/Family: Botryosphaeriales/Botryosphaeriaceae

Synonyms: None

Known distribution: Australia, Madagascar, South Africa, USA and Venezuela

NZ status

- Not reported to be present in NZ (not listed in Biota NZ (2022), accessed 2/11/2021, or PPIN (2021), accessed 21/12/2021)
- Not Listed in ONZPR (2021) (accessed 2/11/2021)

Summary reason for exclusion

Lasiodiplodia mahajangana can be deemed not to be a hazard and excluded from the avocado Germplasm Import Risk Analysis (IRA) because:

- A pathogen was isolated on rotten avocado fruit showing stem-end rot disease symptoms. The isolated pathogen showed some sequence similarity with *Lasiodiplodia mahajangana*
- It has not been associated with avocado budwood.

Supporting Information

A pathogen was isolated on avocado fruits with stem-end disease symptoms in South Africa (Majola 2020). Majola (2020) inferred from molecular analysis that the isolated avocado stem-end rot pathogen was a member of the genus *Lasiodiplodia*, possibly *L mahajangana*, but concluded that the identity of the species requires further investigation and confirmation.

So far, this inconclusive evidence is the only report that associates *L. mahajangana* with avocado.

Lasiodiplodia mahajangana has been associated with about 20 different host plants from five countries. In a study in the Kimberley region of Western Australia, it was the dominant species sampled among native trees in an isolated habitat (Sakalidis et al. 2011). However, the only report that associates *L. mahajangana* with avocado is inconclusive and requires confirmation is strong enough as a reason to exclude this species as a hazard.

Searches using the search term '*Lasiodiplodia mahajangana* on avocado' from Google, Google scholar, CABI and the USDA fungal database found no additional record (other than the references above) of *L. mahajangana* association with avocado.

References

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Sakalidis, M L; Ray, J D; Lanoiselet, V; Hardy, G E S; Burgess, T I (2011) Pathogenic Botryosphaeriaceae associated with *Mangifera indica* in the Kimberley region of Western Australia. *European Journal of Plant Pathology* 130(3): 379-391.

App 1.8 *Microthia havanensis*

Scientific name: *Microthia havanensis* (Bruner) Gryzenh. & M.J.Wingf.

Taxonomic position (order/family): Diaporthales/Cryphonectriaceae

Taxonomic notes: This species was initially described by Bruner (1916) as *Endothia havanensis* Bruner, and later named *Cryphonectria havanensis* (Bruner) M.E. Barr. The results from phylogenetic analyses and morphological comparisons performed by Gryzenhout et al. (2006), demonstrated that *C. havanensis* do not reside under *Cryphonectria* but under the genus *Microthia*. This fungus and *Amphilogia gyrosa* have been considered as synonyms when the latter fungus was still known as *Cryphonectria gyrosa* (Hodges 1980), but Gryzenhout et al. (2006) has demonstrated that these species should be considered as distinct species. The name *C. havanensis* has also been used for collections of a fungus from *Eucalyptus globulus* in Japan (Kobayashi 1970). However, DNA sequence comparisons performed by Myburg et al. (2004) showed that *C. havanensis* in Japan is the same as *Cryphonectria nitschkei* (G.H. Otth) M.E. Barr. It is unclear if *C. havanensis* collected from Cuba is synonym to *C. nitschkei* because Myburg et al. (2004) did not consider this comparison in their study.

NZ status

- The species is not known to be present in New Zealand (not listed in Biota NZ (2022), accessed 8/12/2021, or PPIN (2021), accessed 17/12/2021)
- The species is regulated under the name *Cryphonectria havanensis* (ONZPR 2021) (accessed 8/12/2021)

Summary of reason for exclusion

Microthia havanensis can be excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- There is insufficient evidence that *Microthia havanensis* is associated with the commodity (i.e., healthy looking budwood/leafless cuttings of *Persea americana*).

Supporting information

The initial description of *Microthia havanensis*, performed by Bruner (1916) under the name *Endothia havanensis,* was performed by sourcing the fungus from *Eucalyptus* twigs. The author mentioned that the habitat of this fungus is dead bark of *Eucalyptus* followed by a list of hosts of which *Persea gratissima* (syn. *Persea americana*) is included. There is no indication from this study that *M. havanensis* is associated with the commodity.

Furthermore, Gryzenhout et al. (2006) mentioned that this fungus was found on dead branches of mango (*Mangifera indica*) and avocado (*Persea gratissima*) that were lying on the ground in the vicinity of Eucalyptus trees and refers to Bruner (1916), who first described this fungus.

The author (Roux et al. 2020) mentioned that this fungus is known as a saprobe mainly in *Eucalyptus* but also in other hosts like avocado and also refers to Bruner (1916).

Literature search

Searches of Google Scholar and Google were conducted using the search terms "Microthia havanensis", "Cryphonectria havanensis" and "Endothia havanensis" combined with the search terms "avocado", "aguacate" and "Persea americana". In addition, searches were performed in Index Fungorum, USDA Fungal databases to determine host and commodity association and GBIF database to obtain the taxonomy of the fungus.

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Hodges, C (1980) The taxonomy of *Diaporthe cubensis*. *Mycologia* 72(3): 542-548.

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Roux, J; Kamgan Nkuekam, G; Marincowitz, S; van der Merwe, N; Uchida, J; Wingfield, M, et al. (2020) Cryphonectriaceae associated with rust-infected *Syzygium jambos* in Hawaii. *MycoKeys* 76: 49-79.

App 1.9 *Physalospora abdita* and *Physalospora perseae*

Scientific name: *Physalospora abdita* (Berk. & M.A.Curtis) N.E.Stevens

Taxonomic position (order/family): Amphisphaeriales (Xylariales)/Hyponectriaceae

Taxonomic notes: The exact taxonomic position is uncertain. Currently, it is placed under synonymy with *Physalospora fusca*, *Barriopsis fusca*, and *Botryosphaeria fusca* (Biota NZ 2022) based on Weiss (1950). *Physalospora fusca* was incorrectly recorded from New Zealand based on specimens later identified as *Sphaeropsis citrigena* (Biota NZ 2022).

NZ status

- Uncertain due to taxonomy (Biota NZ 2022).
- Uncertain regulatory status due to taxonomy (ONZPR 2021).

Summary of reason for exclusion

Physalospora abdita can be deemed not to be a hazard and excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- the pathogen reported as *Physalospora abdita* on avocado was described morphologically and there is no sequence data available to clarify its taxonomic position,
- many *Physalospora* species that were described morphologically have since been moved into different genera using molecular methods, such as in *Botryosphaeria, Lasiodiplodia, Diplodia, Mycospaerella, Guignardia, Barriopsis, Phyllosticta, Stigmatula, Colletotrichum, Polystigma, Vestergrenia, Pseudomassaria* and *Phyllachora* (Biota NZ 2022),
- symptoms of dieback and canker on avocado are more likely attributed to other fungal genera, such as *Botryosphaeria, Lasiodiplodia, Diplodia, Mycospaerella, Phyllachora* and *Colletotrichum* (Valencia et al. 2019; Biota NZ 2022),
- a literature search found no reports of *Physalospora* species associated with avocado since the use of molecular identification methods (1980s).

Scientific name: *Physalospora perseae* Doidge 1923

Order/Family: Amphisphaeriales (Xylariales)/Hyponectriaceae

Taxonomic notes: *Physalospora perseae* was described in 1922 as an avocado pathogen but the name has barely been used since (Doidge 1922) and has only been found in some poorly referenced lists of plant diseases. In the 1920s and 30s it was considered to be the pathogen on avocado causing black spot, leaf spot, die-back or wither tip and canker on avocado (Carpenter 1931) and described as the higher stage of "the avocado *Gloeosporium* or *Colletotrichum*". It was intercepted in the United States from South Africa in the 1930s (USDA 2016). In 1934 the species was redescribed, considered to be a typical *Melanops*, and renamed to *Melanops perseae* (Doidge) Petr. 1934. Since then no *Melanops* or *Physalospora* species have been reported from avocado (Jiang et al. 2018). *Physalospora perseae* or *Melanops perseae* have never been used in a New Zealand context (Biota NZ 2022). There is no genetic data available to resolve the exact taxonomic position of the species.

NZ status:

- Uncertain due to taxonomy (Biota NZ 2022).
- Uncertain regulatory status due to taxonomy (ONZPR 2021).

Summary of reason for exclusion

Physalospora perseae can be deemed not to be a hazard and excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- the pathogen reported as *Physalospora perseae* on avocado was described morphologically and there is no sequence data available to clarify its taxonomic position,

- many *Physalospora* species that were described morphologically have since been moved into different genera using molecular methods, such as in *Botryosphaeria, Lasiodiplodia, Diplodia, Mycospaerella, Guignardia, Barriopsis, Phyllosticta, Stigmatula, Colletotrichum, Polystigma, Vestergrenia, Pseudomassaria* and *Phyllachora* (Biota NZ 2022),
- nowadays symptoms of dieback and canker on avocado are more likely attributed to other fungal genera, such as *Botryosphaeria, Lasiodiplodia, Diplodia, Mycospaerella, Phyllachora* and *Colletotrichum* (Valencia et al. 2019; Biota NZ 2022),
- a literature search found no reports of *Physalospora* species associated with avocado since the use of molecular identification methods (1980s).

Supporting information

There is very little information associated with *Physalospora abdita* and *Physalospora perseae*. Both species had been reported as pathogens on avocado prior to molecular identification methods. There is no sequencing data available to clarify their taxonomic position.

When the two species were initially described as pathogens on avocado (Doidge 1922; Weiss 1950) they were referred to as *Physalospora* because their symptoms and morphology resembled these of *Physalospora cydoniae* on the limbs and twigs of apple trees. Many *Physalospora* species that were described morphologically have since been moved into different genera using molecular methods, such as in *Botryosphaeria, Lasiodiplodia, Diplodia, Mycospaerella, Guignardia, Barriopsis, Phyllosticta, Stigmatula, Colletotrichum, Polystigma, Vestergrenia, Pseudomassaria* and *Phyllachora* (NZFungi2 2021).

Literature Search

Searches of Google Scholar, CAB abstracts, CPC, ICMP, USDA fungal database, CBS-KNAW culture collection database and Google were conducted using the search terms *'Physalospora abdita',* '*Sphaeria abdita*', '*Physalospora perseae*' and '*Melanops perseae'* 60. The last *Physalospora* species record on avocado was likely an interception of *Physalospora perseae* from South Africa in 1935 (USDA 2016), although a pathogen resembling *Physalospora perseae* was described from Morocco in the 1960s and synonymised with *Macrophoma allospora* (Rieuf and Yeasca 1970). No other specimen records or cultures have been found.

Symptom development

In the original publication by Doidge (1922) the plants which were inoculated with *Physalospora perseae* spores soon showed signs of infection. The tissues around the incision became discoloured and sunken; in some cases the affected area slowly increased in size for a few weeks until it was about 1 cm. After six months typical conidia were produced. Based on the original publication by Doidge (1922) it seems likely that *Physalospora perseae* would be detected in PEQ due to symptom development within weeks.

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App 1.10 *Plagiostoma perseae*

Scientific name: *Plagiostoma perseae* Hyde (1995)

Taxonomic position (order/family): Diaporthales / Gnomoniaceae

Synonyms: nil

NZ status: Not known to be present in New Zealand: not listed in (NZOR 2021; Biota NZ 2022), or PPIN (2021). Organism is not listed in ONZPR (2021).

Summary of reason for exclusion

Plagiostoma perseae can be excluded as a hazard on avocado budwood (as per the commodity description) because:

- This fungus was described from a single isolate from living leaves of avocado (*Persea americana*) in Papua New Guinea (Hyde 1995). The isolate was collected in 1992 and the aetiology of the disease was not tested. Therefore, it cannot be verified that the fungus detected on the avocado leaves was the cause of disease symptoms.
- There were no further reports found of this fungus being detected on avocado or any other host species. Therefore, there is insufficient evidence that this fungus is pathogenic or has an association with the commodity.

Supporting information:

This fungus was described in association with leaf spots on the upper and lower surfaces of living avocado leaves in Papua New Guinea (Hyde 1995). Irregular lesions were up to 7mm in diameter, brown and necrotic, with blackened centres were reported. Ascomata occurred mostly on the lower surface of the leaf. Hyde (1995) does not report if the fungus was observed on multiple leaves of the avocado plant, or if multiple plants appeared to be infected. No testing of pathogenicity or aetiology was reported.

A literature search in Google and Google Scholar using the search term “*Plagiostoma perseae*” did not yield any other occurrences of this fungus on avocado or any other host.

The genus *Plagiostoma* are microscopic fungi that are often found on the leaves, stems, twigs and branches of a variety of host plants (Mejía et al. 2011). They are most prevalent in temperate regions of the northern hemisphere. Some *Plagiostoma* species are reported to cause plant disease and may infect hosts asymptomatically. They are particularly noted as a cause of cankers on willows (*Salix spp.*) and poplars (*Populus spp.*). The genus may also be an important endophyte. A literature search in Google and Google Scholar using the search terms “*Plagiostoma*” AND “avocado” OR “*Persea americana*” and “*Plagiostoma*” AND “Lauraceae” found no association of fungi within this genus with avocados or any plant species in the Lauraceae family.

References:

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App 1.11 *Pseudomonas syringae* pathovar (causing avocado bacterial blast)

Scientific name: *Pseudomonas syringae* van Hall, 1902

Taxonomic position (order/family): Pseudomonadales / Pseudomonadaceae

Taxonomic notes: Other names include: *Phytomonas vignae; Pseudomonas holci; Pseudomonas japonica; Pseudomonas medicaginis; Pseudomonas striafaciens var. japonica; Pseudomonas syringae pv. japonica; Pseudomonas vignae* (EPPO 2020)(accessed 23/1/2022). The *'P. syringae* complex*'* encompasses up to 10 *Pseudomonas* species (Arnold and Preston 2019), about 400 strains with known genome (Dillon et al. 2017), and over 60 pathogenic variants (pathovars) with different host ranges (Bull et al. 2010; Arnold and Preston 2019). A pathovar (pv.) is defined as a bacterial strain or set of strains with the same or similar characteristics that is differentiated from other strains of the same species or subspecies on the basis of distinctive pathogenicity to one or more plant hosts (Xin et al. 2018). *Pseudomonas syringae pv. syringae* is a pathovar originally isolated from lilac (genus *Syringa*) but which has subsequently been shown to share hosts in common from many dicotyledonous and monocotyledonous hosts. Bradbury (1986) attributed all hosts of *P. syringae* to *P. syringae* pv. *syringae* and in so doing almost certainly included hosts of unidentified but related pathovars (Bradbury 1986). There are more than 100 strains of *P. syringae pv. syringae* (Scortichini et al. 2003).

NZ status:

- *Pseudomonas syringae* is known to be present in New Zealand, with 61 identified strains (potential synonyms) recorded (NZOR 2021) (accessed 24/1/2022)
- *Pseudomonas syringae pv. syringae* is non-regulated (ONZPR 2021) (accessed 24/1/2022,)
- *Pseudomonas syringae,* and all pv's*,* is not assessed, it is non-regulated for strains present in New Zealand and pv. nerii, and it is regulated for strains not present in New Zealand and pv. papulans (ONZPR 2021) (accessed 24/1/2022)

Summary of reason for exclusion:

The *P. syringae* pathovar causing avocado bacterial blast can be excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- Evidence was found that a *P. syringae* pathovar causing bacterial blast is already present in New Zealand.
  - There is no available evidence suggesting the existence of multiple *P. syringae* pv's causing avocado bacterial blast.
  - The current evidence does not justify concerns about potential exotic *P. syringae* avocado pv's causing avocado blast and impacts other than negligible in New Zealand.

Supporting information:

Pseudomoas syringae is ubiquitous, and particularly abundant in habitats associated with water (Morris et al. 2013). It is found in rain, rivers, alpine streams, lakes, wild plants, as well as in epilithic biofilms (Morris et al. 2008), and has an important role in processes leading to rain and snowfall (Morris et al. 2013). As a plant pathogen, *P. syringae* can infect a wide range of species (Bull et al. 2010). There are both non-pathogenic and pathogenic strains of *P. syringae* (Xin et al. 2018). Collectively, the pathogenic strains (pathovars) of *P. syringae* infect almost all economically important crop species, which makes *P. syringae* one of the most common plant pathogens (Xin et al. 2018). Yet, each pathovar infects a characteristic group of host plants (Xin et al. 2018).

There are two avocado diseases associated with *Pseudomonas syringae* found in the literature: avocado blast, affecting the fruit (Lunar and Fucikovsky 1987), and avocado bacterial canker, affecting the plant (trunk and branches) (Korsten and Kotzé 1987; Marais 2004). Both avocado blast and canker were attributed to bacterial complexes including *P. syringae pv. syringae* and other bacteria (Korsten and Kotzé 1987; Lunar and Fucikovsky 1987; Korsten and Towsen 1997; Marais 2004), and *P. syringae pv. syringae* was listed as a potential exotic pest threats to the New Zealand avocado industry (New Zealand Avocado 2021). When first described in California, avocado fruit blast was attributed to *Bacterium citriputeale* (Smith 1926). The correct name for the blast organism was later identified as *P. syringae* (Schroth 1981). After its description in California (Smith 1926), avocado fruit blast symptoms were later described in Israel (Volcani 1946), and in Mexico (Lunar and Fucikovsky 1987).

Lunar and Fucikovsky (1987) reported that the avocado blast was characterized by appearance of dark brown irregular areas which merged and covered a significant part of the maturing avocado fruit. Affected areas took on a variety of forms. In some cases, lesions were raised above the surface of the fruit and were frequently located in the central or distal part. In other cases, lesions were not raised and were frequently depressed. In later stages, lesions cracked, exposing the mesocarp tissue. As these dark lesions enlarged, the mesocarp tissue softened and became sunken. Infected avocado fruits were randomly distributed throughout the crown of 10-year-old trees. In advanced stages of infection, the avocado fruit produced a brown exudate from the lenticels. Dried exudate had a whitish appearance and dissolved rapidly with rain (Lunar and Fucikovsky 1987). The bacteria associated with diseased fruits were predominantly *Pseudomonas syringae pv. syringae*; however, *Erwinia herbicola* (*Pantoea agglomerans*) was also present and could have been accountable for a portion of the damage (Lunar and Fucikovsky 1987).

There is evidence that a *P. syringae* pathovar causing bacterial blast is already present in New Zealand. The International Collection of Microorganisms from Plants (ICMP) includes isolates of *P. syringae pv. syringae* associated to avocado diseases in New Zealand. One culture (ICMP 458) was isolated in Auckland (Mt Albert) from avocado “blast” in 1955. The other culture (ICMP 7405) was isolated in the Bay of Plenty (Te Puke) from avocado canker, a “twig lesion”, in 1981. A third reported record (Bevan Weir pers. comm) refers to an isolate of *P. syringae* associated to avocado canker and not pathogenic to lilac. These records suggest that three *P. syringae* pv’s associated to avocado are already present in New Zealand, with one *P. syringae* pv. isolated from avocado with blast symptoms, and possibly two *P. syringae* pv’s isolated from avocado with canker symptoms.

There is no available evidence suggesting the existence of multiple *P. syringae* pv’s causing avocado bacterial blast. There are no sequence data available to clarify the taxonomic position of the *P. syringae* pv’s associated to the avocado disease symptoms reported overseas and the *P. syringae* pv’s already present in New Zealand (Kerry Everett, Bevan Weir pers. comm). Given the uncertainty, potential testing options were suggested by both Plant And Food Research (Kerry Everett pers. comm) and Maanaki Whenua - Landcare Research (Bevan Weir pers. comm). There could be concerns about the existence of exotic *P. syringae* pv’s causing avocado bacterial blast and other impacts. These concerns could stem from the huge diversity of the *‘P. syringae* complex*’* and its changing taxonomy (Dillon et al. 2017), and from the severe impacts of *Pseudomonas syringae pv. actinidiae (Psa)* on the kiwifruit industry (Vanneste 2017).

The current evidence does not justify concerns about potential exotic *P. syringae* avocado pv’s causing avocado blast and impacts other than negligible in New Zealand. In fact, in almost a century since its description, the disease “avocado blast” was reported sporadically and on a local orchards (Smith 1926; Volcani 1946; Lunar and Fucikovsky 1987), and no original articles referring to avocado blast could be found after Lunar and Fucikovsky’s work published in 1987 (Dann et al. 2013). Avocado blast is not listed in a recent review of the avocado diseases, listing avocado canker (Marais 2004). When avocado blast was first described, it was noted that the disease had limited distribution and little significance (Smith 1926). Also the avocado canker attributed to *P. syringae* pv. *syringae* was described as having little significance (Korsten and Kotzé 1987). Although avocado canker was observed in all major avocado-growing areas of South Africa, it had no economic importance to the industry because of the small percentage of trees infected (Korsten and Kotzé 1984). Neither avocado blast nor avocado canker are listed among the avocado diseases by EPPO (EPPO, accessed 24/1/2022).

Literature search:

Searches of Google Scholar and Google were conducted using the search terms " *Pseudomonas*", " *Pseudomonas syringae*", combined with the search terms "avocado blast" "avocado", “aguacate”, “*Persea*” and "*Persea americana*". In addition, searches were performed in the PPIN and ICPM databases.

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App 1.12 *Rhizoctonia solani*

Scientific name: *Rhizoctonia solani* J. G. Kühn 1858

Taxonomic position (order/family): Cantharellales/Ceratobasidiaceae

Synonyms: *Corticium sasakii* (Shirai) H. Matsumoto 1934, *Corticium solani* (Prill. & Delecr.) Bourdot & Galzin 1911, *Corticium vagum* var. *solani* Burt 1903, *Hypochnus cucumeris* A. B. Frank 1883, *Hypochnus filamentosus* Pat. 1891, *Hypochnus sasakii* Shirai 1906, *Hypochnus solani* Prill. & Delacr. 1891, *Pellicularia filamentosa* (Pat.) D. P. Rogers 1943, *Thanatephorus cucumeris* (A. B. Frank) Donk 1956, *Thanatephorus sasakii* (Shirai) C. C. Tu & Kimbr. 1978

Taxonomic notes: *Rhizoctonia solani* is a large species complex composed of genetically distinct groups with diverse life histories (Cubeta and Vilgalys 1997). These groups are known as anastomosis groups (AGs) and differ in their ability to cause disease on crops (Cubeta & Vilgalys 1997, Windels et al. 1994). There are at least 14 AGs within the *R. solani* complex (Cubeta and Vilgalys 1997; Zheng et al. 2019). AGs have been further subdivided into subgroups that differ in their biochemical, genetic or pathogenic characteristics (Cubeta and Vilgalys 1997).

Thanatephorus cucumeris is the teleomorph of *Rhizoctonia solani* (Adams and Butler 1978; Strauss et al. 2000).

NZ status:

- Recorded as present in New Zealand (Biota NZ 2022).
    - It is unknown which AG the *Persea americana* isolate belongs to.
    - AG-3PT, AG-2-1 and AG-6 are known to be present in New Zealand (Das 2013; Biota NZ 2022)
- Strains of *Thanatephorus cucumeris* that are recorded as present in New Zealand are non-regulated (ONZPR 2021).
- Strains of *Thanatephorus cucumeris* that are recorded as absent from New Zealand are regulated and unwanted (ONZPR 2021).

Summary of reason for exclusion

Rhizoctonia solani can be deemed not to be a hazard and excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- *Rhizoctonia solani* is primarily a soilborne pathogen that has only been reported to infect roots, seeds and young seedlings of *Persea americana*, and
- a literature search found no reports to suggest that *R. solani* can be associated with the commodity, *Persea americana* budwood.

Supporting information

Rhizoctonia solani has an extremely large host range across multiple plant families with symptoms specific to the host (i.e. symptoms are not the same across species) (Chase 1991; PPIN 2021) and has been isolated from 46 species in New Zealand (PPIN 2021). Symptoms can include stem and root rot and leaf blight but this depends on the AG (Chase 1991). Specifically, AG2-1 and AG4 are associated with stem and root rot diseases in dicots (e.g. Brassicaceae species), while AG8 causes root rot in monocots (Williamson-Benavides and Dhingra 2021). In general, the first four AGs (AG-1, -2, -3, and -4) cause important diseases in plants worldwide, whereas the remaining AGs (AG-5, -6, -7, -8, -9, -10, -11, -12) are less destructive and generally have a restricted geographic distribution (Williamson-Benavides and Dhingra 2021).

Rhizoctonia solani was first reported being pathogenic on *Persea americana* in 1960 (Mircetich and Zentmyer 1960). *Rhizoctonia solani* causes root and seed rot in *P. americana* (González-Sánchez et al. 2010; Horst 2013). Specifically, the fungus invades seedlings through cotyledons (leaves at first emergence) and can cause rot of young roots and shoots (Mircetich and Zentmyer 1960; Zentmyer

1961). In some cases the embryo will be killed and germination does not occur (Mircetich and Zentmyer 1960; Zentmyer 1961).

If infection occurs after primary root formation, the pathogen will attack the youngest part of the root causing brown lesions and will spread until the root system has been destroyed (Mircetich and Zentmyer 1960; Zentmyer 1961; Snyman and Darvas 1983). It may occassionally infect small roots on adult trees, but these roots usually regenerate (Mircetich and Zentmyer 1960; Zentmyer 1961). The disease primarly occurs in seed flats in nurseries or in seed beds in the field (Mircetich and Zentmyer 1960; Zentmyer 1961).

Literature Search

Searches of Google Scholar, CAB abstracts, CPC, ICMP, USDA fungal database, CBS-KNAW culture collection database and Google were conducted using the search terms ['*Rhizoctonia solani*' OR '*Thanateporus cucumeris*' AND '*Persea americana*' OR 'avocado']. Results associated with *P. americana* refer to symptoms described above. There have been no significant outbreaks of this pathogen in *P. americana* since the 1960s and 1970s. Current literature focuses on biological control by bacteria (e.g. González-Sánchez et al. 2010).

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App 1.13 *Thielaviopsis* sp.

Scientific name: *Thielaviopsis* sp. Went

Taxonomic position: Microascales/ Ceratocystidaceae

Synonyms: n/a

NZ status: At least one species in this genus, *T. ethacetica*, is present in New Zealand (NZOR 2021). Of the species in this genus listed in ONZPR (2021), *T. quercina* (syn. *Bretziella fagacaerum*) and *T. punctulata* (syn. *Ceratocystis radicicola*) are listed as regulated, *T. thielavioides* (syn. *Chalara thielavioides*) and *T. basicola* are listed as non-regulated, and *T. paradoxa* is listed as not assessed.

Summary of reason for exclusion

Thielaviopsis sp. can be deemed not to be a hazard and excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- There is insufficient evidence that avocado is a host of any species in the genus *Thielaviopsis*.

Supporting information

Thielaviopsis sp. reported as "bark rot" of avocado by Alvarez (1976) is the only record of association with avocado. This association is based on morphological identification from herbarium specimens, so the identity of this fungal isolate cannot be verified. The association of *Thielaviopsis sp.* with *Persea americana* stated in the USDA Fungal Database refers to the same source by Alvarez (1976). Based on the lacking information provided by this source, and that no other instances of an association with avocado being found in the literature, this genus should not be considered a hazard on avocado budwood. Some species in this genus are economically significant pathogens known to cause root or post-harvest rot (i.e., *T. paradoxa* and *T. basicola*), but have not been reported on any part of the avocado plant (Hewajulige and Wijesundera 2014; Zepeda-Giraud et al. 2020; Williamson-Benavides and Dhingra 2021).

Literature search

Searches of Google Scholar and Google were conducted using the search terms "*Thielaviopsis*" AND "avocado" OR "*Persea americana*". Searches in CPC, EPPO and USDA fungal database used the search terms *"Thielaviopsis*".

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App 1.14 *Verticillium alboatrum* (Verticillium wilt)

Scientific name: *Verticillium alboatrum* (*Verticillium albo-atrum*) Reinke and Berthold (1879).

Taxonomic position (e.g., order/family): Glomerellales/Plectosphaerellaceae.

Taxonomic notes: This organism was split into three different species (*V. alboatrum*, *V. alfalfae* and *V. nonalfalfae*) in a recent taxonomic revision of *Verticillium* (Inderbitzin et al. 2011). Following this taxonomic revision some pathogenic isolates previously identified as *V. alboatrum* were confirmed to be *V. nonalfalfae* (Kasson et al. 2014; Mellow et al. 2019). All three *Verticillium* species are known to occur in New Zealand (NZOR 2021).

Synonyms: *Verticillium albo-atrum* var. *caespitosum* Wollenweber (1929), *Verticillium albo-atrum* var. *tuberosum* Rudolphi.

NZ status: Recorded in New Zealand (NZOR 2021) and non-regulated except for some unspecified severe strains (ONZPR 2021).

Summary of reason for exclusion:

Verticillium alboatrum can be excluded from the Avocado Germplasm Import Risk Analysis (IRA) because:

- *V. alboatrum* is known to occur in New Zealand (NZOR 2021).
- The severe regulated strains of *V. alboatrum* are most likely limited to hops (*Humulus lupulus*), hence are not associated with the commodity.
- There are no records of avocado-associated *V. alboatrum* strains which may pose a risk if not present in New Zealand.
- Recent studies of Verticillium wilt disease in avocado associated the disease with *V. dahliae* rather than with *V. alboatrum.*

Supporting information:

A few commodities are known to be associated with different strains of *V. alboatrum* and some of these strains are regulated in New Zealand (ONZPR 2021). The regulated strains are likely related to a group of *V. alboatrum* strains from England and Slovenia which have been observed to be severely pathogenic on hops (Radišek et al. 2003; Radišek et al. 2006; Cregeen et al. 2015); these strains are not known to be present in New Zealand. In addition, no evidence was found in the literature to suggest that the hops-associated strains are pathogenic on avocado or other host plants. While commodities like hops are known to be associated with some strains of *V. alboatrum*, no evidence was found to indicate that there are strains specifically associated with avocado. In contrast, previous reports indicate that *V. alboatrum* from avocado equally infects other hosts e.g. tomato, pepper and eggplant (Zentmyer 1949; McMillan 1976; Zentmyer 1984; Luna and Fucikovsky 1987).

Before 1980, there was no clear distinction between *V. dahliae* and *V. alboatrum* (Fitzell et al. 1980) thus some reports of Verticium wilt disease in avocado may have erroneously been attributed to *V. alboatrum*. Recent studies of Verticillium wilt disease of avocado are associated with *V. dahliae* rather than *V. alboatrum* (Dann et al. 2010; Markakis et al. 2014; Haberman et al. 2020; Ramírez-Gil and Morales-Osorio 2020, 2021) suggesting that *V. alboatrum* is not a significant pathogen of avocado and *V. dahliae* may have been misidentified as *V. alboatrum* in the past.

Literature Search

Google and Google Scholar were used to search for information on *Verticillium alboatrum* and its synonyms (*Verticillium albo-atrum* var. *caespitosum* and *Verticillium albo-atrum* var. *tuberosum*). Search terms include “Verticillium wilt disease”, “Avocado”, “*Persea americana*”, “*Verticillium* *alboatrum*”, “*Verticillium alboatrum* strains”. Additionally, USDA fungal database was also searched for the species and isolates.

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App 1.15 *Diplodia pseudoseriata*

Diplodia pseudoseriata is a fungus with a wide host and environmental range. It can cause disease on many important host plants but can also have an endophytic lifestyle. In avocado, *D. pseudoseriata* causes branch canker and dieback.

App 1.15.1 Summary of PRA conclusions

Given the arguments and evidence presented:

- *Diplodia pseudoseriata* meets the criteria to be a hazard on imported *Persea americana* budwood imported to New Zealand because it:
    - is not known to be present in New Zealand,
    - has the potential to establish in New Zealand and cause unwanted harm, and
    - has the potential to enter New Zealand on *Persea americana* budwood (as described in the commodity description).
- *Diplodia pseudoseriata* poses an overall moderate risk on *P. americana* budwood imported to New Zealand because it:
    - has a high likelihood of entering New Zealand on *P. americana* budwood (with moderate uncertainty) because:
        - *Diplodia pseudoseriata* can be associated with avocado budwood without causing visible symptoms
        - disease symptoms may not develop throughout the entire Post Entry Quarantine (PEQ) period
    - has a high likelihood of transferring from *P. americana* budwood to a suitable host in New Zealand (with low uncertainty) because:
        - imported avocado budwood is intended to generate plants for planting in New Zealand and avocado plants (which are a suitable host for these pathogens) derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments, and
        - if the commodity is infected with this pathogen at the time of their release from PEQ and they are used as propagative material then the pathogen can be transferred to a suitable host on multiple occasions.
    - has a high likelihood of establishing in New Zealand (with low uncertainty) because:
        - hosts (in the form of cultivated avocados and other cultivated crops) are available for the establishment of *D. pseudoseriata*
        - suitable climate is available for the establishment of *D. pseudoseriata*
    - may cause moderate overall impact on New Zealand (with moderate uncertainty) because:
        - *Diplodia pseudoseriata* can affect a broad range of host plants that are of economic importance in Aotearoa New Zealand
        - direct impacts of *D. pseudoseriata* are difficult to assess as the pathogen usually infects plants alongside other pathogens

The following table contains a summary of risk assessment against the criteria:

Criteria	**Rating**	**Uncertainty**
The likelihood of entry on the commodity, given the commodity description	High	Moderate
The likelihood of transferring from the imported commodity onto a suitable host	High	Low
The likelihood of establishing in the New Zealand environment	High	Low
Impact on the New Zealand economy, environment, human health and society	Moderate	Moderate
Overall level of assessed risk to New Zealand	Moderate	Moderate

App 1.15.2 Taxonomic description

Scientific name: *Diplodia pseudoseriata* C.A. Pérez et al. (2010)

Order/Family: Botryosphaeriales/Botryosphaeriaceae

Other names include: *Diplodia alatafructa* Mehl & Slippers, *Diplodia insularis* Linaldeddu, A. Alves & A.J.L. Phillips, *Diplodia pseudoplatani* Wijayaw., A.J.L. Phillips, D.J. Bhat & K.D. Hyde

Taxonomic notes: Names above were synonymised with the current name by Zhang et al. (2021).

App 1.15.3 Hazard identification

Diplodia pseudoseriata is not known to be present in New Zealand as it is not listed in NZOR (2021) or PPIN (2021). The regulatory status of *D. pseudoseriata* has not been assessed (ONZPR 2021). Biota NZ (2022) lists the *D. pseudoseriata* complex as present in New Zealand but it is not known which species within this complex is present.

Diplodia pseudoseriata has the potential to establish (and spread) in New Zealand because:

- *Diplodia pseudoseriata* has a wide host range and known host plants including *P. americana* (avocado) (Valencia et al. 2019) are widely grown in New Zealand, both in home gardens and commercial production.

Diplodia pseudoseriata has the potential to cause harm to New Zealand because:

- *Diplodia pseudoseriata* causes branch canker and dieback in avocado (Valencia et al. 2019) and other host plants such as *Acacia* spp., *Citrus* spp. and *Eucalyptus* sp. (Table 7-2).
- Avocado fruit are an important commercial crop in New Zealand. In the 2019–2020 financial year, export sales of fresh avocados earned NZ$112.3 million and domestic sales earned NZ$50.6 million (Plant & Food Research 2020).

Diplodia pseudoseriata is associated with *P. americana* budwood because:

- *Persea americana* is a known host of *D. pseudoseriata* (Valencia et al. 2019);
- *Diplodia pseudoseriata* occurs in association with stems endophytically or pathogenically causing branch canker (Valencia et al. 2019).

Given the arguments and evidence above, *D. pseudoseriata* meets the criteria to be a hazard on *P. americana* budwood (as in the commodity description) imported to New Zealand.

App 1.15.4 Risk assessment

Biology and epidemiology of Diplodia pseudoseriata

Host range and distribution

Diplodia pseudoseriata was first described in Uruguay on myrtaceous hosts species native to Uruguay (Pérez et al. 2010). It was later isolated from several hosts in 7 families (Table 7-2).

Table 7-2: Host range and distribution of *Diplodia pseudoseriata*

Host	Host family	Country	CMI	Reference
Acacia karroo	Fabaceae	South Africa	0.7–0.8	Jami et al. (2014)
Acca sellowiana	Myrtaceae	Uruguay	0.8	Pérez et al. (2010)
Blepharocalyx salicifolius	Myrtaceae	Uruguay	0.8	Pérez et al. (2010)

Host	Host family	Country	CMI	Reference
Citrus limon	Rutaceae	Spain	0.7–0.9	Bezerra et al. (2021)
Citrus reticulata	Rutaceae	Spain	0.7–0.9	Bezerra et al. (2021)
Citrus sinensis	Rutaceae	Portugal	0.8–0.9	Bezerra et al. (2021)
Eriobotrya japonica	Rosaceae	Spain	0.8	González-Domínguez et al. (2017)
Eugenia involucrata	Myrtaceae	Uruguay	0.8	Pérez et al. (2010)
Eugenia uniflora	Myrtaceae	Uruguay	0.8	Pérez et al. (2010)
Fraxinus angustifolia	Oleaceae	Italy	0.8–0.9	Bezerra et al. (2021)
Hexachlamis edulis	Myrtaceae	Uruguay	0.8	Pérez et al. (2010)
Myrceugenia euosma	Myrtaceae	Uruguay	0.8	Pérez et al. (2010)
Myrcianthes cisplatensis	Myrtaceae	Uruguay	0.8	Pérez et al. (2010)
Myrciaria tenella	Myrtaceae	Uruguay	0.8	Pérez et al. (2010)
Persea americana	Lauraceae	Chile	0.8	Valencia et al. (2019)
Pistacia lenticus	Anacardiaceae	Italy	0.8	Linaldeddu et al. (2016)
Pterocarpus angolensis	Fabaceae	South Africa	0.8	Mehl et al. (2011)
Retama raetam	Fabaceae	Tunisia	0.7–0.8	Hlaiem et al. (2019)
Acacia sp.	Fabaceae	Australia	0.7	Tan et al. (2019)
Eucalyptus globulus	Myrtaceae	Portugal	0.8–0.9	Batista et al. (2020)

Life cycle

Available literature on *D. pseudoseriata* does not specifically focus on life cycle or transmission mode. However, *D. pseudoseriata* is usually isolated alongside other fungi from the Botryosphaeriaceae family and therefore it also likely shares similar life cycle and transmission modes with other fungi in the Botryosphaeriaceae family (Pérez et al. 2010; Linaldeddu et al. 2016; Valencia et al. 2019; Batista et al. 2020; Bezerra et al. 2021).

Healthy plants are usually infected through wounds or natural openings such as stomata (Slippers and Wingfield 2007). Some species of fungi in the Botryosphaeriaceae family can be seed borne or infect seeds, but most infections of woody hosts are through horizontal transmission via spores (Slippers and Wingfield 2007). *Diplodia pseudoseriata* produces conidia (asexual spores) in spherical fruiting bodies (pycnidia) and the sexual form (teleomorph) of the fungus has not yet been observed (Zhang et al. 2021). Similar to other Botryosphaeriaceae fungi, the spores are likely transmitted naturally a short distance by water splashes or rain, but could be transmitted longer distances by anthropogenic means such as grafting or tools (Slippers and Wingfield 2007).

Optimal growth conditions

Optimal temperature for mycelial growth of *D. pseudoseriata* isolates under laboratory conditions was 25℃ with a range of 5–30℃ (Valencia et al. 2019).

Symptoms and asymptomatic infections

Diplodia pseudoseriata can cause branch canker and dieback, necrosis of stems but can also infect host plants without developing any symptoms.

Under field conditions *D. pseudoseriata* was associated with branch canker, dieback and associated wilting of leaves and inflorescence and impeded fruit growth in avocado (Valencia et al. 2019). Brown exudate and necrosis developed in the vascular system 3 weeks after artificial inoculation of 1-year-old avocado plants (Valencia et al. 2019). In Chile, where the disease was first described on avocado, the disease symptoms were likely triggered by a severe drought that affected avocado production (Valencia et al. 2019).

Diplodia pseudoseriata was also isolated from healthy tissues of several hosts in the Myrtaceae family when it was first described (Pérez et al. 2010), indicating its ability to cause asymptomatic infection.

App 1.15.5 Likelihood of entry

This assessment is made on the basis that *D. pseudoseriata* is present in the countries where avocado budwood is being imported from. *Diplodia pseudoseriata* has been reported from several countries (Table 7-2) but the only record of association with *Persea americana* is from Chile (Valencia et al. 2019).

Diplodia pseudoseriata can cause branch canker in avocado, but has also been isolated from healthy (asymptomatic) tissues of several hosts plants (Pérez et al. 2010). It is likely that the pathogen could be associated with avocado budwood without causing visible symptoms.

The routine surface disinfection of budwood entering PEQ, (dipping in 1% sodium hypochlorite for 2 minutes), will not reduce the risk of entry since most studies with *D. pseudoseriata* isolated the pathogen from surface sterilised tissues (Pérez et al. 2010).

Similar to other species in the Botryosphaeriaceae family, *D. pseudoseriata* infection is more likely to become symptomatic when the host plant is stressed (Slippers and Wingfield 2007). In Chile, where the disease was first described on avocado, the rise in disease incidence coincided with a severe drought that affected avocado production (Valencia et al. 2019). However, the appropriate environmental conditions for symptom development may be absent throughout the entire PEQ period and the infection could remain undetected.

Given the arguments and evidence above, that is:

- *Diplodia pseudoseriata* can be associated with avocado budwood without causing visible symptoms
- disease symptoms may not develop throughout the entire PEQ period

the likelihood of *D. pseudoseriata* entering New Zealand associated with *P. americana* budwood is considered to be **high**, with **moderate** uncertainty.

App 1.15.6 Likelihood of exposure

This assessment is made on the basis that *D. pseudoseriata* has entered New Zealand undetected.

When a pest or pathogen arrives in a new area, it usually needs to find or encounter a growing host plant in order to establish. The commodity (imported avocado budwood, as described, see section 2.1) is intended to generate plants for planting in New Zealand. Avocado plants derived from avocado budwood will be planted and nurtured to survive in optimal or near optimal conditions in controlled or natural environments in New Zealand. Avocado (*P. americana*) is a suitable host for *D. pseudoseriata*.

If avocado plants are infected with *D. pseudoseriata* at the time of their release from PEQ they will be planted in New Zealand or used to propagate new plants for planting (see section 2.3 for additional information about exposure).

Given the arguments and evidence above, the likelihood of exposure of *D. pseudoseriata* in New Zealand from *P. americana* budwood is considered high, with low uncertainty.

App 1.15.7 Likelihood of establishment

This assessment is made on the basis that *D. pseudoseriata* has been successfully exposed to a suitable host plant in the New Zealand environment.

Diplodia pseudoseriata has a broad host range (Table 7-2) and many of its hosts are either grown commercially in New Zealand (e.g., *Acacia* spp., *Citrus* spp., *Eucalyptus* sp.) or as backyard plants in many home gardens. Therefore, hosts for the establishment of *D. pseudoseriata* are readily available.

Diplodia pseudoseriata is also found across a broad geographic range from countries (Table 7-2) that have a similar climate to the whole of New Zealand, as well as to the Auckland and Northland regions, as indicated by a climate match index of ≥0.7 (Phillips et al. 2018).

Given the arguments and evidence above, that is:

- hosts (in the form of cultivated avocados and other cultivated crops) are available for the establishment of *D. pseudoseriata*
- suitable climate is available for the establishment of *D. pseudoseriata* (Phillips et al. 2018)

the likelihood of *D. pseudoseriata* establishing in New Zealand is considered high, with low uncertainty.

App 1.15.8 Impacts in New Zealand

Diplodia pseudoseriata can be asociated with a wide range of plants including avocado and other economically important hosts such as citruses, *Eucalyptus* and *Acacia* (Table 7-2). The fungus can cause branch canker and dieback, but can also infect the plants without developing any symptoms. Diplodia pseudoseriata likely spreads naturally by water splashes or rain over short distances, but could be transmitted over longer distances by anthropogenic means such as grafting or tools (Slippers and Wingfield 2007). As such, symptomless cuttings of avocado plants infected with *Diplodia pseudoseriata* can be transmitted and spread through grafting especially in commercial avocado plant nurseries.

Economic impacts

Given the broad host range of the *D. pseudoseriata*, it is likely that avocados would not be the only commercially produced hosts affected by this pathogen (Table 7-2). *Eucalyptus* and *Acacia* species are known hosts of *D. pseudoseriata* and are grown on more than 1.2% area of commercial forests in Aotearoa New Zealand (Forest Owners Association 2020). Lemons, oranges and mandarins are known hosts of *D. pseudoseriata* with a combined value of $55.8 million on the domestic market and $9 million in exports (Plant & Food Research 2020).

Outbreaks of *Diplodia pseudoseriata* could cause stem canker and yield losses of avocado (Valencia et al. 2019). However, assessments of direct impacts of *D. pseudoseriata* are unavailable as the pathogen usually infects host plants alongside other fungi from the family Botryosphaeriaceae (Pérez et al. 2010; Linaldeddu et al. 2016; Valencia et al. 2019; Batista et al. 2020; Bezerra et al. 2021).

Given the arguments and evidence above, that is:

- the broad host range *D. pseudoseriata* includes many plants that are of economic importance in Aotearoa New Zealand (Table 7-2)
- *Diplodia pseudoseriata* may cause disease
- direct impacts of *D. pseudoseriata* are difficult to assess as the pathogen usually infects plants alongside other pathogens

the economic impact of *D. pseudoseriata* to New Zealand is considered to be moderate, with moderate uncertainty.

Environmental impacts

Diplodia pseudoseriata is not known to infect plants native to Aotearoa New Zealand, but given its broad host range, it is likely some native plants, especially in the Myrtaceae family might be susceptible to the pathogen (Table 7-2). Furthermore, fungi from the Botryosphaeriaceae family including several species of *Diplodia* have already established in Aotearoa, New Zealand (Biota NZ 2022) with low impacts on the environment to date, suggesting other members of this genus may do the same.

Given the arguments and evidence above, the impact on the environment from the establishment of *D. pseudoseriata* in New Zealand is considered to be low, with high uncertainty.

Human health impacts

There are no known human health impacts associated with *D. pseudoseriata.*

Given the arguments and evidence above, the human health impact of *D. pseudoseriata* to New Zealand is considered to be negligible, with low uncertainty.

Sociocultural impacts

Plant disease associated with *D. pseudoseriata* could negatively affect fruit trees (e.g., citrus and avocado) growing on residential properties, as well as amenity plants (e.g., *Acacia* spp. and *Eucalyptus* spp.). The extent of these impacts, however, is unknown.

Given the arguments and evidence above, the sociocultural impact of *D. pseudoseriata* to New Zealand is considered to be moderate, with moderate uncertainty.

Overall impact to New Zealand

The overall impact on the New Zealand economy, environment, human health and society is considered to be moderate, with moderate uncertainty.

App 1.15.9 Overall level of assessed risk to New Zealand

Based on the assessments of likelihood and consequence above, that:

- the likelihood of entry is high
- assuming entry, the likelihood of exposure is high
- assuming successful exposure, the likelihood of establishment is high
- the overall impact on the New Zealand economy, environment, human health and society is considered to be moderate

the overall level of assessed risk to New Zealand from *D. pseudoseriata* on imported *P. americana* budwood (as in the commodity description) is considered to be moderate, with moderate uncertainty.

App 1.15.10 Management considerations

Diplodia pseudoseriata can cause branch canker and dieback, necroses of stems but can also infect host plants without developing any symptoms. Similar to other species in the Botryosphaeriaceae family, *D. pseudoseriata* infection is more likely to become symptomatic when the host plant is stressed (Slippers and Wingfield 2007). In Chile, where the disease was first described on avocado, the rise in disease incidence coincided with a severe drought that affected avocado production (Valencia et al. 2019). *Diplodia pseudoseriata* was isolated from branches and inner woody tissues of avocado, therefore testing stem or woody tissues would increase the chances of detecting the pest. Phylogenettic analysis of Internal transcribed spacer region (ITS1-5.8S-ITS2) and the translation elongation factor 1-α (TEF1- α) should be specific enough to identify *D. pseudoseriata* in PCR testing (Valencia et al. 2019).

App 1.15.11 Answers to Risk Management Questions

What factors influence the likelihood of the pest or disease being detected during the growing period (a period of active growth with a minimum of three distinct phenological growing events referred to as 'shoot flush') in 3A PEQ?

- ***What symptoms will the pest or disease exhibit?***

Diplodia pseudoseriata can cause branch canker and dieback, necrosis of stems but can also infect host plants without developing any symptoms.

- ***What are the known environmental conditions conducive to symptom expression?***

Similar to other species in the Botryosphaeriaceae family, *D. pseudoseriata* infection is more likely to become symptomatic when the host plant is stressed (Slippers and Wingfield 2007). In Chile, where the disease was first described on avocado, the rise in disease incidence coincided with a severe drought that affected avocado production (Valencia et al. 2019).

What are the limitations to taking samples for potential testing?

- ***What plant part should be tested in order to maximise the chances of detection of the pest in PEQ in the case of asymptomatic infections?***

Diplodia pseudoseriata was isolated from branches and inner woody tissues of avocado, therefore testing stem or woody tissues would increase the chances of detecting the pest.

- ***What is the optimum season for conducting detection testing?***

Similar to other species in the Botryosphaeriaceae family, *D. pseudoseriata* infection is more likely to become symptomatic when the host plant is stressed.

App 1.15.12 References

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Appendix 2. Summary of taxa excluded at hazard identification

Groups and species identified at the hazard idendification stage as not requiring further assessment and not requiring measures over and above basic measures, and the rationale for exclusion (a total of 382 organisms). DUA are diseases of unknown aetiology. No subspecific taxa highlights that there are no subspecific taxa that increase the risk to New Zealand (e.g. either no subspecific taxa or subspecific taxa is present in New Zealand).

Scientific name	Organism	Presence in New Zealand	Rationale for exclusion	Reference
Cephaleuros virescens	Algae	Present	No subspecific taxa	
Agrobacterium radiobacter rhizogenic strain	Bacteria	Present	No subspecific taxa	
Agrobacterium radiobacter tumorigenic strain	Bacteria	Present	No subspecific taxa	
Bacillus licheniformis	Bacteria	Present	No subspecific taxa	
Bacillus subtilis	Bacteria	Present	No subspecific taxa	
Erwinia aroideae	Bacteria	Present	Synonymy	*Pectobacterium aroidearum*
Erwinia atroseptica	Bacteria	Present	Synonymy	*Pectobacterium atrosepticum*
Pantoea agglomerans	Bacteria	Present	No subspecific taxa	
Pectobacterium aroidearum	Bacteria	Present	No subspecific taxa	
Pectobacterium carotovorum	Bacteria	Present	No subspecific taxa	
Pectobacterium carotovorum carotovorum	Bacteria	Present	No subspecific taxa	
Pseudomonas citriputealis	Bacteria	Present	Synonymy	*Pseudomonas syringae* pv. *syringae*
Pseudomonas syringae bacterial canker-causing strains	Bacteria	Present	No subspecific taxa	
Rhizobium rhizogenes	Bacteria	Present	No subspecific taxa	
Peduncle ringing	DUA	Present	Not applicable	
Acremonium	Fungus	Present (genus)	Insufficient information for host association	Literature search
Acrodontium crateriforme	Fungus	Present	No subspecific taxa	Korsten et al. (1988)
Acrosporium	Fungus	Absent	Not associated with the host	Cline and Farr (2006)
Acrostalagmus luteoalbus	Fungus	Present	No subspecific taxa	
Akanthomyces lecanii	Fungus	Present	No subspecific taxa	

Scientific name	**Organism**	**Presence in New Zealand**	**Rationale for exclusion**	**Reference**
Albonectria rigidiuscula	Fungus	Present	No subspecific taxa	
Alternaria alternata	Fungus	Present	No subspecific taxa	
Alternaria citri	Fungus	Present	No subspecific taxa	
Alternaria mouchaccae	Fungus	Uncertain	Insufficient information for host association	Literature search
Alternaria tenuissima	Fungus	Present	Synonymy	*Alternaria alternata*
Armillaria limonea	Fungus	Present	No subspecific taxa	
Armillaria lutea	Fungus	Absent	Not associated with commodity	Kurbetli et al. (2020)
Armillaria mellea	Fungus	Absent	Not associated with commodity	Luna and Fucikovsky (1987)
Armillaria novae-zelandiae	Fungus	Present	No subspecific taxa	
Arthrinium phaeospermum	Fungus	Present	No subspecific taxa	
Ascochyta citri	Fungus	Absent	Not associated with the host	Literature search
Aspergillus candidus	Fungus	Present	No subspecific taxa	
Aspergillus niger	Fungus	Present	No subspecific taxa	
Asteromella gratissima	Fungus	Absent	Insufficient information for commodity association	Petrak and Ciferri (1930)
Athelia rolfsii	Fungus	Present	No subspecific taxa	
Aureobasidium pullulans	Fungus	Present	No subspecific taxa	
Barriopsis fusca	Fungus	Absent	Insufficient evidence for host association	
Bipolaris sorokiniana	Fungus	Present	No subspecific taxa	
Botryodiplodia	Fungus	Present (genus)	Synonymy	Species associated with *Persea* is a synonym of *Lasiodiplodia theobromae*; Darvas and Kotze (1987)
Botryosphaeria australis	Fungus	Present	No subspecific taxa	
Botryosphaeria dothidea	Fungus	Present	No subspecific taxa	
Botryosphaeria quercuum	Fungus	Absent	Not associated with the host	Literature search
Botryosphaeria stevensii	Fungus	Present	No subspecific taxa	
Botryotinia fuckeliana	Fungus	Present	No subspecific taxa	

Scientific name	**Organism**	**Presence in New Zealand**	**Rationale for exclusion**	**Reference**
Botrytis cinerea	Fungus	Present	No subspecific taxa	
Cadophora luteo-olivacea	Fungus	Present	No subspecific taxa	
Calonectria cylindrospora	Fungus	Present	No subspecific taxa	
Calonectria ilicicola	Fungus	Absent	Not associated with commodity	Dann et al. (2012)
Calonectria insularis	Fungus	Absent	Insufficient information for commodity association	Schoch et al. (1999)
Calonectria kyotensis	Fungus	Present	No subspecific taxa	
Calonectria morganii	Fungus	Present	No subspecific taxa	
Calonectria pauciramosa	Fungus	Present	No subspecific taxa	
Calonectria scoparia	Fungus	Present	No subspecific taxa	
Capnodium citri	Fungus	Present	No subspecific taxa	
Cephalothecium sp.	Fungus	Present	No subspecific taxa	
Ceratocystis fimbriata	Fungus	Present	No subspecific taxa	
Cercospora	Fungus	Present (genus)	Synonymy	*Pseudocercospora*
Ceriporia purpurea	Fungus	Absent	Not associated with the host	Gilbertson et al. (2002)
Chaetomium spirale	Fungus	Present	No subspecific taxa	
Chrysoporthe cubensis	Fungus	Absent	Not associated with commodity	Seixas et al. (2004)
Cladosporium citri	Fungus	Absent	Misidentification	Stevens and Piper (1941)
Cladosporium cladosporioides	Fungus	Present	No subspecific taxa	
Cladosporium herbarum	Fungus	Present	No subspecific taxa	
Clasterosporium roupalae	Fungus	Absent	Not associated with the host	Literature search
Clonostachys rosea	Fungus	Present	No subspecific taxa	
Cochliobolus intermedius	Fungus	Present	No subspecific taxa	
Cochliobolus setariae	Fungus	Present	No subspecific taxa	
Colletotrichum acutatum	Fungus	Present	No subspecific taxa	
Colletotrichum alienum	Fungus	Present	No subspecific taxa	
Colletotrichum aotearoa	Fungus	Present	No subspecific taxa	
Colletotrichum boninense	Fungus	Present	No subspecific taxa	

Scientific name	Organism	Presence in New Zealand	Rationale for exclusion	Reference
Colletotrichum cigarro	Fungus	Present	No subspecific taxa	
Colletotrichum crassipes	Fungus	Present	No subspecific taxa	
Colletotrichum dematium	Fungus	Present	No subspecific taxa	
Colletotrichum fioriniae	Fungus	Present	No subspecific taxa	
Colletotrichum fructicola	Fungus	Border intercept	Not associated with commodity	Sharma et al. (2017); Hofer et al. (2021)
Colletotrichum gigasporum	Fungus	Present	No subspecific taxa	
Colletotrichum gloeosporioides	Fungus	Present	No subspecific taxa	
Colletotrichum godetiae	Fungus	Present	No subspecific taxa	
Colletotrichum kahawae	Fungus	Present	No subspecific taxa	
Colletotrichum karsti	Fungus	Present	No subspecific taxa	
Colletotrichum nupharicola	Fungus	Absent	Misidentification	Johnson et al. (1997); Khodadadi et al. (2020)
Colletotrichum perseae	Fungus	Present	No subspecific taxa	
Colletotrichum siamense	Fungus	Present	No subspecific taxa	
Colletotrichum simmondsii	Fungus	Present	No subspecific taxa	
Colletotrichum theobromicola	Fungus	Present	No subspecific taxa	
Coprinus	Fungus	Present (genus)	Not associated with the host	Baker (1938)
Corallomycetella repens	Fungus	Present	No subspecific taxa	
Coriolopsis occidentalis	Fungus	Absent	Not associated with the host	Literature search
Cornuvesica falcata	Fungus	Present	No subspecific taxa	
Corynespora cassiicola	Fungus	Present	No subspecific taxa	
Cryptococcus gattii	Fungus	Present	No subspecific taxa	
Cryptococcus neoformans	Fungus	Present	No subspecific taxa	
Curvularia lunata	Fungus	Present	No subspecific taxa	
Curvularia senegalensis	Fungus	Absent	Not associated with commodity	Korsten et al. (1988)
Cylindrocarpon didymum	Fungus	Present	No subspecific taxa	
Cylindrocladiella parva	Fungus	Present	No subspecific taxa	
Cylindrocladiella pseudoinfestans	Fungus	Absent	Not associated with commodity	Parkinson et al. (2017b)

Scientific name	**Organism**	**Presence in New Zealand**	**Rationale for exclusion**	**Reference**
Cylindrocladiella stellenboschensis	Fungus	Present	No subspecific taxa	
Cylindrocladium	Fungus	Absent	Not associated with commodity	Raabe et al. (1981)
Cytospora actinidiae	Fungus	Absent	Not associated with the host	Literature search
Dactylonectria anthuriicola	Fungus	Absent	Not associated with commodity	Parkinson et al. (2017b)
Dactylonectria macrodidyma	Fungus	Present	No subspecific taxa	
Dactylonectria novozelandica	Fungus	Present	No subspecific taxa	
Dactylonectria pauciseptata	Fungus	Present	No subspecific taxa	
Desarmillaria tabescens	Fungus	Absent	Not associated with commodity	Horst (2013)
Dialonectria episphaeria	Fungus	Present	No subspecific taxa	
Diaporthe ampelina	Fungus	Present	No subspecific taxa	
Diaporthe australafricana	Fungus	Present	No subspecific taxa	
Diaporthe citri	Fungus	Present	No subspecific taxa	
Diaporthe foeniculacea	Fungus	Absent	Synonymy	*Guignardia foeniculacea*
Diaporthe rudis	Fungus	Present	No subspecific taxa	
Didymostilbe cubensis	Fungus	Absent	Not associated with commodity	Arnold and Ruiz (1984)
Diplodia mutila	Fungus	Present	No subspecific taxa	
Diplodia perseana	Fungus	Absent	Insufficient information for host association	Kobayashi (2007)
Diplodia pseudoseriata	Fungus	Present	No subspecific taxa	
Diplodia seriata	Fungus	Present	No subspecific taxa	
Dipodascus geotrichum	Fungus	Present	No subspecific taxa	
Dothichiza	Fungus	Present (genus)	Insufficient information for host association	Alvarez (1976)
Dothiorella iberica	Fungus	Present	No subspecific taxa	
Elsinoe pyri	Fungus	Present	No subspecific taxa	
Epicoccum nigrum	Fungus	Present	No strains specific to host	Fávaro et al. (2011)
Erysiphe betae	Fungus	Present	No subspecific taxa	
Erythricium salmonicolor	Fungus	Present	No subspecific taxa	
Exserohilum rostratum	Fungus	Present	No subspecific taxa	

Scientific name	**Organism**	**Presence in New Zealand**	**Rationale for exclusion**	**Reference**
Fomitella supina	Fungus	Absent	Not associated with commodity	The Global Fungal Red List Initiative (2021)
Fracchiaea heterogenea	Fungus	Absent	Not associated with commodity	Raymundo et al. (2017)
Fusarium acuminatum	Fungus	Present	Not applicable	
Fusarium avenaceum	Fungus	Present	Not applicable	
Fusarium compactum	Fungus	Recorded in error	Not associated with the host	Frisullo et al. (1994)
Fusarium crookwellense	Fungus	Present	Synonymy	*Fusarium cerealis*
Fusarium equiseti	Fungus	Present	Not associated with commodity	Biota NZ (2022)
Fusarium expansum	Fungus	Absent	Not associated with commodity	Baker (1938)
Fusarium fujikuroi	Fungus	Present	No subspecific taxa	
Fusarium gibbosum	Fungus	Present	Synonymy	*Fusarium equiseti*
Fusarium graminearum	Fungus	Present	No subspecific taxa	
Fusarium incarnatum	Fungus	Present	No subspecific taxa	
Fusarium lateritium	Fungus	Present	No subspecific taxa	
Fusarium oxysporum	Fungus	Present	No subspecific taxa	
Fusarium pallidoroseum	Fungus	Present	Synonymy	*Fusarium incarnatum*
Fusarium roseum	Fungus	Present	No subspecific taxa	
Fusarium scirpi	Fungus	Absent	Insufficient information for host association	Korsten et al. (1988)
Fusicoccum aesculi	Fungus	Present	Synonymy	*Botryosphaeria dothidea*
Ganoderma lucidum	Fungus	Absent	Not associated with commodity	Loyd et al. (2018)
Ganoderma zonatum	Fungus	Absent	Not associated with commodity	CABI (2021)
Ganoderma zonatum sulcatum	Fungus	Not applicable	Misidentification	No record of this name
Gibberella	Fungus	Present (genus)	Synonymy	*Fusarium*
Gliocladiopsis curvata	Fungus	Present	No subspecific taxa	
Gliocladiopsis sumatrensis	Fungus	Absent	Not associated with commodity	Lombard and Crous (2012)
Gliocladiopsis tenuis	Fungus	Present	No subspecific taxa	
Gliocladiopsis whileyi	Fungus	Absent	Not associated with commodity	Parkinson et al. (2017a)
Gloeosporium	Fungus	Present (genus)	Taxonomy	Some species are considered *Colletotrichum* which are assessed

Scientific name	Organism	Presence in New Zealand	Rationale for exclusion	Reference
Glomerella	Fungus	Present (genus)	Taxonomy	Some species are considered *Colletotrichum* which are assessed
Gnomonia	Fungus	Present (genus)	Not associated with the host	EPPO (2020)
Gracilistilbella clavulata	Fungus	Present	No subspecific taxa	
Helminthosporium	Fungus	Present (genus)	Taxonomy	Genus has been split into multiple genera
Hendersonia	Fungus	Present (genus)	Taxonomy	Genus has been split into multiple genera
Hexagonia rigida	Fungus	Absent	Not associated with commodity	Lee et al. (2012)
Humicola	Fungus	Present (genus)	Not associated with the host	Rashmi et al. (2019)
Ilyonectria destructans	Fungus	Present	No subspecific taxa	
Ilyonectria liriodendri	Fungus	Present	No subspecific taxa	
Laetiporus sulphureus	Fungus	Absent	Not associated with commodity	Horst (2013)
Lasiodiplodia theobromae	Fungus	Present	No subspecific taxa	
Lentinus stuppeus	Fungus	Absent	Insufficient information for host association	Literature search
Leptosphaeria gratissitnae	Fungus	Absent	Insufficient information for commodity association	Rieuf and Yeasca (1970)
Leptosphaerulina trifolii	Fungus	Present	No subspecific taxa	
Macrophoma perseae	Fungus	Absent	Insufficient information for commodity association	Index Fungorum (2022)
Macrophomina phaseolina	Fungus	Present	No subspecific taxa	
Macrosporium	Fungus	Present (genus)	Synonymy	*Alternaria*
Marasmiellus scandens	Fungus	Absent	Insufficient information for host association	Peregrine and Ahmad (1982)
Marssonina	Fungus	Present (genus)	Taxonomy	Genus has been split into multiple genera
Microporus affinis	Fungus	Absent	Insufficient information for commodity association	Gilbertson et al. (2002)
Monilia	Fungus	Present (genus)	Taxonomy	Genus has been split into multiple genera
Monochaetia	Fungus	Present (genus)	Taxonomy	Genus has been split into multiple genera
Mucor	Fungus	Present (genus)	Taxonomy	Genus has been split into multiple genera

Scientific name	**Organism**	**Presence in New Zealand**	**Rationale for exclusion**	**Reference**
Mycoacia kurilensis	Fungus	Absent	Not associated with the host	Index Fungorum (2022)
Mycosphaerella tassiana	Fungus	Present	Synonymy	*Cladosporium herbarum*
Nectria cinnabarina	Fungus	Present	No subspecific taxa	
Nectria pseudotrichia	Fungus	Present	No subspecific taxa	
Nectricladiella infestans	Fungus	Present	No subspecific taxa	
Neocosmospora ramosa	Fungus	Absent	Not associated with commodity	Sandoval-Denis and Crous (2018)
Neocosmospora solani	Fungus	Present	No subspecific taxa	
Neofusicoccum australe	Fungus	Present	No subspecific taxa	
Neofusicoccum cryptoaustrale	Fungus	Present	No subspecific taxa	
Neofusicoccum luteum	Fungus	Present	No subspecific taxa	
Neofusicoccum mangiferae	Fungus	Present	No subspecific taxa	
Neofusicoccum mediterraneum	Fungus	Absent	Not associated with the host	CABI (2021)
Neofusicoccum parvum	Fungus	Present	No subspecific taxa	
Neofusicoccum ribis	Fungus	Present	No subspecific taxa	
Neofusicoccum stellenboschiana	Fungus	Present	No subspecific taxa	
Neofusicoccum vitifusiforme	Fungus	Absent	Not associated with the host	Phillips et al. (2013)
Neofusioccum luteum	Fungus	Not applicable	Misidentification	No record in Mycobank (Robert et al. 2005) or Index Fungorum (2022)
Neonectria ditissima	Fungus	Present	No subspecific taxa	
Neopestalotiopsis clavispora	Fungus	Present	No subspecific taxa	
Neoscytalidium dimidiatum	Fungus	Present	No subspecific taxa	
Nigrospora oryzae	Fungus	Present	No subspecific taxa	
Nothoramichloridium perseae	Fungus	Absent	Insufficient information for commodity association	Crous et al. (2019)
Ovularia	Fungus	Present (genus)	Taxonomy	Genus has been split into multiple genera
Oxychaete cervinogilva	Fungus	Absent	Insufficient information for host association	Literature search
Oxyporus latemarginatus	Fungus	Absent	Not associated with commodity	Ploetz (2003)

Scientific name	Organism	Presence in New Zealand	Rationale for exclusion	Reference
Parencoelia myriostylidis	Fungus	Absent	Insufficient information for host association	Literature search
Penicillium chrysogenum	Fungus	Present	No subspecific taxa	
Penicillium digitatum	Fungus	Present	No subspecific taxa	
Penicillium expansum	Fungus	Present	No subspecific taxa	
Penicillium italicum	Fungus	Present	No subspecific taxa	
Penicillium simplicissimum	Fungus	Present	No subspecific taxa	
Periconia byssoides	Fungus	Present	No subspecific taxa	
Periconia cambrensis	Fungus	Present	No subspecific taxa	
Pestalotia eriobotryae-japonicae	Fungus	Absent	Insufficient information for host association	Literature search
Pestalotiopsis adusta	Fungus	Present	No subspecific taxa	
Pestalotiopsis aloës	Fungus	Absent	Insufficient information for host association	Literature search
Pestalotiopsis disseminata	Fungus	Present	No subspecific taxa	
Pestalotiopsis gracilis	Fungus	Absent	Not associated with the host	Granados-Montero et al. (2018)
Pestalotiopsis guepinii	Fungus	Present	No subspecific taxa	
Pestalotiopsis leprogena	Fungus	Absent	Insufficient information for host association	Literature search
Pestalotiopsis mangiferae	Fungus	Absent	Not associated with the host	Kamhawy et al. (2011)
Pestalotiopsis phoenicis	Fungus	Absent	Insufficient information for host association	Literature search
Pestalotiopsis versicolor	Fungus	Present	No subspecific taxa	
Peyronellaea obtusa	Fungus	Absent	Insufficient information for host association	Literature search
Phanerochaete australis	Fungus	Present	No subspecific taxa	
Phellinus gilvus	Fungus	Present	No subspecific taxa	
Phellinus grenadensis	Fungus	Absent	Insufficient information for host association	Literature search
Phlebia acanthocystis	Fungus	Present	No subspecific taxa	
Phoma persicae	Fungus	Present	No subspecific taxa	

Scientific name	**Organism**	**Presence in New Zealand**	**Rationale for exclusion**	**Reference**
Phomopsis achilleae	Fungus	Absent	Insufficient information for host association	Literature search
Phomopsis phaseoli	Fungus	Present	No subspecific taxa	
Phyllosticta capitalensis	Fungus	Present	No subspecific taxa	
Phyllosticta citricarpa	Fungus	Recorded in error	Insufficient information for host association	Meyer et al. (2001)
Phyllosticta micropuncta	Fungus	Absent	Synonymy	*Phyllosticta perseae*
Phymatotrichopsis omnivora	Fungus	Absent	Not associated with commodity	Bragard et al. (2019)
Phymatotrichum	Fungus	Absent	Synonymy	*Phymatotrichopsis*
Pionnotes capillacea	Fungus	Absent	Insufficient information for host association	Teodoro (1937)
Pithomyces graminicola	Fungus	Absent	Not associated with commodity	Thomas et al. (1994)
Podoconis anacardii	Fungus	Absent	Not associated with commodity	Shaw (1984)
Podosphaera tridactyla	Fungus	Present	No subspecific taxa	
Polyporus sanguineus	Fungus	Present	No subspecific taxa	
Pseudopestalotiopsis theae	Fungus	Absent	Insufficient information for host association	Liu (1977)
Pseudopithomyces chartarum	Fungus	Present	No subspecific taxa	
Pseudopithomyces maydicus	Fungus	Uncertain	Not associated with commodity	Baker (1938)
Puccinia scimitriformis	Fungus	Absent	Insufficient information for host association	Literature search
Purpureocillium lilacinum	Fungus	Present	No subspecific taxa	
Pyrrhoderma lamaoense	Fungus	Absent	Not associated with commodity	Khan et al. (2020)
Rhizomorpha subcorticalis	Fungus	Absent	Synonymy	*Armillaria mellea* (anamorph of)
Rhizopus arrhizus	Fungus	Present	No subspecific taxa	
Rhizopus stolonifer	Fungus	Present	No subspecific taxa	
Rigidoporus microporus	Fungus	Uncertain	Not associated with commodity	Shaw (1984)
Rigidoporus ulmarius	Fungus	Absent	Not associated with commodity	Ploetz (2003)
Rosellinia bunodes	Fungus	Absent	Insufficient information for commodity association	Lavaire and Morazán (2013); Ploetz et al. (1994)
Rosellinia necatrix	Fungus	Present	No subspecific taxa	

Scientific name	**Organism**	**Presence in New Zealand**	**Rationale for exclusion**	**Reference**
Rosellinia pepo	Fungus	Uncertain	Insufficient information for commodity association	Lavaire and Morazán (2013); Ploetz et al. (1994)
Rugonectria rugulosa	Fungus	Absent	Insufficient information for commodity association	Samuels and Brayford (1994)
Sarocladium strictum	Fungus	Present	No subspecific taxa	
Schizophyllum commune	Fungus	Present	No subspecific taxa	
Sclerostagonospora	Fungus	Present (genus)	Insufficient information for host association	Korsten et al. (1988)
Sclerotinia sclerotiorum	Fungus	Present	Not applicable	
Septoria	Fungus	Absent	Synonymy	*Mycosphaerella*
Sesquicillium	Fungus	Present (genus)	Synonymy	*Clonostachys*
Sphaceloma purea	Fungus	Not applicable	Taxonomy	Invalid name
Sphaerostilbe cinnabarina	Fungus	Present	Synonymy	*Nectria pseudotrichia*
Stemphylium	Fungus	Present (genus)	Synonymy	
Stilbella	Fungus	Present (genus)	Synonymy	
Stomiopeltis citri	Fungus	Absent	Misidentification	Smith et al. (1985)
Strigula smaragdula	Fungus	Present	No subspecific taxa	
Trametes coccinea	Fungus	Present	No subspecific taxa	
Trametes elegens	Fungus	Absent	Insufficient information for host association	Teodoro (1937)
Trametes hirsuta	Fungus	Present	No subspecific taxa	
Trametes nivosa	Fungus	Present	No subspecific taxa	
Trametes versicolor	Fungus	Present	No subspecific taxa	
Trichocladium	Fungus	Present (genus)	Synonymy	*Chaetomium*
Trichoderma harzianum	Fungus	Present	No subspecific taxa	
Trichoderma koningii	Fungus	Present	No subspecific taxa	
Trichoderma viride	Fungus	Present	No subspecific taxa	
Trichomerium ornatum	Fungus	Absent	Not associated with commodity	Chomnunti et al. (2012)
Trichothecium roseum	Fungus	Present	No subspecific taxa	
Tubercularia lateritia	Fungus	Present	Synonymy	*Nectria pseudotrichia*

Scientific name	**Organism**	**Presence in New Zealand**	**Rationale for exclusion**	**Reference**
Venturia carpophila	Fungus	Present	No subspecific taxa	
Verticillium dahliae	Fungus	Present	No subspecific taxa	
Xenasma tulasnelloideum	Fungus	Present	No subspecific taxa	
Xenosporium berkeleyi	Fungus	Present	No subspecific taxa	
Zygosporium	Fungus	Present (genus)	Insufficient information for host association	Literature search
Aphelenchoides bicaudatus	Nematode	Present	No subspecific taxa	
Aphelenchus avenae	Nematode	Absent	Not associated with commodity	Walker (1984)
Aphelencoides sp.	Nematode	Uncertain	Insufficient information for commodity association	Williams (1980)
Boleodorus	Nematode	Absent	Not associated with commodity	Munawar et al. (2021)
Criconema demani	Nematode	Absent	Not associated with commodity	Siddiqi (2000b)
Criconema mutabile	Nematode	Present	No subspecific taxa	
Criconemoides	Nematode	Absent	Not associated with commodity	Siddiqi (2000b)
Discocriconemella perseae	Nematode	Absent	Not associated with commodity	Siddiqi (2000b)
Ditylenchus sp.	Nematode	Uncertain	Insufficient information for commodity association	Siddiqi (2000a)
Geocenamus brevidens	Nematode	Absent	Not associated with commodity	Siddiqi (2000d)
Helicotylenchus cavenessi	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Helicotylenchus dihystera	Nematode	Present	No subspecific taxa	
Helicotylenchus erythrinae	Nematode	Present	No subspecific taxa	
Helicotylenchus indicus	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Helicotylenchus microcephalus	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Helicotylenchus mucronatus	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Helicotylenchus multicinctus	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Helicotylenchus pseudorobustus	Nematode	Present	No subspecific taxa	
Hemicriconemoides cocophillus	Nematode	Absent	Not associated with commodity	Siddiqi (2000b)
Hemicriconemoides mangiferae	Nematode	Absent	Not associated with commodity	Siddiqi (2000b)

Scientific name	Organism	Presence in New Zealand	Rationale for exclusion	Reference
Hemicriconemoides strictathecatus	Nematode	Absent	Not associated with commodity	Siddiqi (2000b)
Hemicycliophora penetrans	Nematode	Absent	Not associated with commodity	Chitambar and Subbotin (2014)
Heterodera zeae	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Longidorus africanus	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Longidorus laevicapitatus	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Meloidogyne enterolobii	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Meloidogyne javanica	Nematode	Present	No subspecific taxa	
Mesocriconema denoudeni	Nematode	Absent	Not associated with commodity	Siddiqi (2000b)
Mesocriconema sosamossi	Nematode	Absent	Not associated with commodity	Siddiqi (2000b)
Nanidorus minor	Nematode	Present	No subspecific taxa	
Paratrichodorus porosus	Nematode	Present	No subspecific taxa	
Paratylenchus hamatus	Nematode	Absent	Not associated with commodity	Ghaderi (2019)
Pratylenchus brachyurus	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Pratylenchus coffeae	Nematode	Present	No subspecific taxa	
Pratylenchus goodeyi	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Pratylenchus neglectus	Nematode	Present	No subspecific taxa	
Pratylenchus penetrans	Nematode	Present	No subspecific taxa	
Pratylenchus thornei	Nematode	Present	No subspecific taxa	
Pratylenchus vulnus	Nematode	Present	No subspecific taxa	
Radopholus similis	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Rotylenchulus reniformis	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Rotylenchus breviglans	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Rotylenchus colbrani	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Rotylenchus uniformis	Nematode	Present	No subspecific taxa	
Scutellonema brachyurus	Nematode	Present	No subspecific taxa	
Scutellonema clathricaudatum	Nematode	Absent	Not associated with commodity	Siddiqi (2000c)
Trichodorus	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Tylenchorhynchus clarus	Nematode	Absent	Not associated with commodity	Siddiqi (2000d)

Scientific name	**Organism**	**Presence in New Zealand**	**Rationale for exclusion**	**Reference**
Tylenchorhynchus claytoni	Nematode	Absent	Not associated with commodity	Siddiqi (2000d)
Tylenchulus semipenetrans	Nematode	Present	No subspecific taxa	
Xiphinema americanum	Nematode	Present	No subspecific taxa	
Xiphinema brasiliense	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema brevicollum	Nematode	Present	No subspecific taxa	
Xiphinema bricolensis	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema californicum	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema diffusum	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema diversicauadatum	Nematode	Present	No subspecific taxa	
Xiphinema elongatum	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema ensiculiferum	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema inaequale	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema insigne	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema intermedium	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema pachtaicum	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema rivesi	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema tarjanense	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema turcicum	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Xiphinema vuittenezi	Nematode	Absent	Not associated with commodity	Decraemer and Geraert (2006)
Globisporangium debaryanum	Oomycete	Uncertain	Not associated with commodity	Radwan and Hassan (2018)
Globisporangium irregulare	Oomycete	Present	No subspecific taxa	
Globisporangium rostratum	Oomycete	Present	No subspecific taxa	
Globisporangium splendens	Oomycete	Present	No subspecific taxa	
Globisporangium ultimum	Oomycete	Present	No subspecific taxa	
Phytophthora boehmeriae	Oomycete	Present	No subspecific taxa	
Phytophthora cactorum	Oomycete	Present	No subspecific taxa	
Phytophthora cambivora	Oomycete	Present	No subspecific taxa	

Scientific name	Organism	Presence in New Zealand	Rationale for exclusion	Reference
Phytophthora capsici	Oomycete	Absent	Insufficient information for host association	Literature search
Phytophthora cinnamomi	Oomycete	Present	No subspecific taxa	
Phytophthora citricola	Oomycete	Present	No subspecific taxa	
Phytophthora citrophthora	Oomycete	Present	No subspecific taxa	
Phytophthora cryptogea	Oomycete	Present	No subspecific taxa	
Phytophthora kernoviae	Oomycete	Present	No subspecific taxa	
Phytophthora megakarya	Oomycete	Absent	Not associated with commodity	Bailey et al. (2016)
Phytophthora megasperma	Oomycete	Present	No subspecific taxa	
Phytophthora multivora	Oomycete	Present	No subspecific taxa	
Phytophthora nicotianae	Oomycete	Present	No subspecific taxa	
Phytophthora niederhauseri	Oomycete	Present	No subspecific taxa	
Phytophthora ramorum	Oomycete	Absent	Insufficient information for host association	Literature search
Phytopythium vexans	Oomycete	Present	No subspecific taxa	
Pythium afertile	Oomycete	Present	No subspecific taxa	
Pythium coloratum	Oomycete	Present	No subspecific taxa	
Pythium cucurbitacearum	Oomycete	Not applicable	Taxonomy	Invalid name (Robert et al. 2005; Index Fungorum 2022)
Pythium deliense	Oomycete	Absent	Not associated with commodity	CABI (2021)
Pythium oligandrum	Oomycete	Present	No subspecific taxa	
Pythium torulosum	Oomycete	Present	No subspecific taxa	
Trachysphaera fructigena	Oomycete	Absent	Insufficient information for commodity association	Dade (1940)
Tomato apical stunt viroid	Viroid	Absent	Insufficient information for host association	Literature search
Avocado 3 alphacryptovirus	Virus	Not applicable	Not applicable	Not a legitimate pathogen
Cucumber leaf spot virus	Virus	Absent	Not associated with the host	CABI (2021)
Cucumber mosaic virus	Virus	Present	No subspecific taxa	
Papaya mosaic virus	Virus	Absent	Not associated with the host	CABI (2021)

Scientific name	Organism	Presence in New Zealand	Rationale for exclusion	Reference
Persea americana alphaendornavirus 1	Virus	Present	No subspecific taxa	
Persea americana chrysovirus	Virus	Present	No subspecific taxa	
Tobacco mosaic virus	Virus	Present	No subspecific taxa	

References for Appendix 2

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Appendix 3. Hosts of *Grovesinia moricola*

Family	Species	Common name	Reference
Adoxaceae	*Viburnum recognitum*	Arrowwood	Trolinger et al. (1978)
Adoxaceae	*Viburnum* sp.	Viburnum	Trolinger et al. (1978)
Amaranthaceae	*Chenopodium ambrosioides*	Mexican tea	Trolinger et al. (1978)
Anacardiaceae	*Toxicodendron pubescens*	Poison oak	Trolinger et al. (1978)
Annonaceae	*Annona muricata*	Soursop	Bezerra et al. (2008)
Annonaceae	*Asimina triloba*	Pawpaw	Trolinger et al. (1978)
Asteraceae	*Bidens frondosa*	begger-ticks	Trolinger et al. (1978)
Asteraceae	*Eupatorium coelestinum*	Mistflower	Trolinger et al. (1978)
Asteraceae	*Eupatorium rugosum*	white snakeroot	Trolinger et al. (1978)
Asteraceae	*Solidago canadensis*	goldenrod	Trolinger et al. (1978)
Balsaminaceae	*Impatiens pallida*	pale touch-me-not	Trolinger et al. (1978)
Batulaceae	*Alnus incana*	Grey alder	Trolinger et al. (1978)
Bignoniaceae	*Campsis radicans*	trumpet vine	Trolinger et al. (1978)
Bignoniaceae	*Catalpa speciosa*	Catalpa	Trolinger et al. (1978)
Campanulaceae	*Lobelia inflata*	Indian tobacco	Trolinger et al. (1978)
Campanulaceae	*Lobelia siphilitica*	Blue cardinal flower	Trolinger et al. (1978)
Cannabaceae	*Celtis laevigata* or *Celtis occidentalis*	Hackberry	Trolinger et al. (1978)
Cannabaceae	*Humulus japonicus*	Japanese hop	Cho et al. (2013)
Commelinaceae	*Commelina diffusa*	Dayflower	Trolinger et al. (1978)
Convolvulaceae	*Ipomoea hederacea*	Morning glory	Trolinger et al. (1978)
Convolvulaceae	*Ipomoea lacunosa*	Morning glory	Trolinger et al. (1978)
Convolvulaceae	*Ipomoea quamoclit*	Cypress vine	Trolinger et al. (1978)
Cornaceae	*Cornus amomum*	red-willow	Trolinger et al. (1978)
Cornaceae	*Cornus florida*	Flowering dogwood	Trolinger et al. (1978)
Ericaceae	*Oxydendron arboreum*	Sourwood	Blake et al. (2015)
Ericaceae	*Vaccinium* sp.	Blueberry	Trolinger et al. (1978)
Euphorbiaceae	*Aleurites fordii*	tung	Trolinger et al. (1978)
Fabaceae	*Arachis hypogaea*	Peanut	Niedbalski et al. (1979), Vyas et al. (1982)
Fabaceae	*Arachis hypogaea*	peanut	Trolinger et al. (1978)
Fabaceae	*Desmodium canescens*	Tick clover	Trolinger et al. (1978)
Fabaceae	*Glycine max*	Soybean	Cho et al. (2012)
Fabaceae	*Lespedeza cyrtobotrya*	Leafy bush-clover	Park et al. (2019)
Fabaceae	*Millettia pinnata*	Indian beech	Chang (1995)
Fabaceae	*Phaseolus lunatus*	Fordhook lima bean	Niedbalski et al. (1979)
Fabaceae	*Phaseolus lunatus*	lima bean	Trolinger et al. (1978)
Fabaceae	*Phaseolus lunatus var. Harvester*	snap bean	Trolinger et al. (1978)
Fabaceae	*Vigna angularis*	Adzuki bean	Cho et al. (2017)
Fabaceae	*Vigna vexillata* var. *tsusimensis*	Wild cowpea	Lee & Kim (2007)
Hydrangeaceae	*Hydrangea petiolaris*	Hydrangea	Trolinger et al. (1978)

Family	Species	Common name	Reference
Juglandaceae	*Carya illinoiensis*	Pecan	Niedbalski et al. (1979)
Juglandaceae	*Carya illinoiensis*	pecan	Trolinger et al. (1978)
Juglandaceae	*Juglans cinerea*	Butternut	Trolinger et al. (1978)
Juglandaceae	*Juglans nigra*	Black walnut	Niedbalski et al. (1979)
Juglandaceae	*Juglans nigra*	Black walnut	Trolinger et al. (1978)
Lamiaceae	*Perilla frutescens*	perilla	Trolinger et al. (1978)
Lauraceae	*Cinnamomum kanehirae*	camphor tree	Fu & Lin (2012)
Lauraceae	*Lindera benzoin*	Spicebush	Trolinger et al. (1978)
Lauraceae	*Persea americana*	Avocado	Bezerra et al. (2008)
Lauraceae	*Persea americana*	Avocado	Trolinger et al. (1978)
Lauraceae	*Sassafras albidum*	Sassafras	Trolinger et al. (1978)
Lythraceae	*Cuphea viscosissima*	Blue waxweed	Trolinger et al. (1978)
Lythraceae	*Lagerstroemia indica*	Crape myrtle	Blake et al. (2015)
Magnoliaceae	*Liriodendron tulipifera*	tulip-poplar	Trolinger et al. (1978)
Magnoliaceae	*Magnolia fraseri*	Mountain magnolia	Trolinger et al. (1978)
Magnoliaceae	*Magnolia tripetala*	umbrella magnolia	Trolinger et al. (1978)
Malvaceae	*Abelmoschus esculentus*	Okra	Trolinger et al. (1978)
Malvaceae	*Gossypium hirsutum*	cotton	Trolinger et al. (1978)
Malvaceae	*Hibiscus cannabinus*	kenaf	Trolinger et al. (1978), Colyer et al. (1992)
Malvaceae	*Sida spinosa*	Prickly mallow	Trolinger et al. (1978)
Malvaceae	*Tilia americana*	American basswood	Trolinger et al. (1978)
Meliaceae	*Melia azedarach*	Chinaberry tree	Chang (1995)
Menispermaceae	*Cocculus orbiculatus*	red-berried moonseed	Trolinger et al. (1978), Lee & Kim (2002)
Menispermaceae	*Menispermum canadense*	Moonseed	Trolinger et al. (1978)
Moraceae	*Artocarpus altilis*	Breadfruit	Hu et al. (2002)
Moraceae	*Broussonetia papyrifera*	Paper mulberry	Chang (1995)
Moraceae	*Ficus pumila*	Creeping fig	Wang et al. (2020)
Myrtaceae	*Eucalyptus benthamii*	Camden white gum	Aubrey et al. (2017)
Oleaceae	*Fraxinus americana*	White ash	Trolinger et al. (1978)
Passifloraceae	*Passiflora incarnata*	passion-flower	Trolinger et al. (1978)
Phytolaccaceae	*Phytolacca americana*	Pokeweed	Trolinger et al. (1978)
Plantanaceae	*Plantanus acerifolia*	London planetree	Trolinger et al. (1978)
Plantanaceae	*Platanus occidentalis*	Sycamore	Trolinger et al. (1978)
Polygonaceae	*Polygonum pensylvanicum*	Pinkweed	Trolinger et al. (1978)
Polygonaceae	*Polygonum scandens*	Climbing false buckwheat	Trolinger et al. (1978)
Polygonaceae	*Rumex crispus*	Yellow dock	Trolinger et al. (1978)
Rhamnaceae	*Zizyphus mauritiana*	Indian jujube	Su & Leu (1983)
Rosaceae	*Amelanchier arborea*	Serviceberry	Grand & Vernia (2004)
Rosaceae	*Malus domestica*	Apple	Trolinger et al. (1978)
Rosaceae	*Malus sylvestris*	Crab apple	Grand & Vernia (2004)
Rosaceae	*Prunus persica* var. *nectarina*	Nectarine	Trolinger et al. (1978)
Rosaceae	*Prunus subhirtella*	Japanese flowering cherry	Trolinger et al. (1978)

Family	Species	Common name	Reference
Rubiaceae	*Paederia foetida*	Chinese fevervine	Trolinger et al. (1978)
Sapindaceae	*Acer negundo*	Boxelder	Niedbalski et al. (1979)
Sapindaceae	*Acer nigrum*	Black maple	Trolinger et al. (1978)
Sapindaceae	*Acer plantanoides*	Norway maple	Trolinger et al. (1978)
Sapindaceae	*Acer pseudoplatanus*	Sycamore maple	Niedbalski et al. (1979)
Sapindaceae	*Acer rubrum*	Red maple	Trolinger et al. (1978)
Sapindaceae	*Acer saccharum*	Sugar maple	Trolinger et al. (1978)
Simaroubaceae	*Ailanthus altissima*	Tree-of-heaven	Trolinger et al. (1978)
Smilacaceae	*Smilax herbacea*	Carrion flower	Grand & Vernia (2004)
Solanaceae	*Solanum lycopersicum*	Tomato	Trolinger et al. (1978)
Solanaceae	*Solanum lycopersicum*	Tomato	Dillard et al. (1995)
Solanaceae	*Solanum tuberosum*	Potato	Trolinger et al. (1978)
Styracaceae	*Halesia diptera*	Siverbell	Holcomb (1994)
Ulmacae	*Ulmus parvifolia*	Chinese elm	Trolinger et al. (1978)
Vitaceae	*Parthenocissus quinquefolia*	Virginia creeper	Trolinger et al. (1978)
Vitaceae	*Parthenocissus* sp.	Virgin ivy	Trolinger et al. (1978)
Vitaceae	*Parthenocissus tricuspidata*	Boston ivy	Shin et al. (2019)
Vitaceae	*Vitis aestivalis*	Summer grape	Trolinger et al. (1978)
Vitaceae	*Vitis labrusca*	Fox grape	Trolinger et al. (1978)
Vitaceae	*Vitis palmata*	Catbird-grape	Trolinger et al. (1978)
Vitaceae	*Vitis rotundifolia*	Muscadine grape	Brennerman at al. (1993)
Vitaceae	*Vitis* sp.	Grape	Takahashi & Tanba (1980)
Vitaceae	*Vitis vinifera*	Grape	Trolinger et al. (1978), NARO (2013), Jayawardena et al. (2018)

References for Appendix 3

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Appendix 4. Glossary and Abbreviations

Term or abbreviation	Definiton
CMI	Climate (or composite) match index, a value that indicates the similarities between a location overseas and New Zealand (Phillips et al. 2018).
commercial production	Definition for the purposes of risk assessment: a process/system where activities, such as in-field monitoring, in-field pest control activities, harvesting, cleaning, sorting and grading have been undertaken to produce a commodity that is free of defects such as broken skin, rot and damage. Depending on the systems in place, these activities can be undertaken at any stage from the point of planting to the point of export.
commodity description	The commodity description defines the form of the commodity that is covered by the IRA, e.g. the commodity species and the countries under consideration. The commodity description may also include commercial production and/or grading requirements or other commodity quality specifications.
conditional non-host	Fruit or vegetable at a specified maturity and specified physical condition that cannot support the complete development of a particular pest species.
CPC	*Crop Protection Compendium*. A CABI database that provides information about crop pests.
disease	A disorder of structure or function in a plant, especially one that affects a specific location and is not simply a direct result of physical injury.
endemic	An animal, plant, pest, or disease that is native to and is not naturally found outside a defined geographical area.
establishment	Perpetuation, for the foreseeable future, of an organism or disease within an area after entry.
ERS	Emerging Risk System, an MPI workflow system that allows MPI to receive, process and store information about new biosecurity risks and to manage those risks in a timely fashion.
exposure	The process of the hazard organism moving from the commodity it arrived on to another host (also called “transfer”).
exotic	This word has different meanings in different fields, but in this document, it is defined as an animal, plant, pest or disease that is not indigenous to New Zealand.
hazard	A hazard is a pest (including arthropods and pathogens) or disease that is not present in the PRA area, e.g. New Zealand (or is present but still represents a biosecurity risk, e.g. is capable of vectoring pathogens not present in the PRA area), has the ability to establish and cause harm in the PRA area and is associated with the entry pathway (commodity/country of origin) under consideration.
indigenous	Native; organism originating or occurring naturally in a specified area.
inspection	Official visual examination of plants, plant products or other regulated articles to determine if pests are present and/or to determine compliance with phytosanitary regulations (FAO 2012).
introduced	Not indigenous, not native to the area in which it now occurs, having been brought into this area directly or indirectly by human activity.

Term or abbreviation	Definiton
IPPC	International Plant Protection Convention, a 1951 multilateral treaty overseen by the Food and Agriculture Organization that aims to secure coordinated, effective action to prevent and to control the introduction and spread of pests of plants and plant products.
ISPM	International Standards for Phytosanitary Measures, international standards adopted by the Conference of the Food and Agriculture Organization, the Interim Commission on phytosanitary measures or the Commission on phytosanitary measures, established under the IPPC.
IHS	Import Health Standard (See “Import Health Standard” for a definition.)
Import Health Standard	An import health standard (IHS) is a document issued under section 24A of the Biosecurity Act 1993. It states the requirements that must be met before risk goods can be imported into New Zealand.
IRA	Import Risk Analysis, an administrative process through which quarantine policy is developed or reviewed, incorporating risk assessment, risk management and risk communication.
MPI	Ministry for Primary Industries, New Zealand
non-host	Fruit or vegetable that will not support the complete development of a particular pest species regardless of the stage of maturity and physical characteristics.
NPPO	National Plant Protection Organisation, official service established by a government to discharge the functions specified by the IPPC.
NZOR	New Zealand Organisms Register, an actively maintained compilation of all organism names relevant to New Zealand: indigenous, endemic or exotic species or species not present in New Zealand but of national interest.
ONZPR	Official New Zealand Pest Register. MPI database that records organisms that may be associated with plants or plant products that are imported into New Zealand. Plant and plant product importers can use ONZPR to find out the quarantine status of a species – regulated or non-regulated.
pest	Any species, strain or biotype of plant, animal, or pathogenic agent injurious to plants or plant products.
pest-free area	An area in which a specific pest does not occur as demonstrated by scientific evidence and in which, where appropriate, this condition is being officially maintained.
pest-free place of production	A place of production in which a specific pest does not occur as demonstrated by scientific evidence and in which, where appropriate, this condition is being officially maintained for a defined period.
PFA	Pest-free area (See “Pest-free area” for a definition.)
PFPP	Pest-free place of production (See “Pest-free place of production” for a definition.)
PPIN	Plant Pest Information Network database (MPI), a record of organisms collected in New Zealand and identified by MPI’s Plant Health and Environment Laboratory.
PRA	Pest Risk Assessment or Pest Risk Analysis
QuanCargo	New Zealand border transaction database, detailing commercial consignments and interceptions of pests made by quarantine inspection (MPI).

Term or abbreviation	Definiton
quarantine pest	A pest of potential economic importance to the area endangered thereby, e.g. New Zealand, and not yet present there, or present but not widely distributed and being officially controlled.
systems approach	The integration of different pest risk management measures, at least two of which act independently, and which cumulatively achieve the appropriate level of phytosanitary protection. Measures can be undertaken at any stage from the point of planting to the point of export.
vector	An organism or object that transfers a pest, parasite, pathogen or disease from one area or host to another.