SCIENTIFIC OPINION

Scientific Opinion on the risk of *Phyllosticta citricarpa* (*Guignardia citricarpa*) for the EU territory with identification and evaluation of risk reduction options[1]

EFSA Panel on Plant Health (PLH) [2,3]

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ABSTRACT

The Panel conducted a risk assessment of *Phyllosticta citricarpa* (synonym *Guignardia citricarpa*) for the EU, also evaluating risk reduction options. *P. citricarpa* causes citrus black spot (CBS) and is absent from the EU. Under the worst case scenario of no risk reduction options, entry (and generally also transfer to host by ascospores and/or pycnidiospores) was rated as likely for citrus plants for planting and citrus fruit with leaves, moderately likely for citrus fruit without leaves, unlikely for citrus leaves and very unlikely for Tahiti lime fruit without leaves. Establishment was rated as moderately likely: hosts are widespread with long periods of susceptibility, current fungicide treatments do not prevent establishment, the climate is considered regularly suitable (with high uncertainty) for *P. citricarpa* ascospore production, dispersal and infection in many EU citrus production areas in September and October and in some locations in May (in Cyprus, Greek islands, Malta, South Italy and Southern Spain), and this is favoured locally by the use of sprinkler and micro-sprinkler irrigation. Spread with trade was rated as moderately likely. Model results indicated that CBS would mainly affect lemons and late maturing sweet orange and mandarin varieties with moderate consequences increased by the environmental impacts of additional fungicide treatments and extra quality controls and/or establishment of pest free areas required to meet export phytosanitary requirements. Consequences would be minor for early maturing citrus varieties (except in areas with potential spring infections or with sprinkler/micro-sprinkler irrigation) and minimal for citrus for processing. Uncertainty concerning the consequences is high mainly due to the lack of data on critical climate response parameters for the pathogen but also on impacts in areas at the limits of the current distribution. Since eradication and containment are very difficult, phytosanitary measures should focus on preventing entry. Current EU measures are judged to be effective.

KEY WORDS

Phyllosticta citricarpa, *Guignardia citricarpa*, citrus black spot, European Union, pest risk assessment, risk reduction options

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SUMMARY

The European Commission requested EFSA to prepare a pest risk assessment of the citrus black spot (CBS) fungus *Guignardia citricarpa* Kiely (all strains pathogenic to Citrus), to identify risk reduction options and to evaluate their effectiveness in reducing the risk to plant health posed by this organism in the EU territory[4]. EFSA was also requested to carry out an evaluation of the effectiveness of the present EU requirements[5] for *Guignardia citricarpa* in reducing the risk of introduction of this harmful organism into the EU. Furthermore, EFSA was requested to assess the risk associated with Citrus latifolia plants, including fruit, for the entry of this organism into the Union.

The Panel on Plant Health (PLH) conducted the risk assessment following the guidance documents for producing standardized assessments of pest risk (EFSA Panel on Plant Health (PLH), 2010) and risk reduction options (EFSA Panel on Plant Health (PLH), 2012). The Panel conducted the risk assessment in the absence of current and potential new risk reduction measures in place. The risk assessment therefore express the full risk posed by *P. citricarpa* to the EU territory corresponding to a situation where all current EU citrus requirements listed in Council Directive 2000/29/EC (in Annexes II, III, IV and V) and Commission Decisions 2004/416/EC and 2006/473/EC are lifted without being replaced by any other risk reduction measures.

The risk assessment covers *Guignardia citricarpa* Kiely, which has since been renamed *Phyllosticta citricarpa* (McAlpine) Aa. Other Phyllosticta species associated with citrus are not included.

After consideration of the evidence, the Panel reached the following conclusions:

With regard to the pest categorisation:

P. citricarpa is absent from the EU and has a potential for establishment and spread and for causing consequences in the risk assessment area.

With regard to the assessment of the risk to plant health of for the EU territory:

Under the worst case scenario in which all current EU citrus and *P. citricarpa* requirements listed in Council Directive 2000/29/EC and Commission Decisions 2004/416/EC and 2006/473/EC would be lifted, the conclusions of the pest risk assessment are as follows:

<u>Entry</u>

The probability of entry is rated as:

- moderately likely for the citrus fruit trade pathway (medium uncertainty),
- very unlikely for the Tahiti lime (*Citrus latifolia*) fruit trade pathway (high uncertainty),
- unlikely for citrus fruit import by the passenger traffic pathway (medium uncertainty),
- likely for the citrus fruit with leaves trade pathway (medium uncertainty),
- likely for the citrus plants for planting trade pathway (low uncertainty),
- likely for the Tahiti lime (*Citrus latifolia*) plants for planting trade pathway (high uncertainty),

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[4] The request was made pursuant to Article 29(1) and Article 22(5) of Regulation (EC) No 178/2002.

[5] The current requirements are listed in Annexes III, IV and V of Council Directive 2000/29/EC. as well as in Commission Decisions 2004/416/EC and 2006/473/EC,

- likely for the citrus plants for planting import by the passenger traffic pathway (medium uncertainty),
- unlikely for the citrus leaves pathway (medium uncertainty)

Establishment

The probability of establishment is rated as moderately likely because of:

- the widely availability of suitable hosts (no uncertainty),
- the climate suitability for ascospore maturation, dispersal and infection in many EU citrus growing areas in September and October and also for specific locations in May (high uncertainty),
- cultural practices (fungicides) not preventing establishment (low uncertainty),
- sprinkler and micro-sprinkler irrigation (still used in parts of the EU citrus growing areas) favouring establishment (low uncertainty),
- the simultaneous occurrence of host susceptibility and of weather conditions suitable for ascospore production and release, as well as for ascospore germination and infection (high uncertainty).

Overall, uncertainty on the probability of establishment is rated as high, mainly due to lack of knowledge on how *P. citricarpa* will respond to the EU climatic conditions. Although the environmental factors that are important in the various stages of the life cycle are known, scientific evidence is lacking concerning the precise thresholds for what values the organism requires, e.g. for the temperature and the wetness levels and durations. Further validation of the models applied by the Panel, especially for marginal areas within the current distribution of the CBS disease would be needed to reduce the uncertainty on the probability of establishment of *P. citricarpa* in the EU.

Spread

Natural spread of *P. citricarpa* is known to mainly occur by airborne ascospores. The distances the pathogen can spread by natural means is poorly known, The pathogen is very likely to spread with commercial fruit and plants for planting pathways. However, because spread is defined as the expansion of the geographical distribution of a pest within an area, the rate of spread depends not only on the rapidity of movement and the number of spread pathways but also the likelihood of finding a suitable environment for establishment. When the proportion of the citrus growing areas identified as potentially suitable for *P. citricarpa* is taken into account, the Panel considered that a rating of moderately likely is most appropriate for spread.

There is uncertainty about the potential natural spread of ascospores carried by wind over long distances, but this uncertainty does not concern the two main pathways of spread (intra-European trade in commercial fruit and plants for planting). Although there is uncertainty about the structure of the EU intra-trade network for the citrus plants for planting owing to lack of data, this does not influence the conclusions above.

Endangered area

The risk assessment has identified parts of the EU where host plants are present and where, based on simulation results, the climatic conditions are suitable for ascospore maturation and release followed by infection.

Conclusions from simulations of the release of ascospores based on gridded interpolated climate data from the EU citrus producing areas show that, in almost all years (for the 95 percentile), ascospore release in areas of Cyprus, Crete, Southern Greece, Italy, Spain and Portugal will start early enough to coincide with climatic conditions that are conducive to infection in September and October as simulated by EFSA (2008). However the same simulations indicate that the onset of ascospore release in most areas will start too late to coincide with the climatic conditions conducive to infection in April-May. Therefore, early maturing orange varieties might generally be infected in autumn only, which is when the availability of inoculum coincides with suitable conditions for infection. Due to the long incubation time, fruits from these early varieties will be harvested before symptoms appear. However, under such scenario, the late maturing orange varieties and lemons are expected to show CBS symptoms.

There are some areas, however, such as locations in Cyprus, the Greek islands, Malta and Southern Spain, where the onset of ascospores is also expected in May in half of the years simulated. In those years, it is expected that symptoms can develop on the fruit before harvest, and therefore have an impact on fruit quality.

The results from the simulations with interpolated (grid-based) weather data are consistent with the simulations based on weather data measured by agrometereological stations. The uncertainty is high as indicated in the establishment section.

Consequences

The results from the simulation of ascospore maturation, release and infection show that CBS will mainly develop and express symptoms in late maturing sweet orange varieties and on lemons grown within the endangered area. The expected consequences will be moderate for fresh fruit of late maturing citrus varieties and on lemons. There would be a potential for the reduction of disease incidence by chemical treatments, but this would cause environmental impact owing to the fact that in most EU citrus growing areas fungicides are not widely applied and that the most effective fungicide products are not currently registered for use on citrus in the EU MSs. In addition, to export citrus fruit to areas where CBS is regulated, additional fungicide treatments in the orchards, official inspections, quality controls in packinghouses and/or establishment of pest free areas might be needed to meet the phytosanitary requirements of these countries.

The consequences for fresh fruit of early maturing citrus varieties are assessed as minor. The impact on early maturing varieties would be sporadic in time and space, limited to years with rainy springs and/or to specific locations. However the impact could be higher in areas where spring infection, based on simulation results, is expected to be more frequent, such as some locations in Southern Spain, Cyprus, Malta and Greek islands.

The consequences would be minimal for citrus for processing, as external lesions or spots on citrus fruit are not a quality issue for the citrus for processing.

As for establishment, the uncertainties concerning the consequences are high due to: the lack of information on key parameters in the epidemiological models and on the incubation period, the lack of knowledge about the rate of disease build-up for this pathogen, the limited information available about the impact and the fungicides treatments in marginal areas within the current area of CBS distribution, e.g. the Eastern Cape Province of South Africa, where environmental conditions are more similar to those in the pest risk assessment area.

With regard to risk reduction options, the Panel notes that, to reduce the probability of entry of *P. citricarpa*, prohibition and import from pest-free areas have overall high to very high effectiveness with moderate to high feasibility for all pathways. Prohibition of parts of the host also has high effectiveness and very high feasibility with regard to the prohibition of citrus fruit with leaves and peduncles. For the fruit pathway, systems approaches as well as induction of precocious symtom expression also have high effectiveness and feasibility. For plants for planting, certification and preand post-entry quarantine systems were also found to have high effectiveness and feasibility (see Table 50 in Appendix).

To reduce the probability of entry and spread, the application of strict waste processing measures would be highly effective in reducing the transfer of *P. citricarpa* from infected citrus fruit, although with low feasibility. The effectiveness of eradication, as well as of containment, is assessed as low. The application of drip irrigation practices, cover crops and mulching will moderately reduce the probability of establishment.

Current EU phytosanitary measures appear to be focusing on the correct strategy to reduce the risk of *P. citricarpa* introduction. Once established, CBS is reported to be very difficult to eradicate or contain. The Panel thus concludes that every effort should be made to avoid the entry of the pathogen. Should the disease be reported from the risk assessment area, limited options are available to reduce the risk of establishment and spread. Current EU measures are overall judged to be effective in preventing the introduction of *P. citricarpa*.

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BACKGROUND AS PROVIDED BY THE EUROPEAN COMMISSION

The current European Union plant health regime is established by Council Directive 2000/29/EC on protective measures against the introduction into the Community of organisms harmful to plants or plant products and against their spread within the Community (OJ L 169, 10.7.2000, p.1).

The Directive lays down, amongst others, the technical phytosanitary provisions to be met by plants and plant products and the control checks to be carried out at the place of origin on plants and plant products destined for the Union or to be moved within the Union, the list of harmful organisms whose introduction into or spread within the Union is prohibited and the control measures to be carried out at the outer border of the Union on arrival of plants and plant products.

Citrus black spot is a serious disease of cultivated citrus plants caused by strains pathogenic to *Citrus* of the fungus *Guignardia citricarpa* Kiely. It is mainly a fruit disease and the unsightly lesions that develop on fruits do not cause post-harvest decay but render the fruits unmarketable. This pathogen is not known to occur in the EU.

Guignardia citricarpa (all strains pathogenic to *Citrus*) is a regulated harmful organism in the EU, listed in Annex IIAI of Council Directive 2000/29/EU. Annexes III, IVAI and VB of this Directive list requirements for the introduction into the EU of citrus plants, including fruits, which could be a pathway for the entry of this pathogen. In addition, temporary emergency measures are in place which impose additional requirements for the import of certain citrus fruits from Brazil in connection with *Guignardia citricarpa* (all strains pathogenic to *Citrus*) (Commission Decision 2004/416/EC; OJ L 151, 30.4.2004, p. 76). Certain third countries, as well as certain areas of third countries, are recognised as being free from *Guignardia citricarpa* (all strains pathogenic to *Citrus*) by Commission Decision 2006/473/EC (OJ L 187, 8.7.2006, p. 35).

In spite of the present import requirements against *Guignardia citricarpa* (all strains pathogenic to Citrus), infested citrus fruit is often intercepted during import inspections. In order to carry out an evaluation of the present EU requirements against *Guignardia citricarpa* (all strains pathogenic to *Citrus*), a pest risk analysis covering the whole territory of the EU is needed, which takes into account the latest scientific and technical knowledge for this organism, including the work on citrus black spot funded by EFSA in the context of the recent Prima Phacie project ('Pest risk assessment for the European Community plant health: A comparative approach with case studies'). EFSA has already worked on *Guignardia citricarpa* in the past, when it prepared a scientific opinion on a pest risk analysis and additional documentation on *Guignardia citricarpa* provided by South Africa (Question number: EFSA-Q-2008-299; doi:10.2903/j.efsa.2009.925). A recently published scientific paper (Yonow T, Hattingh V and de Villiers M, 2013. CLIMEX modelling of the potential global distribution of *Guignardia citricarpa* and the risk posed to Europe. Crop protection 44, 18-28) has modelled the potential global distribution of *Guignardia citricarpa* with the CLIMEX software also discussing the conclusions of the above mentioned EFSA scientific opinion (2008).

It is also important that the risk assessment provides clarity regarding the risk posed by *Citrus latifolia* plants, including fruit, for the introduction of *Guignardia citricarpa* into the Union. The Brazilian Phytosanitary Authorities have recently informed the Commission that they consider that *Citrus latifolia* is not a host of this fungus in field conditions, and that therefore the trade of *Citrus latifolia* fruit poses only a low risk for the introduction of *Guignardia citricarpa*. The Brazilian Phytosanitary Authorities have indicated that the following three documents, which are made available to EFSA for information, support their position:

- Pathogenicity, colony morphology and diversity of isolates of *Guignardia citricarpa* and *G. mangiferae* isolated from *Citrus* spp.. R. Baldassari et al., Eur J Plant Pathol (2008) 120:103-110

- Patogenicidade, morfologia de colônias e diversidade de isolados de *Guignardia citricarpa* e *G. mangiferae* obtidos de *Citrus* spp.. R. Baldassaeri, Doctoral thesis, June 2005
- Reporte sobre la evaluación de riesgos de *Guignardia citricarpa* Kiely en frutos cítricos; COSAVE 2004

TERMS OF REFERENCE AS PROVIDED BY THE EUROPEAN COMMISSION

EFSA is requested, pursuant to Article 29(1) and Article 22(5) of Regulation (EC) No 178/2002, to provide a pest risk assessment of *Guignardia citricarpa* (all strains pathogenic to *Citrus*), to identify risk reduction options and to evaluate their effectiveness in reducing the risk to plant health posed by this harmful organism. The area to be covered by the requested pest risk assessment is the EU territory. In the risk assessment EFSA is also requested to provide an opinion on the effectiveness of the present EU requirements against *Guignardia citricarpa* (all strains pathogenic to *Citrus*), which are listed in Annex III, IV and V of Council Directive 2000/29/EC, as well as in Commission Decision 2004/416/EC and Commission Decision 2006/473/EC, in reducing the risk of introduction of this pest into the EU territory. In its scientific opinion EFSA is requested to indicate what is the risk posed by *Citrus latifolia* plants, including fruit, for the introduction of this organism into the Union.

ASSESSMENT

1. Introduction

1.1. Purpose

This document presents a pest risk assessment prepared by the EFSA Scientific Panel on Plant Health (hereinafter referred to as the Panel) for *Phyllosticta citricarpa* (synonymous *Guignardia citricarpa*) in response to a request from the European Commission. The opinion includes the identification and evaluation of risk reduction options in terms of their effectiveness in reducing the risks posed by this organism.

1.2. Scope

This risk assessment is for *Phyllosticta citricarpa* (McAlpine) Aa, which was previously named *Guignardia citricarpa* Kiely (see Section 3.1.1.1).

The species *Phyllosticta citriasiana* Wulandari, Crous & Gruyter, that has recently been associated with tan spot on pomelo (*Citrus maxima* (Burm.) Merr.) fruit, and *Phyllosticta capitalensis* Henn., that is not pathogenic to citrus, as well as other citrus-associated *Phyllosticta* species, are not included in this pest risk assessment (see Section 3.1.1).

The pest risk assessment area is the territory of the European Union (hereinafter referred to as the EU) with 28 Member States (hereinafter refrerred to as EU MSs)[6], restricted to the area of application of Council Directive 2000/29/EC, which excludes Ceuta and Melilla, the Canary Islands and the French overseas departments.

2. Methodology and data

2.1. Methodology

2.1.1. The guidance documents

In order to maximise transparency and consistency, the risk assessment has been conducted in line with the principles described in the document 'Guidance on a harmonised framework for pest risk assessment and the identification and evaluation of pest risk management options' (EFSA PLH Panel, 2010). The evaluation of risk reduction options (also referred as risk management options) has been conducted in line with the principles described in the above mentioned guidance (EFSA PLH Panel, 2010), as well as with the 'Guidance on methodology for evaluation of the effectiveness of options to reduce the risk of introduction and spread of organisms harmful to plant health in the EU territory' (EFSA PLH Panel, 2012).

Harmonized rating descriptors used in this opinion that follow the EFSA guidance documents are presented in Appendix A.

When expert judgement and/or personal communication have been used, justifications and evidence are provided to support the statements. Personal communications have been considered only when in written form and supported by evidence, and when other sources of information have not been publicly available.

[6] When the dataset utilised do not yet include Croatia (that joined the EU on July 2013), it is specified that they refer only to the EU 27.

2.1.2. Methods used for conducting the risk assessment

The Panel conducted the risk assessment considering the absence of current requirements listed in Annexes II, III, IV and V of Council Directive 2000/29/EC. and in Commission Decisions 2004/416/EC and 2006/473/EC, but under the assumption of a standard citrus disease management in the country of origin to comply with fruit quality standards, However, all the data on imports and interceptions presented in this document were obtained under the regulations currently in place in the EU. These data should be interpreted with caution because quantities of imported products will probably change if the regulations are removed and because interception numbers depend on the procedure of import control currently in place at the EU borders.

The conclusions for entry, establishment, spread and impact are presented separately. The descriptors for qualitative ratings given for the probabilities of entry and establishment and for the assessment of impact are shown in Appendix A.

2.1.3. Methods used for evaluating the risk reduction options

The Panel identifies potential risk reduction options and evaluates them with respect to their effectiveness and technical feasibility, i.e., consideration of technical aspects which influence their practical application. The evaluation of efficiency of risk reduction options in terms of the potential cost-effectiveness of measures and their implementation is not within the scope of the Panel evaluation.

The descriptors for qualitative ratings given for the evaluation of the effectiveness and technical feasibility of risk reduction options are shown in Appendix A.

2.1.4. Level of uncertainty

For the risk assessment conclusions on entry, establishment, spread and impact and for the evaluation of the effectiveness of the risk reduction options, the levels of uncertainty have been rated separately.

The descriptors for qualitative ratings given for the level of uncertainty are shown in Appendix A.

2.2. Data

2.2.1. Data collection

2.2.1.1. Data on cultivation areas and trade

Data on cultivation areas and trade (imports or/and exports) were collected from Eurostat and extracted from January to May 2013. In detail:

- Data on cultivation areas were collected from the apro_cpp database[7];
- Trade data were collected from the Comext database[8] for data since 1988 and from the Nimexe database for data from 1976 to 1987.

[7] http://epp.eurostat.ec.europa.eu/portal/page/portal/agriculture/data/database

[8] http://epp.eurostat.ec.europa.eu/newxtweb/mainxtnet.do

Table 1: The following trade commodities of the HS and CN classifications were used for the trade data since 1988:

Code	Name
0805	Citrus fruit, fresh or dried
080510	Fresh or dried oranges
080520	Fresh or dried mandarins incl. Tangerines and satsumas, clementines, wilkings and similar citrus hybrids
080540	Fresh or dried grapefruit
08055010	Fresh or dried lemons "citrus limon, citrus limonum"
08055090	Fresh or dried limes "citrus aurantifolia, citrus latifolia"
080590	Fresh or dried citrus fruit (excl. Oranges, lemons 'citrus limon, citrus limonum & citrus hybrids)

Table 2: The following trade commodities were used for the trade data during the period 1976-1987:

Code	Name
802	Citrus fruit, fresh or dried
80,202	Fresh sanguines and semi-sanguines from 1 april to 30 april
80,203	Fresh navels, navelines, navelates, salustianas, vernas, valencia lates, maltese, shamoutis, ovalis,trovita and hamlins from 1 april to 30 april, other than sanguines and semi-sanguines
80,205	Other fresh, sweet oranges from 1 april to 30 april except navels, navelines, navelates, salustianas, vernas, valencia lates, maltese, shamoutis, ovalis, trovita, hamlins, sanguines and semi-sanguines
80,206	Fresh sanguines and semi-sanguines from 1 may to 15 may
80,207	Fresh navels, navelines, navelates, salustianas, vernas, valencia lates, maltese, shamoutis, ovalis,trovita and hamlins from 1 may to 15 may other than sanguines and semi-sanguines
80,209	Other fresh, sweet oranges from 1 may to 15 may except navels, navelines, navelates, salustianas, vernas,valencia lates maltese, shamoutis, ovalis, trovita, hamlins and sanguines and semi-sanguines
80,212	Fresh sanguines and semi-sanguines from 16 may to 15 october
80,213	Fresh navels, navelines, navelates, salustianas, vernas, valencia lates, maltese, shamoutis, ovalis, trovita and hamlins except sanguines and semi-sanguines from 16 may to 15 october
80,215	Other fresh, sweet oranges from 16 may to 15 october except navels, navelines, navelates, salustianas, vernas, valencia lates, maltese, shamoutis, ovalis, trovita, hamlins and sanguines and semi-sanguines
80,216	Fresh sanguines and semi-sanguines from 16 october to 31 march
80,217	Fresh navels, navelines, navelates, salustianas, vernas, valencia lates, maltese, shamoutis, ovalis,trovita and hamlins except sanguines and semi-sanguines from 16

	october to 31 march
80,219	Other fresh, sweet oranges from 16 october to 31 march except navels,navelines, navelates, salustianas, vernas,valencia lates, maltese, shamoutis, ovalis, trovita, hamlins and sanguines and semi-sanguines
80,224	Oranges, other than sweet, fresh oranges from 1 april to 15 october
80,227	Oranges, other than sweet, fresh oranges from 16 october to 31 march
80,228	Clementines
80,229	Monreales and satsumas
80,231	Mandarins and wilkings
80,232	Clementines
80,234	Tangerines
80,237	Other similar citrus hybrids except monreales, satsumas, mandarins, wilkings, clementines
80,250	Lemons
80,270	Grapefruit
80,290	Citrus fruit, fresh or dried, other than oranges, mandarins and hybrids, lemons and grapefruit

2.2.1.2. Interception data

The extraction of the citrus interceptions data from the Europhyt database was conducted on 19 April 2013.

2.2.1.3. Climate and weather data

Weather data from agrometeorological stations and interpolated climate data from JRC, as described in the previous opinion by the EFSA Panel on Plant Health (PLH) (2008), were used for simulations of ascospore production and release. The interpolated climate data grids of CRU CL 1.0 (New et al., 1999) and CRU CL 2.0 (New et al., 2002) as well as climate data grids covering the EU produced by JRC were used for CLIMEX simulations.

2.2.2. Literature search

The literature on CBS and *P. citricarpa* up to April 2013 was searched using the following search engines: Web of Science, CAB Abstracts, and Google Scholar. The keywords used were "*Phyllosticta citricarpa*", "*Guignardia citricarpa*", "Citrus Black Spot", and "citricarpa". For the meta-analysis of published treatment experiments against *P. citricarpa*, the last two keywords were first combined with "fungicide", and then with "trial". The literature cited in the papers retrieved was inspected and papers citing retrieved papers were examined. The Panel took advantage of the extensive bibliographic collection on CBS already gathered for the scientific opinion of the EFSA Panel on Plant Health (PLH) in 2008 and focused the literature search on publications that have appeared since then.

3. Pest risk assessment

3.1. Pest categorisation

3.1.1. Identity of pest

3.1.1.1. Taxonomic position

Citrus black spot disease (CBS) was first described in Australia (Cobb, 1897; Kiely, 1948). The causal agent of CBS was identified as *Guignardia citricarpa* Kiely (anamorph *Phyllosticta citricarpa* (McAlpine) Aa), which was also detected on asymptomatic citrus trees as well as on other hosts in

Australia and South Africa (Kiely 1948; Wager 1952). For many years, the coexistence of pathogenic and non-pathogenic strains of *G. citricarpa* was assumed. However, based on pathogenicity tests, McOnie (1964a) demonstrated that the non-pathogenic strains belonged to other *Guignardia* species that did not play a role in the causation of CBS. More recently, based on morphological, molecular and physiological analyses, Baayen et al. (2002) identified isolates obtained from CBS-affected fruits as *G. citricarpa* and isolates from asymptomatic citrus and other hosts as *G. mangiferae* A.J. Roy (anamoph *P. capitalensis* P. Hennings). Baldassari et al. (2008) demonstrated by means of field inoculations that only those isolates identified as *G. citricarpa* were pathogenic to sweet orange (*C. sinensis* Osbeck) fruit. In this study, isolates of *G. citricarpa* were also obtained from asymptomatic Tahiti lime (*C. latifolia* Tanaka) fruit, that had previously been considered to be resistant to CBS, but is apparently an asymptomatic host.

In 2011, a new code for fungal nomenclature was approved by the International Botanical Congress in Melbourne. The current ‘Melbourne Code’ abolishes the dual nomenclature for fungi, and gives priority to the oldest name irrespective of whether it is teleomorphic (sexual reproduction) or anamorphic (asexual reproduction) (Norvell, 2011). In the case of the CBS pathogen, the anamorph name *P. citricarpa* has priority over the teleomorph name *G. citricarpa*, and it should be now used as the only identifier of this species. Since the original type material of *P. citricarpa* has been lost, Glienke et al. (2011) designated a new type specimen for this species (epitype). An epitype for the non-pathogenic *P. capitalensis* was also designated, but it was defined as a different species to *G. mangiferae*, that was previously considered to be its teleomorph and it is currently regarded as a pathogen of mango and not associated with citrus (Glienke et al., 2011; Wikee et al., 2011; 2013). Glienke et al. (2011) defined a new species, *P. citribraziliensis* C. Glienke & Crous, sp. nov., based on three isolates obtained from asymptomatic citrus leaves (*Citrus* sp.) in Brazil. Another new species, *P. citriasiana* Wulandari, Crous & Gruyter, was detected in diseased fruits of pomelo (*Citrus maxima* (Burm.) Merr.) in intercepted consignments imported into the EU from Asia. This fungus was associated with a disease known as citrus tan spot, but confirmatory pathogenicity test have not been published so far (Wulandari et al.l 2009). In extensive surveys conducted in China, Wang et al. (2012) found a new species on citrus, *P. citrichinaensis* X.H. Wang, K.D. Hyde & H.Y. Li, that was associated with leaf and fruit spots of citrus, but the results of confirmatory pathogenicity tests are not available to date. Thus, while new knowledge on the *Phyllosticta* species associated with citrus is continuously emerging, the current knowledge supports the conclusion that only *P. citricarpa* has proven to be pathogenic to citrus and a potential threat to citrus cultivation in regions that are suitable for this pathogen.

3.1.1.2. Biology and life cycles

The primary infection cycle of the CBS pathogen is driven by ascospores formed into sexual fruiting bodies (pseudothecia) in the leaf litter. Citrus leaves drop all year around and mature pseudothecia are formed between 23 to 180 days after leaf fall depending on the temperature and humidity (Lee and Huang 1973; Kotzé 1981). Ascospores of *P. citricarpa* in spore traps are morphologically indistinguishable from those of the non-pathogenic species *P. capitalensis*, which is widespread in CBS-affected areas. Thus, most data on ascospore dynamics available in the literature are based on mixed populations with unknown proportions of both species and should be interpreted with caution. Studies from South Africa and Taiwan indicated that maturation of ascospores occurs practically simultaneously in early summer on infected leaves abscised during late autumn, winter and early spring (Kotzé, 1963; McOnie, 1964b; Lee and Huang, 1973). Once mature, ascospores are mainly released during rain events. Studies conducted in the Mpumalanga province in South Africa indicated that at least 3 mm of precipitation are required for a significant release of ascospores (McOnie 1964b). The presence of frequent dews was associated with ascospore production in Australia, but its role in ascospore release was not confirmed (Kiely, 1948). Irrigation might also trigger ascospore release, but all the studies available were conducted in regions where citrus are seldom irrigated during the time of ascospore production.

In Sao Paulo, Brazil, comparatively low to moderate numbers of ascospores were produced from October to March with peak production in January and February (Reis et al., 2003). In the Limpopo province of South Africa, ascospore release occured mainly from November to March with the highest numbers from December to January (McOnie 1964b; 1964c). Recent studies in the Limpopo province in South Africa indicated a similar period of ascospore availability from October to March. Degree-day models have been developed based on these data to predict the onset and duration of ascospore release in this region in South Africa (Fourie et al., 2013) and as a function of temperature and wetness in Misiones in Argentina (Dummel et al., 2012). Once released, ascospores are disseminated by air currents and infect susceptible leaves and fruit. Under artificial inoculation conditions, leaves of lemon (*C. limon* (L.) Osbeck) cv. Eureka were susceptible for at least 10 months and sweet orange (*C. sinensis*) cv. Valencia for up to 8 months (Truter et al., 2004). In South Africa, sweet orange fruit was considered to be susceptible to infection up to four months after fruit set (Kotzé, 1981), but field studies conducted in Brazil and Ghana indicated a susceptibility period of 6 and 7 months after fruit set, respectively (Reis et al., 2003; Baldassari et al., 2006; Brentu et al., 2012), although longer periods were not evaluated.

CBS occurs mainly in subtropical citrus-growing regions characterized by a summer rainfall pattern (Kotzé, 1981; 2000) and high annual precipitation. However, the disease is also present in drier areas such as the Eastern Cape province in South Africa (Paul et al., 2005) with an annual rainfall of about 400 mm. The full range of temperatures and humidities suitable for ascospore infection have not been determined experimentally, and only ascospore germination rates and field infection data are available in the literature. According to Kotzé (1963), the conditions required for ascospore germination varied from 15-29.5 °C and 15-38 hours of wetness. McOnie (1967) found that ascospores were able to infect with at least 15 hours of continuous wetness. In field studies conducted in Sao Paulo, Brazil, sweet orange fruit were infected with nearly 14 h of wetness per day and 22 to 25 °C, but temperatures out of this range were not evaluated (Reis et al., 2006).

The secondary infection cycle of *P. citricarpa* is caused by pycnidiospores (conidia) formed into asexual fruiting bodies (pycnidia) on lesions in fruit, twigs and leaf litter. Pycnidiospores are splash-dispersed or washed-off by rain to relatively short distances, infecting susceptible leaves and fruit. Under *in vitro* conditions, pycnidiospores of *P. citricarpa* can germinate and form appressoria between 10-40 ºC and 12–48 hours of wetness (Noronha, 2002). Although the role of pycnidiospores in CBS epidemics was recognized in pioneering works in Australia and Zimbabwe (Kiely, 1948; Whiteside, 1967), pycnidispores were later considered insignificant as a source of inoculum in South Africa (Kotzé, 2000). Pycnidiospores produced in fruit lesions were indicated as a potential source of inoculum only where out-ofseason fruit or late-hanging fruit with lesions remain on the trees after blossoming and fruit set (Kotzé, 1981). In a study conducted in Brazil, it was observed that pycnidiospores of *P. citricarpa* that had formed on the lesions of sweet orange fruit from the previous harvest did not significantly increase the severity of disease on the fruits of the subsequent harvest period (Baldassari et al., 2006). However, recent spatio-temporal epidemiological studies in Sao Paulo, Brazil, demonstrated that under field conditions rain-dispersed pycnidiospores formed in fruit and twigs have an important role in increasing the severity of CBS in sweet orange trees in this region (Sposito et al., 2007, 2008, 2011).

The disease is characterized by a relatively long incubation period and fruit symptoms become visible several months after infection. The duration of the incubation period is affected by environmental factors. In general, high temperatures and increased exposure to sunlight reduce the duration of the incubation period and augment disease severity. The disease is more severe on old and drought stressed trees than in young and vigorous trees (Kotzé, 1963; Brodrick and Rabie, 1970; Kotzé, 1981; Ninin et al., 2012). The incubation period is also affected by the growth stage in which the fruit was infected. In artificial inoculations conducted under greenhouse conditions, the incubation period ranged from over 200 days for 3 cm diameter sweet orange fruit to about 50 days for 7 cm diameter fruit (Aguiar et al., 2012). Foliar lesions of CBS appear as small sunken necrotic spots surrounded by a dark brown ring. However, they are rare and only present in lemons or trees in poor condition (Kotzé, 1981; 2000). One feature that has been observed for CBS is that the pathogen may be present for many years in a region before the disease reaches epidemic proportions. In Mpumalanga province in South Africa, symptoms were present for over three decades before control measures became necessary (Kotzé, 1981). Whilst the existence of a lag phase following an initial introduction to a new area is a general feature of a pathogen's epidemiology at various scales, this process has not been studied in detail for *P. citricarpa*. Another general feature of pathogen's is that they can be sporadic in time and space. During a lag phase, inoculum is built up through multiplication in small scale epidemics where the fungus is present mostly as latent mycelia in asymptomatic citrus fruit and leaves. When sufficient inoculum has been built up and if weather conditions become suitable at a specific location, epidemics can develop and cause severe disease impact. Therefore, estimates of disease progression, even in drier regions, such as the Eastern Cape in South Africa where CBS emerged more recently (McOnie 1964; Paul et al., 2005), should be interpreted with caution.

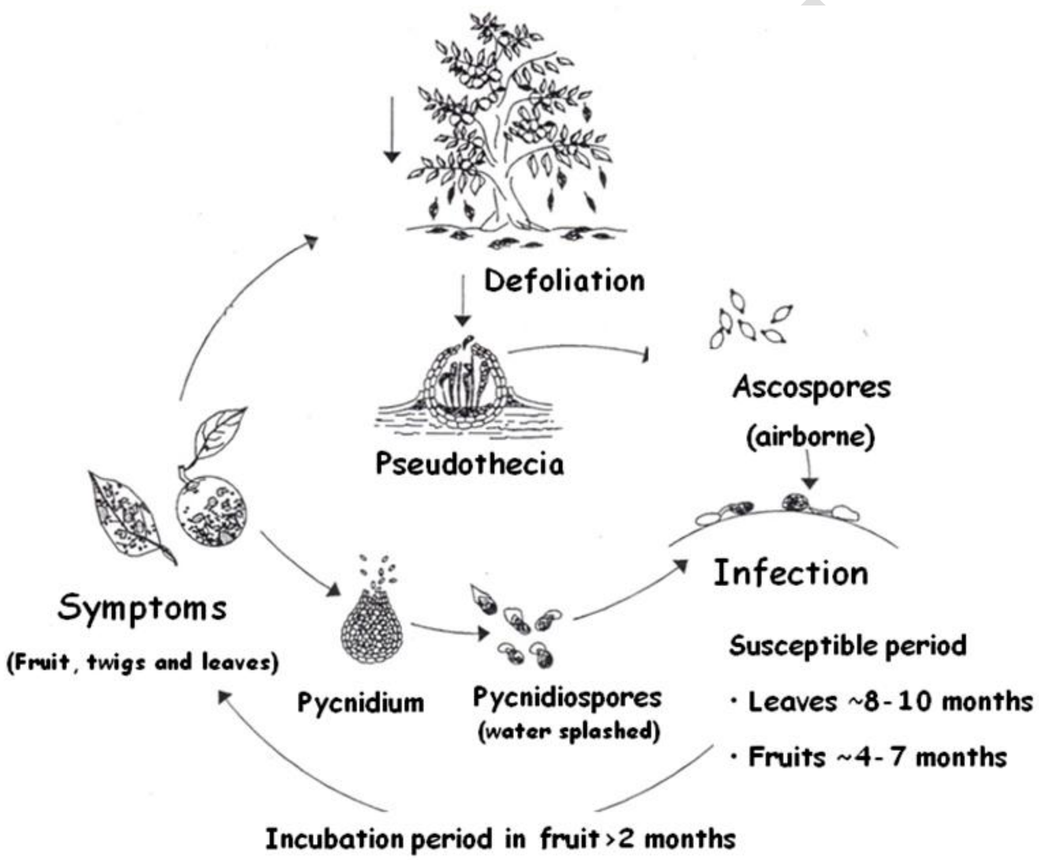


Figure 1: Life cycle of *Phyllosticta citricarpa* (adapted from Timmer (1999) © APS press and modified according to Reis et al. (2003), Truter et al. (2004), Aguiar et al. (2012) and Brentu et al. (2012)).

3.1.1.3. Detection and identification

The formation of CBS lesions in affected asymptomatic fruit can be induced by treatment with ethephon and storage under continuous light and warm temperatures (Baldassari et al., 2007). The pathogen can be isolated from fruit lesions by plating fragments of affected tissues in agar media. The formation of pycnidia and pycnidiospores can be induced by maintaining fruit lesions under high humidity conditions (EPPO, 2003). Some differential morphological and physiological characteristics have been associated with the colonies of *P. citricarpa*, such as pycnidiospores with barely visible mucoid sheaths, production of infertile perithecia and the formation of a yellow pigment on oatmeal agar (Baayen et al., 2002). However, the use of molecular procedures is generally required for an accurate identification of the pathogen. Several PCR methods are available for *P. citricarpa* (Bonants et al., 2003; Meyer et al., 2006; Peres et al., 2007; van Gent-Pelzer et al., 2007; Stringari et al., 2009). Nevertheless, the specificity of these methods needs to be reassessed considering the recent description of new *Phyllosticta* species on citrus (Wulandari et al., 2009; Glienke et al., 2011; Wang et al., 2012). New molecular methods have recently been proposed (Wang et al., 2012; Stammler et al., 2013; Tomlinson et al., 2013), but systematic performance tests still need to be completed.

3.1.1.4. Citrus taxonomy and host range of *P. citricarpa*

The vast majority of citrus fruits and their wild relatives are native to south-eastern Asia, the East Indian Archipelago, New Guinea, Melanesia, New Caledonia, and Australia; another group occurs in tropical Africa. The commonly cultivated citrus fruits belong to three genera: *Citrus*, *Fortunella* and *Poncirus* that are all closely related and belong to the subtribe Citrinae, the tribe Citreae, the orange subfamily Aurantioideae and the plant family Rutaceae. All the genera have persistent unifoliolate or simple leaves except the monotypic genus *Poncirus*, which has trifoliolate, deciduous leaves.

The genus *Fortunella* (kumquat) includes species of small trees and shrubs. All species have small leaves and orange coloured fruits of small size.

The genus *Poncirus* includes a single species, *P. trifoliata*, with trees of small size and trifoliate leaves. It differs from all the other true citrus fruit trees, which are found only in tropical or subtropical regions. Having penetrated far into the temperate zone in northeastern Asia, it has become a deciduous tree with small leaf buds and larger scale-covered flower buds (formed in early summer) that pass the winter on the leafless terminal twigs and open before (and sometimes with) the leaves early in the following spring. *Poncirus* hybridizes freely with *Citrus*. Such hybrids, called citranges, are often used as rootstocks.

The genus *Citrus* is divided into two very distinct subgenera, *Citrus* and *Papeda*, that are easily distinguished by leaf, flower, and fruit characteristics. The subgenus *Citrus* includes all the commonly cultivated species of citrus, all of which have fruit with pulp-vesicles filled with juice, free, or almost free, from droplets of oil, which are located in the rind. Species from the genus *Citrus* are the most important from an agronomical point of view. The botanical classification within this genus is not unique. Nowadays, the classification in common use is that established by Swingle (1967).

Table 3: Main citrus species cultivated worldwide.

Botanical name	Common English name
Fortunella spp.	Kumquat
Poncirus trifoliata (L.) Raf.	Trifoliate orange
Citrus medica L.	Citron
Citrus limon (L.) Burm.f.	Lemon
Citrus aurantifolia (Christm.) Swingle	Key lime
Citrus latifolia Tanaka	Tahiti lime
Citrus limettioides Tanaka	Sweet lime
Citrus hystrix DC	Kaffir lime
Citrus aurantium L.	Sour orange
Citrus sinensis Osbeck	Sweet orange
Citrus reticulata Blanco	Mandarin
Citrus unshiu (Swingle) Marcow.	Satsuma mandarin
Citrus maxima (Burm.) Merr.	Pomelo
Citrus paradisi Macfad.	Grapefruit

All commercial citrus species and cultivars are considered to be susceptible to CBS, except for sour orange (*C. aurantium*) (Kotzé, 1981) and Tahiti lime (*C. latifolia*) (Baldassari et al., 2008). In the case of sour orange, *P. citricarpa* was isolated from asymptomatic leaves in Brazil (Wickert et al., 2009). Isolates obtained in this country from CBS lesions and other fruit blemishes were reported by several studies (Baayen et al., 2002; Wulandari et al., 2009; Glienke et al., 2011), although no evidence of reproduction on this citrus species was found. Tahiti lime is reported not to exhibit CBS symptoms under field conditions, even in areas with high inoculum pressure. However, *P. citricarpa* was isolated in Sao Paulo, Brazil, from asymptomatic fruit and leaves of Tahiti lime (Baldassari et al., 2008; Wickert et al., 2009). Although there is no documented evidence of *P. citricarpa* reproduction on Tahiti lime fruit, it can colonize and form viable ascospores in Tahiti lime leaves suggesting that this citrus species may well play a role in CBS epidemiology (Baldassari et al., 2008).

Lemon (*C. limon*) is considered to be the citrus species that is most susceptible to CBS and it has been stated that the first disease outbreaks in a region always occurred in lemon orchards and later on spread to adjacent citrus orchards (Kotzé, 1981). However, CBS emerged recently in Florida (USA) directly in sweet orange orchards (Schubert et al., 2010). Late maturing cultivars of sweet orange were considered more susceptible than early maturing ones (Timmer, 1999). However, cultivar field trials conducted in Brazil as well as studies comparing the rate of disease progress indicated that cultivar reaction to the disease is more linked to the interaction of environmental factors with the dynamics of fruit maturation (Sposito et al., 2004; Sousa and de Goes, 2010).

Recent surveys conducted in China indicated that pomelo (*C. maxima*) is not affected by *P. citricarpa* (Wang et al., 2012). However, more data from other geographic regions as well as proper pathogenicity tests are needed to completely exclude this citrus species as a potential host.

3.1.1.5. Reports of impact in the area of current distributions

In most of the the area of its current distribution, *P. citricarpa* is reported to cause severe quality and yield losses to citrus fruit production. The apparent absence of severe impact at specific locations, e.g. in Addo, Eastern Cape, South Africa, where the pathogen is reported to “persist but not flourish” (Yonow et al., 2013), could be due to the relatively recent emergence of the disease as well as to the fungicide schedules currently in place. Several types of CBS symptoms including hard spot, virulent spot, and false melanose occur on the rind of affected fruit (Fig. 2), reducing its commercial value for the fresh market (Kotzé, 2000). Premature fruit drop due to CBS causes significant yield loss in Brazil, and probably in other citrus regions of the world (Reis et al., 2006). Leaf lesions are seldom seen in well-managed sweet orange orchards and they appear more commonly on lemons (Kotzé, 2000). In order to obtain more information about disease impacts, the Panel undertook a meta-analysis of recorded disease incidence in untreated and fungicide-treated plots from published field trials for the control of CBS. The results from this meta-analysis are described in section 3.6.1.1.

Figure 2: Left: fruits of sweet orange with symptoms of citrus black spot caused by *Phyllosticta citricarpa*; right: lesions of citrus black spot in a lemon fruit with pycnidia of *P. citricarpa*.

3.1.2. Current distribution

Reports of *P. citricarpa*, from EPPO PQR (EPPO, 2013), from scientific and technical literature and from interception records by EU MSs, are given in the table 2 below.

P. citricarpa (as *G. citricarpa* (all strains pathogenic to citrus)) is listed in the EU Directive 2000/29/EC as not known to occur in the EU and is reported in the EPPO PQR as absent from all the citrus producing EU MSs (Croatia, Cyprus, France, Greece, Italy, Malta, Portugal and Spain).

Table 4: Reports of Phyllosticta citricarpa from the EPPO PQR (EPPO, 2013), interception records (Europhyt, online), scientific and technical literature.

Country	State/region	Reports	Source
Africa			
Benin		Absent, Nevertheless detected in 3 consignments exported to France	EPPO, 2013; Europhyt (online)
Cameroon		Absent However detected in 2 consignments exported to UK (2006 on *C. sinensis*) and to CH (in 2012 on *C. maxima*)	EPPO, 2013; Europhyt (online)
Ghana	Eastern	Widely distributed	Brentu et al., 2012
Ghana	Ashanti	Widely distributed	Brentu et al., 2012
Guinea		Absent, However detected in 2 consignments exported to France (1 in *C. maxima* in 1999; 1 in *C. sinensis* in 2000)	EPPO, 2013; Europhyt (online)
Kenya		Present, no details	EPPO, 2013
Mozambique		Present, no details	EPPO, 2013
South Africa		Present, restricted distribution	EPPO, 2013
South Africa	KwaZulu-Natal		Carstens et al., 2012
South Africa	Mpumalanga		Carstens et al., 2012
South Africa	Limpopo		Carstens et al., 2012
South Africa	North West		Carstens et al., 2012
South Africa	Eastern Cape		Carstens et al., 2012
Swaziland		Absent However detected in 26 consignments to EU MSs	EPPO, 2013, Europhyt (online)
Uganda		Present, few occurrences	EPPO, 2013
Zambia		Present, no details	EPPO, 2013
Zimbawe		Present, no details	EPPO, 2013
America			
Argentina		Present, restricted distribution	EPPO, 2013
Brazil		Present, restricted distribution	EPPO, 2013
Brazil	Rio Grande do Sul	Present, no details	EPPO, 2013
Brazil	Rio de Janeiro	Present, no details	EPPO, 2013
Brazil	Sao Paulo	Present, no details	EPPO, 2013
Cuba		Present, no details	EPPO, 2013
United States of America		Present, few occurrences	EPPO, 2013
United States of America	Florida	Present, few occurrences	EPPO, 2013
Uruguay		Present, no details. Detected in 3 consignments to EU MSs from 2001 to 2010 (2 on *C. sinensis*, 1 on *C. reticulate*	USDA APHIS, 2012a; EPPO, 2013; Europhyt (online)
Asia			
Bangladesh		Absent Detected in 20 consignments to UK	EPPO, 2013; Europhyt (online)
Bhutan		Present, no details	EPPO, 2013
China		Present, restricted distribution	EPPO, 2013
China	Fujian	Present, no details	EPPO, 2013
China	Guangdong	Present, no details	EPPO, 2013

China	Sichuan	Present, no details	EPPO, 2013
China	Xianggang (Hong Kong)	Present, no details	EPPO, 2013
China	Yunnan	Present, no details	EPPO, 2013
China	Zhejiang	Present, no details	EPPO, 2013
Indonesia		Present, no details	EPPO, 2013
Indonesia	Java	Present, no details	EPPO, 2013
Philipppines		Present, no details	EPPO, 2013
Taiwan		Present, no details	EPPO, 2013
Thailand		Deetected in 12 consignments to EU MSs	Europhyt (online)
Vietnam		Detected in 8 consignments to the Netherlands on *C. maxima*	Europhyt (online)
Oceania			
Australia		Present, restricted distribution	EPPO, 2013
Australia	(coastal) New South Wales	Present, no details	EPPO, 2013; Miles et al., 2013
Australia	Queensland	Present, no details	EPPO, 2013
Australia	Northern Territory	Cited as present in references	Miles et al., 2013 (although Northern Territories are recognised as being free from *G citricarpa* by Commission Decision 2006/473/EC)
Australia	Victoria	Present, no details	EPPO, 2013
New Zealand		Present, no details	EPPO, 2013
Vanuatu		Present, no details	EPPO, 2013

3.1.3. Regulatory status in the EU

3.1.3.1. History of regulatory status in the citrus producing EU MSs

In most EU MSs growing citrus, the import of citrus plants and plant parts, including fruit, has been historically forbidden following national plant quarantine rules, until, after joining the European Community/EU, common EC/EU phytosanitary measures introducing also particular requirements for citrus fruit and *P. citricarpa* were implemented.

In Spain, the import of fresh fruit, live plants and plant parts of citrus and other woody fruit species from Japan, USA, Canada and New Zealand has been prohibited since 1929 (Real Orden Nº 976, Gaceta de Madrid 114: 464-465). In 1934, this prohibition was expanded to plant material imported from Portugal, Argentina, Brazil, Mexico and South Africa (Orden, Gaceta de Madrid 228: 1526). These regulations were derogated in 1987 (Orden 7366, BOE 71:8395-8411) when the European Directive 77/93/CEE was implemented in Spain. This new regulation prohibited the import of all kind of citrus material from any country. The import of citrus fruit in Spain was first allowed in 1993, but with specific provisions to avoid the introduction of *P. citricarpa* and other harmful organisms, when the European Directive 77/93/CEE was implemented (Real Decreto 2071/1993, BOE 300:35603-35603) and later the Directive 2000/29/CE (Real Decreto 58/2005, BOE 19:2583-2665).

Similarly, in Italy the import of fresh fruit (with the exception of grapefruit), live plants and plant parts of citrus have been forbidden since the 1930s by national law (L. 18 June 1931, n. 987). After implementation of the European Directive 77/93/EC (Ministerial Decree D.M. 31 January 1996), the import of citrus fruit was still forbidden as Italy was recognized as protected zone. Only in 1999 was the protected zone status for Italy removed (D.M. 8 July 1999) and since then, the import of citrus fruit from Third Countries has been allowed provided that the requirements of Directive 77/93/CEE and later Directive 2000/29/EC have been met.

3.1.3.2. History of the citrus fruit trade in the citrus producing EU MSs

Citrus fruit trade into the citrus producing EU MS was limited until the 1990s. For example, Italy imported less than 50,000 tonnes of citrus fruit until 1992, with about a five-fold increase over the following decade, mostly due to imports from Spain (Fig. 3). Focusing on imports of citrus fruit from South Africa and Argentina to Italy, it is clear that historically there has been little import of this commodity from these two countries (Fig. 3). It should be also noted that, until 1998, Italian imports of citrus fruit from Third Countries such as Argentina and South Africa were only of grapefruit. The same applies for other extra-European countries where *P. citricarpa* is present, such as Uruguay.

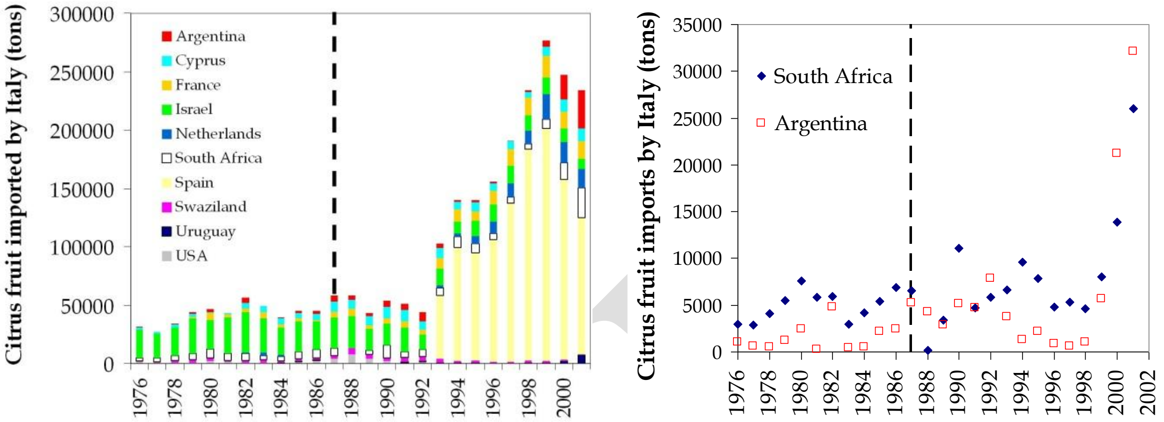


Figure 3: Annual citrus fruit imports by Italy (1976-2001) (left-hand panel) from the ten major exporter countries, and (right-hand panel) from South Africa and Argentina (Eurostat, online). Until 1998, all citrus fruit imports by Italy from South Africa and Argentina were of grapefruit.

It is important to note that trade data for the periods 1976-1987 and 1988-2001 come from two different datasets and so might not be entirely comparable. However, the jump in the imports between 1992 and 1993 in the left-hand panel of Fig. 3 did not coincide with the change-over between the two datasets, but with Spain and Portugal joining the EU.

A similar process can be observed for Spain and Portugal, two other major citrus-growing EU countries (Fig. 4). Spain moved from a situation of absence of citrus fruit imports from Third Countries at the beginning of the 1990s to more than 200,000 tons of citrus fruit imports in 2001 mainly from Argentina, Brazil, Morocco, the Netherlands, South Africa and Uruguay. As far as Portugal is concerned, most citrus fruit imports have traditionally come from Spain (Fig. 4).

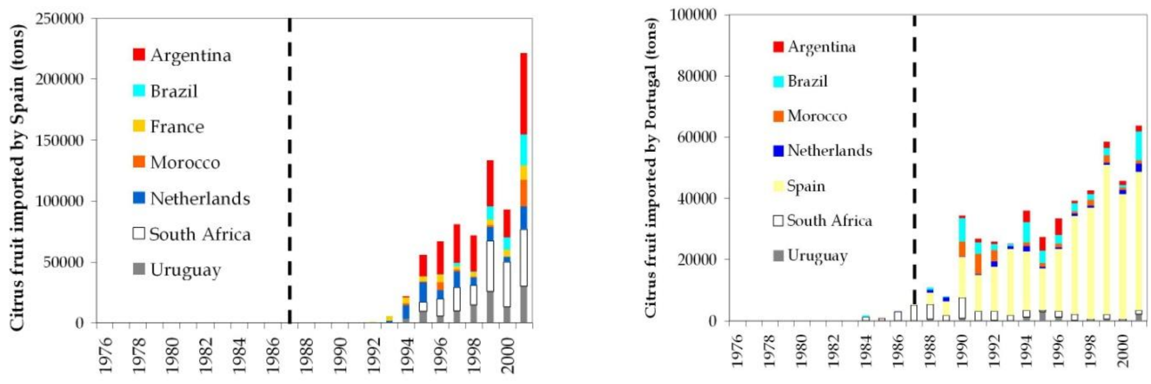


Figure 4: Annual citrus fruit imports from Third Countries (1976-2001) by (left-hand panel) Spain and (right-hand panel) Portugal (Eurostat, online).

Historically, imports of citrus fruit by the major EU citrus-growing countries (Greece, Italy, Portugal, Spain) from the major exporting Third Countries where CBS is present (Argentina, Australia, Brazil, South Africa and Swaziland) were very limited until the mid of the 1990s. Indeed, Spain only started to import citrus fruit from Third Countries in the 1990s (Fig. 5).

Therefore, the argument that European citrus-growing areas are not suitable for the introduction of CBS because there have been plenty of opportunities for introduction due to decades of massive citrus fruit imports into such areas from CBS-affected regions (Kotzé, 2000) is not supported by the trade data. The analysis of historical trade statistics shows that the import of significant amounts of citrus fruit from CBS-affected countries into the EU citrus-growing areas started only recently in the mid 1990s), i.e. after the integration of the Mediterranean countries into the EU. All these imports of citrus fruits were done fulfilling the current European phytosanitary regulations on *P. citricarpa* (section 3.1.3.3), implemented by the Mediterranean countries after their integration of the into the EU.

Structural change during the 1990s in the citrus fruit trade into EU MSs can be observed also in the increase in the number of sources. Again taking Italy as an example, citrus fruit was imported from 15 countries in 1991, but from 32 countries in 2001 (Fig. 6). Similar recent structural changes in the trade of plant commodities have been also documented for other horticultural sectors in Europe (Dehnen-Schmutz et al., 2010).

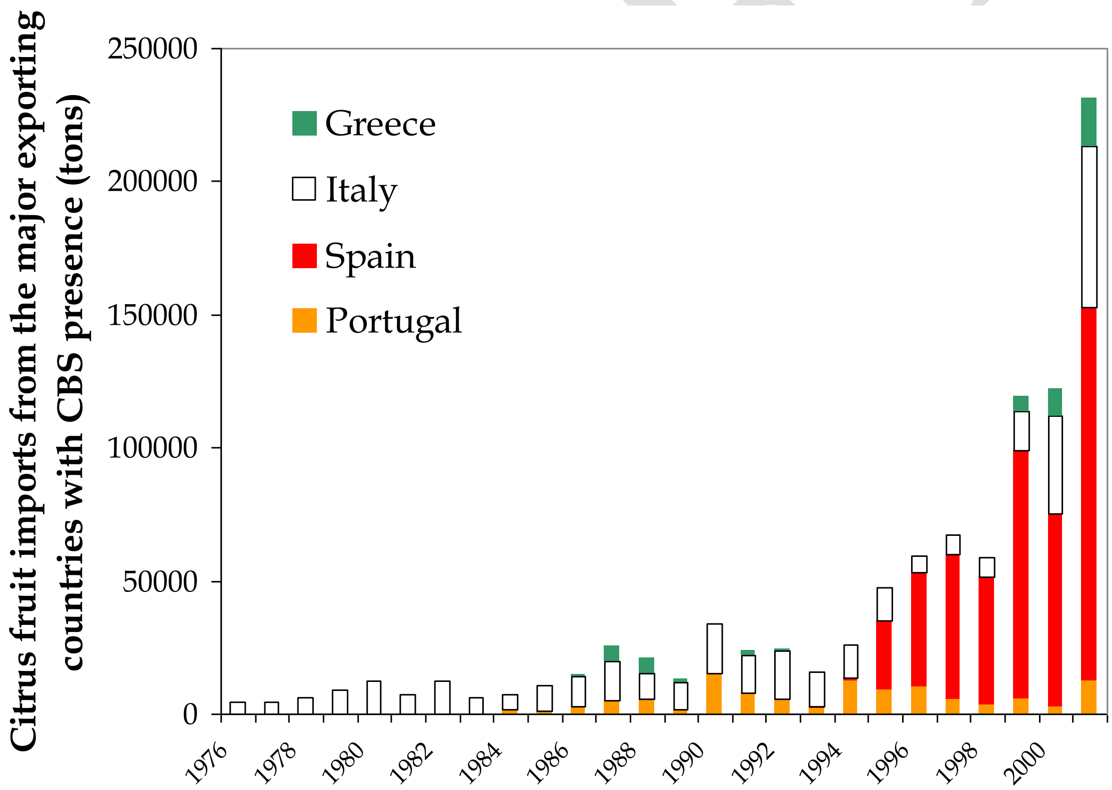


Figure 5: Volume of citrus fruit imported during the period 1976-2001 by the major EU citrus-growing countries (Greece, Italy, Spain and Portugal) from the five major exporters, where CBS is present (Argentina, Australia, Brazil, Swaziland and South Africa) (Eurostat, online). Note that, until 1998, citrus fruit imports by Italy from these Third Countries were exclusively of grapefruit.

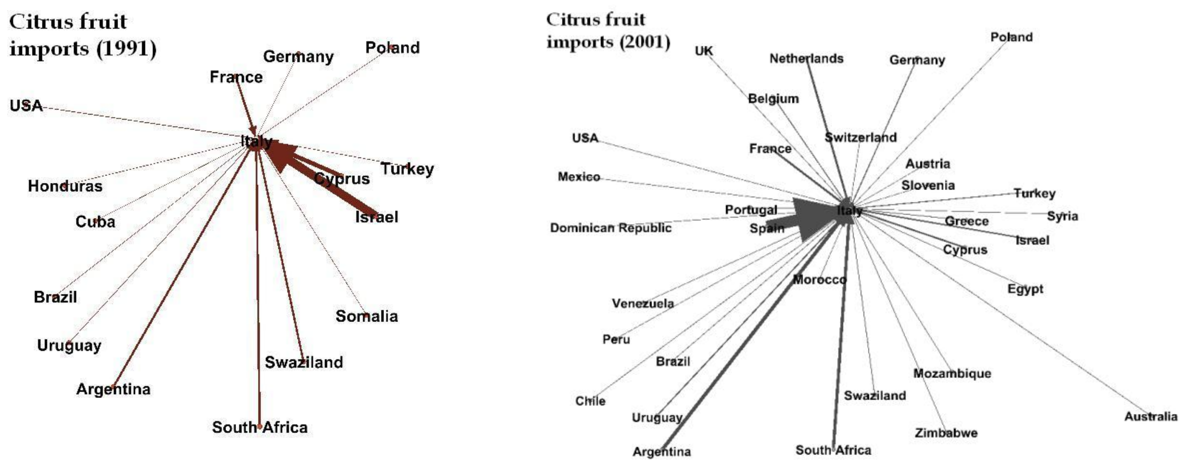


Figure 6: Network of citrus fruit trade imports for Italy in (left-hand panel) 1991 and (right-hand panel) 2001. The imported quantities increased approximately five-fold. Arrows are weighted in relation to the traded volumes. The network does not show the citrus fruit exported or re-exported by Italy to other countries.

3.1.3.3. Current EU regulatory status

Guignardia citricarpa (all strains pathogenic to *Citrus*) is listed in the EU Council Directive 2000/29/EC in Annex II, Part A, Section I. This is the list of organisms harmful to plants and plant products that are not known to occur in the EU and are relevant for the entire EU, whose introduction into, and spread within, all EU MSs is banned if they are present on certain plants or plant products. In particular, *Guignardia citricarpa* (all strains pathogenic to *Citrus*) is banned if present on "plants[9] of *Citrus* L., *Fortunella* Swingle, *Poncirus* Raf., and their hybrids, other than seeds", i.e. if this pathogen is present on living plants or part of plants, as fruit, branches with foliage or flowers, plant tissue culture.

Annex IV, Part A. Section I, paragraph 16.4 of the Council Directive 2000/29/EC describes the special requirements related to *Guignardia citricarpa* for the introduction into the Community of fruits originating in Third Countries of *Citrus* L., *Fortunella* Swingle, *Poncirus* Raf., and their hybrids, other than fruits of *Citrus aurantium* L. According to these requirements, the imported fruit should be accompanied by an official statement that:

- the fruit should originate in a country recognised as being free[10] from *Guignardia citricarpa* Kiely (all strains pathogenic to *Citrus*);

- or the fruit should originate in an area recognised as being free[11] from *Guignardia citricarpa* Kiely (all strains pathogenic to *Citrus*);

- or no symptoms of *Guignardia citricarpa* Kiely (all strains pathogenic to *Citrus*), have been observed in the field of production and in its immediate vicinity since the beginning of the last cycle of vegetation, and none of the fruits harvested in the field of production has shown, in appropriate official examination, symptoms of this organism;

[9] In Art 2 of Council Directive 2000/29/EC, "plants" are considered to mean: living plants and living parts thereof, including seeds; living parts of plants are considered to include: fruit (in the botanical sense, other than preserved by deep freezing), vegetables (other than preserved by deep freezing), tubers, corms, bulbs, rhizomes, cut flowers, branches with foliage, cut trees retaining foliage, plant tissue cultures.

[10] in accordance with the procedure laid down in Article 18 of Council Directive 2000/29/EC.

[11] in accordance with the procedure laid down in Article 18, and mentioned on the certificates referred to in Articles 7 or 8 of this Directive.

- or the fruit originate in a field of production subjected to appropriate treatments against *Guignardia citricarpa* Kiely (all strains pathogenic to *Citrus*), and none of the fruits harvested in the field of production has shown in appropriate official examination, symptoms of this organism.

In addition, temporary emergency measures are in place that impose additional requirements for the import of certain citrus fruits from Brazil in connection with *Guignardia citricarpa* (all strains pathogenic to *Citrus*) (Commission Decision 2004/416/EC; OJ L 151, 30.4.2004, p. 76).

Commission Decision 2006/473/EC (OJ L 187, 8.7.2006, p. 35) lists the Third Countries, as well as certain areas of Third Countries, recognised as being free from *Guignardia citricarpa* (all strains pathogenic to *Citrus*).

In Council Directive 2000/29/EC, other requirements are listed for citrus plants and fruit that are not specific to *P. citricarpa*.

Annex III, Part A, (9) prohibits the introduction of plants of *Citrus* L., *Fortunella* Swingle, *Poncirus* Raf., and their hybrids, other than fruit and seeds from Third Countries into all MSs. This prohibition therefore regards living plants, branches with foliage or cut flowers and plant tissue culture. However, citrus plants for research or breeding programmes can still be introduced by following the conditions listed in Commission Directive 95/44/EC.

Annex IV, Part A, Section I, point 16.1 of , states that fruit of *Citrus* L., *Poncirus* Raf. and *Fortunella* Swingle as well as their hybrids originating in Third Countries shall be free from peduncles and leaves and the packaging shall bear an appropriate origin mark.

- Annex V, Part B, point 3, states that fruits of *Citrus* L., *Poncirus* Raf. and *Fortunella* Swingle and their hybrids originating outside EU shall be subjected to a plant health inspection in the country of origin or the consignor country, before being permitted to enter the Community.

Guignardia citricarpa is in the A1 List of the European and Mediterranean Plant Protection Organization (EPPO, 2013).

3.1.4. Regulatory status in Third Countries

Outside the EU, according to the EPPO PQR database (EPPO, 2013), *P. citricarpa* is in the A1 List of the Caribbean Plant Protection Commission (CPPC) and in the A2 Lists of Asia and Pacific Plant Protection Commission (APPC), Comitè de Sanidad Vegetal del Cono Sur (COSAVE), Interafrican Phytosanitary Council (IAPSC) and Pacific Plant Protection Organisation (PPPO). In America, it is a quarantine pest in the United States and is in the A1 List of Chile, Paraguay and Uruguay. In Asia and Europe, it is in the A1 List in Turkey and is a quarantine pest in Israel and Jordan. In Oceania, it is a quarantine pest in New Zealand

3.1.5. Potential for establishment and spread in the pest risk assessment area

Host plants of *P. citricarpa* are widely grown in orchards of the Southern EU MSs (see Table 6). In a previous scientific opinion (2008), the EFSA PLH Panel did not agree with the conclusion by Paul et al. (2005) that the climate of the EU is unsuitable for the establishment of *P. citricarpa*. Therefore the Panel concludes that there is a potential for establishment and spread in the risk assessment area that should be evaluated.

3.1.6. Potential for consequences in the pest risk assessment area

The pathogen causes different degrees of yield and quality losses in citrus orchards in the area of its current distribution (see Sections 3.1.1.5 and 3.6.1.1). Therefore the Panel concludes that there is a potential for consequences in the risk assessment area that should be evaluated.

3.1.7. Conclusion of pest categorisation

P. citricarpa is absent from the EU and has a potential for establishment and spread and for causing consequences in the risk assessment area. For this reason, a risk assessment for *P. citricarpa* is needed for the EU territory.

3.2. Probability of entry

As stated above (section 2.1.2), the Panel conducted the risk assessment considering the absence of current requirements listed in Annexes II, III, IV and V of Council Directive 2000/29/EC. and in Commission Decisions 2004/416/EC and 2006/473/EC, but under the assumption of a standard citrus disease management in the country of origin to comply with fruit quality standards, However, all the data on imports and interceptions presented in this document were obtained under the regulations currently in place in the EU. These data should be interpreted with caution because quantities of imported products will probably change if the regulations are removed and because interception numbers depend on the procedure of import control currently in place at the EU borders.

3.2.1. Identification of pathways

The Panel identified the following pathways for entry of *P. citricarpa* into the EU:

i. Citrus fruit commercial trade
ii. Tahiti lime fruit (*Citrus latifolia*) commercial trade
iii. Citrus fruit import by passenger traffic
iv. Citrus fruit with leaves and peduncles commercial trade
v. Citrus plants for planting
vi. Tahiti lime (*Citrus latifolia*) plants for planting
vii. Citrus plants for planting import by passenger traffic
viii. Citrus plants and plant parts not for planting, excluding fruit

Seeds have not been considered as a pathway for *P. citricarpa* in this opinion. According to current knowledge, *P. citricarpa* infections are limited to the rind (exocarp and mesocarp) of citrus fruit (Kotzé, 1981). Seeds are located in the internal juice sacs (endocarp), which are not colonized by the pathogen. Seeds could hypothetically be affected by *P. citricarpa* if extensive rotting occurred in harvested fruit but this has not been reported.

Citrus flowers are not known to be infected or colonized by *P. citricarpa*, so they are also not considered as a potential pathway in this opinion. Citrus branches with flowers and/or leaves for ornamental purposes are a theoretical entry pathway, however, as the Panel could not find any information or data regarding such a trade, this pathway is not dealt in this opinion.Ornamental citrus grown in pots are instead included in the pathway V regarding the citrus plants for planting.

Infected citrus twigs are known to be a source of inoculum of *P. citricarpa* (Sposito et al., 2011) but there are no reports of infection or colonization of lignified wood tissues such as large branches. In fact, severe pruning to leave only a framework of branches has been proposed as an alternative eradication method to the removal of entire trees (Whiteside, 1967) (Section 3.4.4). Citrus wood has therefore not been evaluated as a potential pathway for *P. citricarpa* in this opinion.

3.2.2. Entry pathway I: citrus fruit commercial trade

This pathway (graphically illustrated in Fig. 7) concerns the importation of fruit without leaves and peduncles of citrus species from Third Countries where *P. citricarpa* is present (see Table 1), into the EU. All species and varieties of citrus species are considered in this pathway including sweet oranges, mandarines and clementines (*C. reticulata*), lemons, limes (*C. latifolia*, *C. aurantifolia* and *C.*

limettioides), satsumas (*C. unshiu*) and grapefruit (*C. paradisi*). Recent surveys conducted in China indicated that the pomelo (*C. maxima*) is not affected by *P. citricarpa* (Wang et al., 2012) but more data from other regions and proper pathogenicity tests would be needed to completely exclude this citrus species as a potential host (see Section 3.1.1.4 on host range).

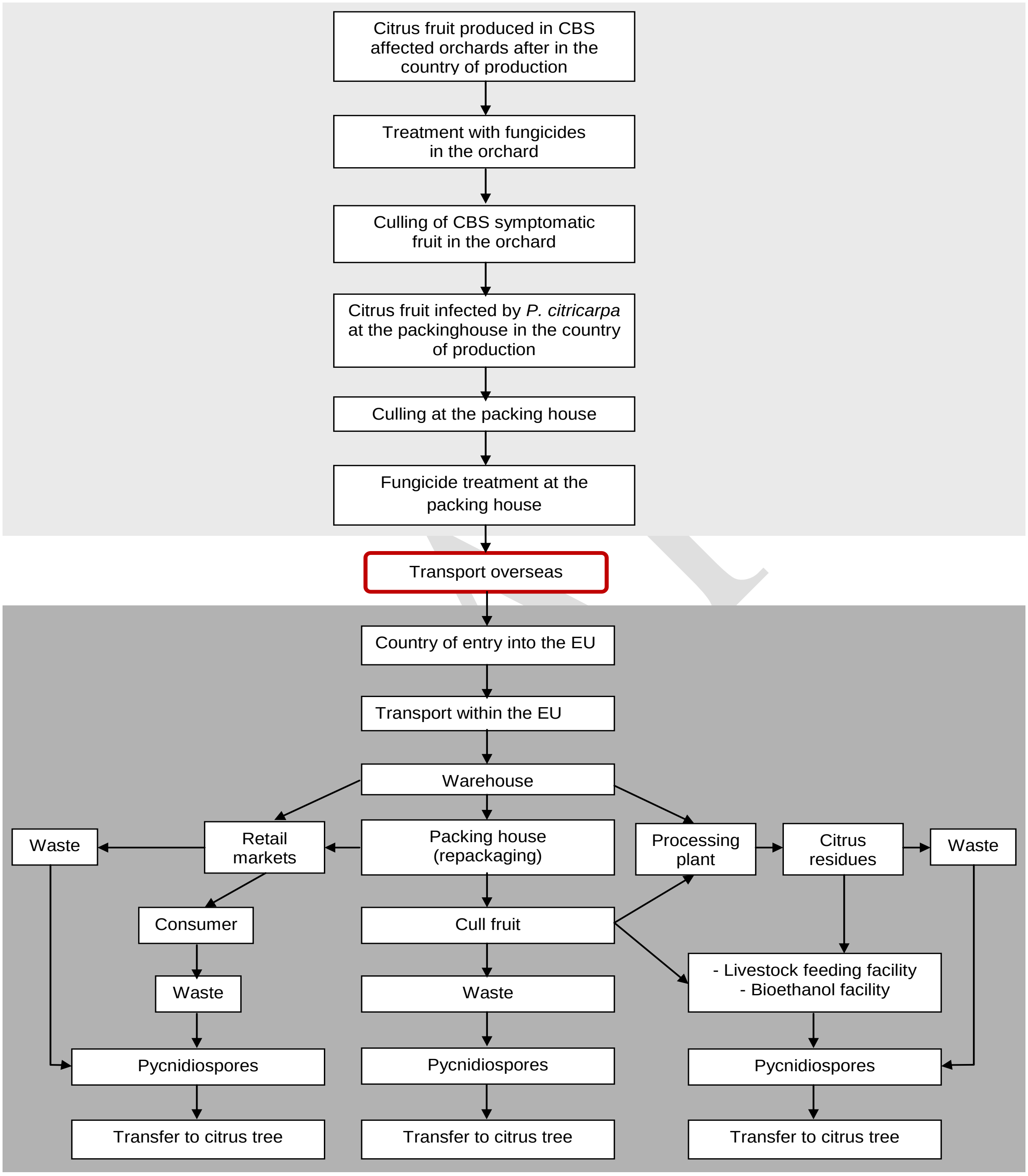


Figure 7: Graphical pathway model illustrating steps in the entry pathway of *Phyllosticta citricarpa* with commercial trade of citrus fruit. The pathway starts in CBS-affected orchards in a country of origin outside the EU and ends with the transfer of spores of the pathogen to a susceptible host within the EU. The scheme is illustrative and departures from the depicted sequence may apply in some countries of origin and certain EU MSs, depending upon local characteristics of citrus production, trade and processing. For instance, in current practice (with the EU legislation in place), there is import inspection, but, in the scenarios considered in this opinion, there is no inspection specifically targeted at CBS.

The pathway of *P. citricarpa* entry with imported citrus fruit has previously been analysed in pest risk assessment documents made by the Republic of South Africa (Hattingh et al., 2000), the Southern Cone Plant Health Committee (COSAVE) (Cortese et al., 2004), the European Food Safety Authority (EFSA Panel on Plant Health (PLH), 2008), the United States Department of Agriculture (USDA APHIS, 2010a) and the EFSA cooperation project on "Pest risk assessment for the European Community plant health: a comparative approach with case studies" (Prima phacie) (MacLeod et al., 2012). The evidence cited in these documents has been considered by the Panel and where there are differences in the conclusions these are discussed below.

The reader should bear in mind that, as stated above, the Panel assessed the probability of entry in the absence of current EU regulations.

Living stages of *P. citricarpa* are frequently found on imported citrus fruit during border inspections at the EU points of entry (see Figure 8). This shows that *P. citricarpa* is associated with the citrus fruit pathway and is able to survive transport and storage as well as existing pest management procedures. During 1999-2012 there were 963 interceptions of *P. citricarpa* on citrus fruit consignments from Third Countries to the EU. There were also three interceptions of *P. citricarpa* on plants for planting (including one bonsai), 67 interceptions of *Phyllosticta* spp. (without identification of the species; mostly from China) and 2 interceptions of *P. citriasiana* (both from China). These interceptions have not been included in the following graphs for the fruit pathway. On average, nearly 70 interceptions were reported per year, with a minimum of 19 (2000) and a maximum of 155 interceptions (2008). Most interceptions were made by the Netherlands (67 %), but approximately 17 % (161) were from Spain, and a few interceptions were made by France, Greece and Portugal, three other EU citrus-growing countries.

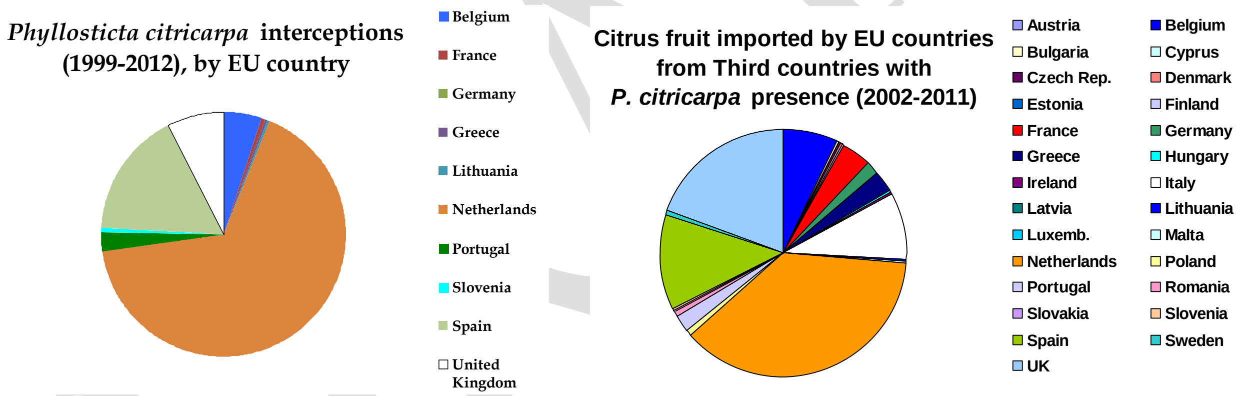


Figure 8: Distribution by EU country of: (left-hand panel) the 961 *Phyllosticta citricarpa* EU interceptions on citrus fruit consignments imported from Third Countries (1999-2012), and (right-hand panel) citrus fruit imported from Third Countries with *P. citricarpa* presence (2002-2011).

For the 11 EU MSs which intercepted *P. citricarpa* over the period 1999-2012, there is a strong correlation between the number of *P. citricarpa* interceptions reported by the EU MS and the volume of citrus fruit imported by the same MS from Third Countries with reported presence of *P. citricarpa* (Fig. 9).

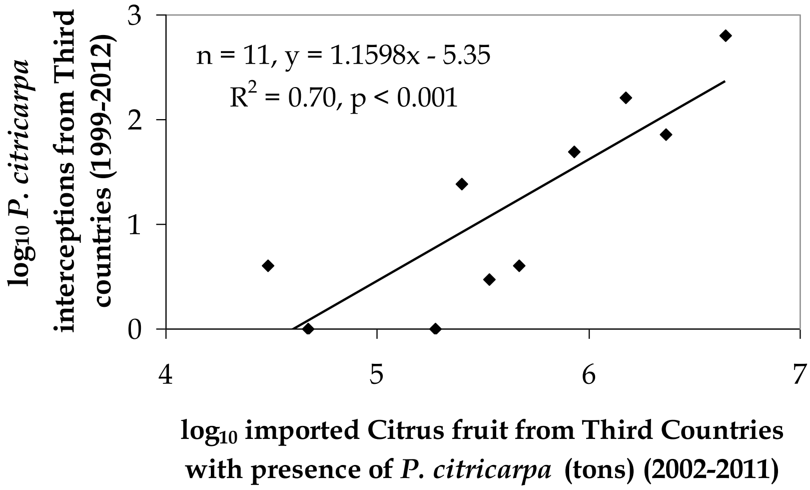


Figure 9: Log-log correlation of number of *Phyllosticta citricarpa* interceptions made by EU MSs (1999-2012) and imported volumes of citrus fruit from Third Countries with reports of *P. citricarpa* (2002-2011), for the 11 EU MSs which intercepted *P. citricarpa* at their borders.

3.2.2.1. Probability of association with the pathway at origin

The association with the citrus pathway varies with the citrus species: lemons and late-maturing sweet orange cultivars are generally considered to be more susceptible (Kotzé, 1981), mostly because they hang on the tree for a longer period and are therefore more exposed to inoculum and environmental factors and have more time for symptom development. Early-maturing sweet orange cultivars are considered less susceptible as they are harvested earlier (Timmer, 1999; Sposito et al., 2004; Sousa and de Goes, 2010).

Most (approximately 75%) *P. citricarpa* interceptions on citrus fruit consignments imported into the EU from Third Countries were made on shipments of sweet orange. About 7 % (70) interceptions were made on shipments of lemon (Fig. 10), the citrus species most susceptible to *P. citricarpa*, of which more than half (43) originated from South Africa.

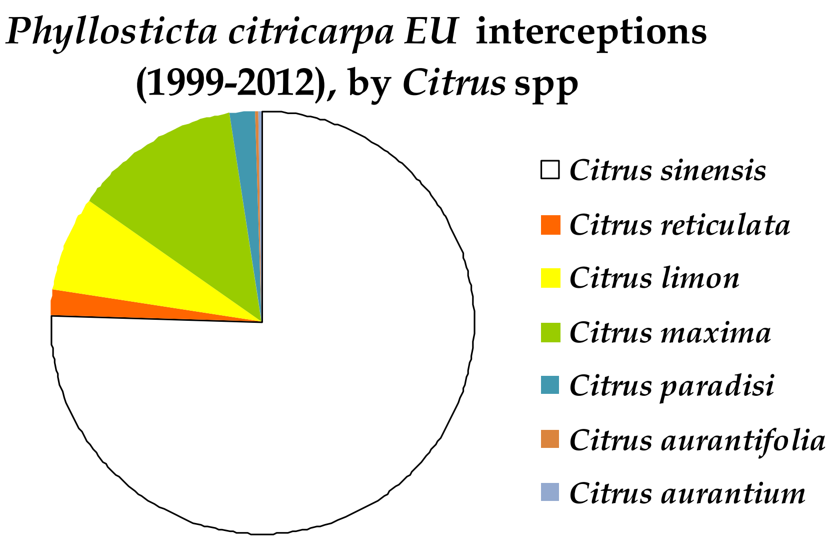


Figure 10: Distribution by citrus species of the 961 *Phyllostica citricarpa* EU interceptions on citrus fruit consignments imported from Third Countries between 1999 and 2012.

In the countries where *P. citricarpa* is present, fungicide treatments and some cultural practices are currently applied for the management of CBS (Kotzé, 1981; Timmer, 1999, Miranda-Bellote et al., 2013). However, in Brazil, cultural practices such as the early harvest of symptomatic citrus fruit and the removal of leaf litter from the orchard floor have been shown incapable of reducing CBS incidence and severity to satisfactory levels (Spósito et al., 2011). Pre-harvest applications of fungicides reduce CBS incidence or delay symptom development in citrus fruit in storage, but they do not seem to eradicate quiescent infections completely (Seberry et al., 1967; Andrade et al., 2001; Agostini et al., 2006). A meta-analysis of available data from fungicide trials against CBS (see section 3.6.1.) shows that fungicide treatments are unable to reduce the level of infection of citrus fruit to negligible levels if disease pressure is high, as is usually the case in orchards in which fungicide trials are done.

The efficacy of culling fruit in the field and/or in the packinghouse is limited due to the presence of latent infections in asymptomatic fruit that may develop symptoms after harvest during transport and storage (Kotzé, 1981; Agostini et al., 2006; Baldassari et al., 2007). In addition, symptoms on fruit are variable and unspecific, with the exception of hard spot with pycnidia. Some lesions are very small (1 to 3 mm, in diameter) and may therefore be confused with those caused by other citrus pathogens, as well as by mechanical or insect damage (Snowdon, 1990; Kotzé, 2000).

Low storage temperatures (8 °C), waxing or hot water treatments of fruit may reduce or delay the post-harvest development of CBS symptoms, but they are unlikely to eliminate the pathogen (Seberry et al., 1967; Korf et al., 2001; Agostini et al., 2006). Agostini et al. (2006) also showed that post-harvest fungicide dips and waxes were ineffective in controlling CBS. The same authors also reported that once quiescent infections are present in the fruit, it appears to be difficult to prevent the development of CBS symptoms after harvest. Washing and brushing of fruit are procedures approved by APHIS for citrus packinghouses located in CBS quarantined areas in Florida (APHIS, 2012). Although these measures will most probably remove any pycnidiospores present on its surface, they are unlikely to affect the latent mycelium inside the fruit peel or the pycnidiospores embedded within pycnidia.

In conclusion, cultural practices, pre- and/or post-harvest treatments applied in the current area of *P. citricarpa* distribution may reduce the incidence and severity of CBS-infected citrus fruit imported into the PRA area, but they will not completely eliminate the pathogen.

Based on the above, and in agreement with MacLeod et al. (2012), the probability of association with the pathway at origin for *P. citricarpa* on fresh citrus fruit imported from infested areas into the PRA area is assessed as likely, with a **medium** uncertainty.

Volume of the movement along the pathway

There is a high volume of citrus fruit imported every year into the EU from Third Countries where *P. citricarpa* is present. Data for each exporting Third Country for the period 2002-2011 are shown in the Appendix. The main exporters of citrus fruit into the EU are: Argentina, Brazil, China, the United States, Uruguay, South Africa and Zimbabwe. Minor imports originate from Australia, Cuba, Ghana, Mozambique and New Zealand. Very small quantities of citrus fruit have been imported into the EU from Kenya, the Philippines, Taiwan, Uganda and Zambia.

Most EU interceptions of *P. citricarpa* on citrus fruit consignments imported from Third Countries over the period 1999-2012 originated from Brazil and South Africa (Fig. 11). The number of countries from which interceptions originated (16) provides evidence that citrus fruit can be considered as a major potential pathway of entry for the pathogen.

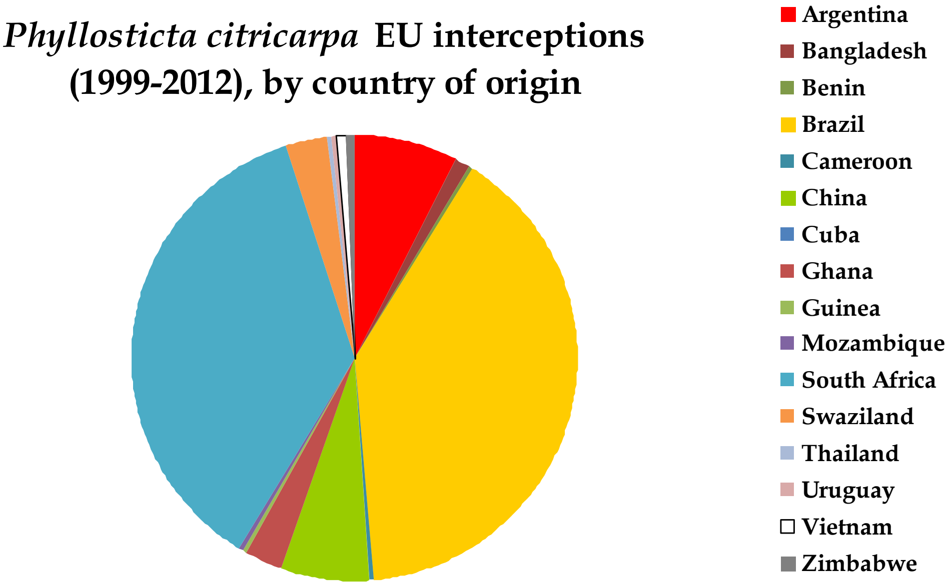


Figure 11: Distribution by country of origin of the 961 *Phyllostica citricarpa* EU interceptions on citrus fruit consignments imported from Third Countries between 1999 and 2012.

Based on the above, and in agreement with MacLeod et al. (2012), the volume of citrus fruit imported into the PRA area from Third Countries where the pest is present is **massive**, with **low** uncertainty.

Frequency of the movement along the pathway

The frequency of imports of citrus fruit into the PRA area varies between different years, citrus species, exporting countries and the importing EU MSs (Eurostat, online). Generally, citrus fruit consignments from Third Countries where *P. citricarpa* is present are imported into the EU throughout the whole year, with the main import period between March and November (Eurostat, online) with the volumes decreasing in relation to the beginning of the EU harvest season (MacLeod et al., 2012).

Most *P. citricarpa* interceptions on citrus fruit consignments imported from Third Countries into the EU were made during the late summer and autumn in Europe, mainly in September and October, as exemplified by data from South Africa, Brazil and China (all years, all receiving EU countries, all *Citrus* spp.) (Fig. 12). This timing has implications for the probability of transfer of the pathogen to a suitable host (see below), which would be much lower if affected consignments were imported during the European winter, particularly for shipments going directly to EU citrus-growing countries, but also in case of re-exported consignments from, e.g. the Netherlands to Spain and other Mediterranean EU countries.

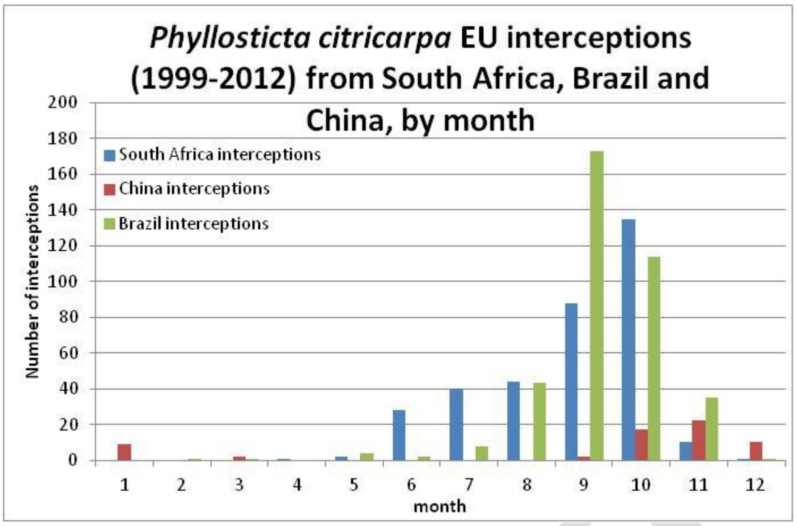


Figure 12: Distribution by month of the *Phyllosticta citricarpa* EU interceptions on citrus fruit consignments imported from South Africa, Brazil and China between 1999 and 2012.

The seasonality in imports appears to be consistent across different years, as shown by the relatively small error bars in the frequency distribution of imports of sweet oranges from the three major exporting Third Countries (South Africa, Brazil and Argentina) to the EU (these monthly data do not include Croatia) over the period 2002-2011 (Fig. 13). A similar pattern is observed for mandarins. The pattern is different for grapefruit, because two of the major exporters into the EU (the USA and China) are located in the northern hemisphere. The major exporter of lemons into the EU is Argentina, but the seasonality of lemon imports into the EU from the three major exporters (South Africa, Brazil and Argentina) is similar to that for sweet oranges (Fig. 13).

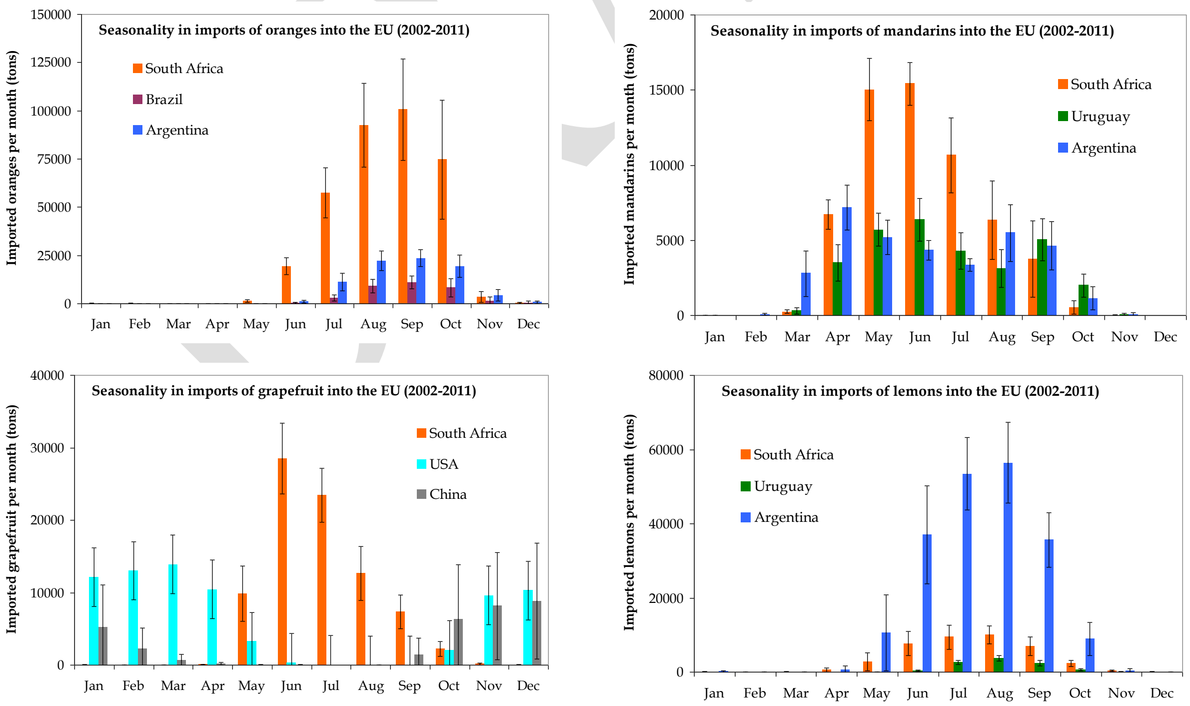


Figure 13: Quantity of sweet oranges, mandarins, grapefruit and lemons imported monthly into the EU-27 MSs from South Africa, Argentina and Brazil, the three major exporters of oranges into the EU, during the period 2002-2011 (Eurostat, online). Error bars are standard deviations.

In agreement with MacLeod et al. (2012), and based on the data above, citrus fruit is imported **very often** into the PRA area from Third Countries where the pest is present, with **medium** uncertainty due to the seasonal variation in imports (mainly from May to November).

Based on the above ratings, the Panel concludes that, overall, the probability of association of *P. citricarpa* with the commercial fruit pathway at origin is rated as likely, with **medium** uncertainty mainly due to different incidence and severity of CBS in affected citrus fruit from different locations and years and to the difficulties in ensuring that fruit is disease-free if it originates from countries where the disease is endemic, due to the limited efficacy of fungicides, as indicated by the meta-analysis of control trials presented later in section 3.6.1 of this opinion.

3.2.2.2. Probability of survival during transport or storage

In general, the transport of commercial citrus fruit takes place under cool conditions (Wills et al., 1998). Whilst sweet oranges and mandarins are typically shipped at 1 °C and 4 °C, respectively, lemons and limes are usually shipped at 10ºC, because of their sensitivity to chilling injury. Depending on the time of the year, the conditions of harvested trees, and fruit conditions, grapefruit is shipped at 10 or 15 ºC (Wardowski, 1981). Such low temperatures during transport and storage are likely to prolong the survival of *P. citricarpa* pycnidia and pycnidiospores on CBS lesions. Mature pycnidiospores of *P. citricarpa* produced on infected citrus fruit were still shown to be viable after three weeks storage of the fruit at 4.5 or 10 ºC, but apparently lost their viability at 25 ºC (Korf et al., 2001). Similarly, freshly exuded pycnidiospores of *P. citricarpa* incubated at 25 ºC decreased their viability between 60 to 100% after four days and three months, respectively (Kiely, 1948).

In addition, the survival of latent *P. citricarpa* mycelium is not affected by the low temperatures typical used to transport and store citrus fruits: CBS symptoms develop rapidly when fruit with quiescent infections encounters higher temperatures (Kotzé, 1981; Agostini et al., 2006; Baldassari et al., 2007). The isolation of *P. citricarpa* was successful from CBS lesions in citrus fruit kept for more than 40 days under various moisture conditions at 8 ºC or ambient temperatures of 15-25ºC (Agostini et al., 2006). More than 85 % positive isolations of *P. citricarpa* where also obtained from CBS lesions in sweet orange fruit after three weeks maintained at 4.5 °C, 10 ºC and 25 ºC (Korf et al., 2001). These findings imply that the pathogen is likely to remain viable long after fruit stored in such conditions have become unmarketable. This has important implications for the likelihood of transfer to suitable hosts (see below).

Based on the above, and in agreement with MacLeod et al. (2012), the Panel concludes that, in terms of duration and conditions of transport and storage, *P. citricarpa* in the form of (i) pycnidiospores within pycnidia in fruit lesions and/or (ii) latent mycelium present in asymptomatic fruit, is **very likely** to survive transport and storage conditions (with **low** uncertainty).

In 2008/2009, citrus fruit imported by the EU from, Argentina, Brazil, China, South Africa and Uruguay by sea was three orders of magnitude greater than imports by air (MacLeod et al., 2012). Little information is available on the time required for citrus fruit to be shipped from other continents to Europe (three weeks or longer are reported for shipments from South Africa (Terblanche, 1999)). Although they would be very valuable, no data are available on the incidence and severity of CBS on citrus fruit consignments (proportion of infected fruit and number of lesions per fruit) imported by EU countries, because the consignments with CBS-affected fruit are rejected without evaluating further.

Likelihood of the pest multiplying/increasing in prevalence during transport/storage

Since the optimal temperature for the hyphal growth of *P. citricarpa* in synthetic medium is 25-28 ºC (Chiu, 1955; Kotzé, 1981) and the pathogen remains virtually inactive at temperatures lower than 15 °C (Chiu, 1955), in agreement with MacLeod et al. (2012), it is **very unlikely** (with **low** uncertainty) that the pathogen will multiply or increase in prevalence during transport/storage of infected citrus fruit, which normally occurs at low temperatures.

3.2.2.3. Probability of surviving existing pest management procedures

The management of CBS in its current area of distribution (cultural practices and chemical treatments applied pre- and post-harvest) can reduce the level of disease in the orchard or delay symptom development in transit and storage but does not eliminate the pathogen, particularly quiescent infections on citrus fruit (Kotzé, 1981). Similarly, physical treatments of citrus fruit in packinghouses can reduce or delay the post-harvest development of CBS symptoms but without eliminating the pathogen (Seberry et al., 1967; Korf et al., 2001; Agostini et al., 2006).

The detection of the pathogen is made difficult by the long incubation period (2 to 12 months) during which latently infected fruit remains asymptomatic (McOnie, 1967; Kellerman and Kotzé, 1977; Kotzé, 1981; Aguiar et al., 2012). Culling will thus not detect the latently infected fruit, which will also escape any potential border inspection.

Given their variability, CBS symptoms on fruit can be confused with those caused by other citrus pathogens, mechanical or insect damage (Snowdon, 1990; Kotzé, 2000), although living stages of *P. citricarpa* continue to be intercepted on citrus fruit consignments imported into the EU.

Reliable detection and identification of the organism on citrus fruit can be made only after laboratory testing (EPPO, 2003).

Based on the above, and in agreement with MacLeod et al. (2012), the Panel concludes that *P. citricarpa* is **very likely** (with **low** uncertainty) to survive and remain undetected during existing pest management procedures, particularly on latently infected (asymptomatic) fruit and fruit with low disease incidence and severity.

3.2.2.4. Probability of transfer to a suitable host

Large quantities of citrus fruit are imported every year from CBS-infested Third Countries into all the EU MSs, including the citrus-growing EU MSs (i.e. Spain, Italy, Greece, Cyprus, France and Portugal) (Tables 20 in Appendix B) (Eurostat, online). In addition, some EU MSs (e.g. Belgium and The Netherlands) re-distribute within the EU large quantities of fresh citrus fruits imported from CBS-infested countries (see sections 3.4.2 and 3.4.3) .

As an example, in 2009 the Netherlands imported approximately 450,000 tons of sweet orange from various CBS-infested countries (including Argentina, Brazil and Uruguay) and re-distributed almost 200,000 tons of sweet orange to other EU MSs, including citrus-producing EU MSs (Eurostat, 2008). Fresh citrus fruit are destined for human consumption or processing. Thus, once fruit consignments enter the PRA area, they are sent to packing houses, processing plants, wholesale and retail fresh fruit markets before being sold to the end users.

Therefore, and in agreement with MacLeod et al. (2012), it is expected that the imported citrus fruit will be **very widely** distributed within the PRA area (with **low** uncertainty).

The main period of import of citrus fruit from Third Countries is between March and November (Fig. 13), when there is little availability of European fresh citrus fruit (Agustí, 2012). Examples of the seasonality of imports are shown in Figure 13. A varying, but significant proportion of citrus fruit imports to the EU takes place during April-May and September-October, the period when weather conditions are potentially favourable for infection (EFSA PLH Panel, 2008). Moreover, *P. citricarpa* interceptions in imported citrus fruit are mainly concentrated in late summer and autumn (Section 3.2.2.1, Fig. 12).

Based on the above, citrus fruit consignments imported into the PRA area from Third Countries with presence of the pest are **likely** to arrive during a time of the year potentially suitable for disease establishment, with **low** uncertainty.

Most of the citrus fruit consignments imported into the pest risk assessment area arrive at ports because they are transported by sea (Eurostat, online; Europhyt, online). Citrus species susceptible to CBS are widely grown in the southern EU MSs in a variety of locations (commercial orchards, nurseries, smallholdings, private gardens for family consumption, public gardens and along the roadsides both in urban and rural regions). Commercial citrus orchards and nurseries are located mainly in coastal areas, next to rivers (Agustí, 2012) and in some cases in close proximity to ports. Given that fresh citrus fruit imports are destined for human consumption and processing, they will be very widely distributed to packing houses, processing plants and fresh fruit markets in urban and rural regions of all EU MSs.

Most of the sweet oranges imported by the EU from South Africa go to non citrus-producing countries, but the quantities of South African sweet oranges imported by citrus-producing EU countries are not negligible (Table 20 in Appendix B). Interestingly, mandarins from South Africa go nearly exclusively to non citrus-producing EU countries. Also grapefruit from South Africa is mostly imported by non-citrus producing EU countries. Most lemons exported by South Africa to the EU go to non citrus-producing countries, but the quantities imported by citrus-producing EU countries are not negligible (Table 20 in Appendix B).

The risk of pathogen transfer associated with citrus fruit imported into the PRA area from CBS-infested Third Countries is mainly due to discarded unmarketable whole fruit or peel produced by packing houses, processing plants, fresh fruit markets, households, etc. and their subsequent management. Packinghouses and processing plants are usually located within the citrus-growing regions of the PRA area and are often in close proximity and even adjacent to commercial citrus orchards (EFSA PLH Panel, 2008; NPPO of Italy, 2010, cited by Mc Leod et al., 2012).

Citrus pulp is the residue generated by pressing fresh citrus fruit for juice extraction. During this process, 45 to 60% of their weight remains in the form of peel, rag and seeds. Fresh citrus pulp is characterized by a high moisture content which favours microbial degradation (Cerisuelo et al., 2010). For this reason, ensiled or dried citrus pulp residue is the citrus by-product that is most extensively used for livestock/animal feeding (Bampidis and Robinson, 2006; Caparra et al., 2007; NPPO of Italy, 2010, cited by Mc Leod et al., 2012). Whole marketable and non-marketable citrus fruit can also be withdrawn from the market and turned into citrus waste (Piquer et al., 2009b) either because they do not meet the requirements for fresh produce (2 %; unmarketable fruits) or because they have been withdrawn from the market in order to maintain prices (Piquer et al., 2009a). A maximum of 5% commercialized fruits is withdrawn from the market (EU Regulation- 2200/96). Boluda-Aguilar et al. (2010) estimated that 1.5 million tonnes of citrus waste are produced each year in the Mediterranean Basin. The average yearly production of citrus peel wastes in Spain and Italy was estimated to be about 500,000 tonnes (Caparra et al, 2007; Boluda-Aguilar et al., 2010). Citrus waste is also used in ethanol production facilities in the EU to obtain biofuel, together with other co-products such as limonene, galacturonic acid and pectin (Boluda-Aguilar et al., 2010; NPPO of Italy, 2010, cited by Mc Leod et al., 2012; Lanfranchi, 2012). Citrus waste from fruit markets or households may also be discharged in the vicinity of citrus trees either where landfills are located close to commercial or abandoned citrus orchards, or where citrus waste are discharged uncontrolled in the vicinity of citrus trees.

Symptomatic citrus fruit and peel can be a source of *P. citricarpa* pycnidiospores, which may remain viable for a relatively long time (Korf et al., 2001; Agostini et al., 2006). Therefore, if symptomatic citrus fruit, fruit peel or other citrus by-products with pycnidia are disposed close to host plants (grown in nurseries, commercial orchards, private and public gardens, roadsides, etc.) in the risk assessment area (Fig. 14), the mature pycnidiospores exuded from pycnidia under wet conditions could be splash-dispersed by rain (Whiteside, 1967; Spósito et al., 2011) onto the lower parts of the canopy infecting leaves, twigs and fruit at a susceptible stage. This splash-dispersal of pycnidiospores could potentially take place during rain events at the beginning (April-May) and the end (September-October) of the main period of import of citrus fruit into the citrus-growing regions of the risk assessment area. Rain events in the citrus-growing areas of the EU are common in those months (EFSA PLH Panel, 2008).

Moreover, water drops formed on leaves due to fog, mist or dew occurring during the night in the coastal citrus-growing regions and irrigation (overhead or micro-sprinkler) applied during the dry periods may cause drip-splash of *P. citricarpa* pycnidiospores produced on infected fruit/peel discarded near to citrus plants in the PRA area. Drip-splash can be as efficient as direct rain-splash for the dispersal of mucilaginous conidia (Fitt et al., 1989).

Studies conducted in South Africa indicated that pycnidispores of *P. citricarpa* from pure cultures, symptomatic CBS sweet orange fruit and peelings were not able to colonize lemon leaf litter on the orchard floor (Truter et al., 2007). However, susceptible live tissues (leaves, twigs and fruit on the canopy) of citrus trees in commercial orchards in the risk assessment area are normally very close to the soil, and leaves and fruit very often directly touch the orchard floor (see Fig. 14 below and Section 3.3.3.3), depending on the cultivation technique and the season. Branches bearing many mature fruits tend to bend closer to the orchard floor before harvest. In addition, during the last few years, there is a trend towards the cultivation of shorter citrus trees by grafting onto dwarfing rootstocks (section 3.3.3.3).
Preliminary results from ongoing experiments on splash dispersal of *P. citricarpa* pycnidiospores from artificially infected sweet orange fruit, show in completely still air conditions a maxiumum height of splashed droplets of 47.4 cm, which was reached 20 cm horizontally from the orange, and a maximum horizontal distance splashed of 50 cm. Further work is in progress to investigate splash dispersal in combination with different wind speeds (Perryman and Jones, 2013). These preliminary results are in accordance with the knowledge that conidia covered with mucilage are splash-dispersed up to a height of not more than 50 cm and less than 1 m from the inoculum source with their numbers decreasing steeply with increasing height or distance (Fitt et al., 1989). Pycnidiospores of *P. citricarpa* present on citrus fruit/peel discarded close to citrus trees can thus be splash-dispersed by rain or irrigation water to infect young leaves, twigs and fruit in the lower parts of the citrus tree canopy.

Under windy conditions, the small *P. citricarpa* pycnidiospores (9.4-12.7 x 5-8.5 μm) (Baayen et al., 2002) carried in small splash drops can potentially become windborne as an aerosol of fine spray, spreading *P. citricarpa* pycnidiospores over longer distances. Although not yet investigated, *P. citricarpa* pycnidiospores produced on discarded CBS-affected fruit/peels might be transported by insects, birds, and other organisms and deposited on susceptible hosts grown at a considerable distance (Kiely, 1948; MacLeod et al., 2012).

Based on the above together with the information provided by MacLeod et al. (2012), the Panel considers that citrus fruit imported into the EU from infested Third Countries are very widely distributed in both citrus-growing and non-citrus-growing EU MSs. If CBS-affected citrus fruit, peels or other citrus by-products with pycnidia of *P. citricarpa* are discarded underneath or in close proximity to susceptible citrus trees, the pathogen can be dispersed by natural means to infect susceptible plant tissues.

In agreement with MacLeod et al. (2012) the Panel concludes that the pest is **moderately likely** to transfer from the fruit pathway to a suitable host or habitat, with a **medium** level of uncertainty.

The uncertainties are associated with the frequency and quantity of infected fruit/peel being discarded in close enough proximity to a host in the citrus-growing regions of the risk assessment area, and the time taken for discarded asymptomatic whole fruit or peel to produce pycnidiospores before their decomposition (MacLeod et al., 2012).

Figure 14: a), b) Processing of citrus pulp residues and whole citrus fruit in close proximity to citrus orchards; c) uncontrolled citrus waste discharged in the vicinity of neglected citrus trees; and d) sweet orange orchard with low hanging branches and fruit (Valencia, Spain).

The import of fresh citrus fruit into the PRA area occurs mainly during the European late spring, summer and early autumn period, when there is little if any local production (Agustí, 2012). Because citrus fruit is imported for processing and direct human consumption, it is expected that the commodity will be widely distributed in both urban and rural areas of the EU, both in citrus- and non-citrus-growing regions. The risk of transfer to a suitable host posed by citrus fruit imported from CBS-infested Third Countries is associated with the discarded fruit, peels, pulp or other citrus fruit by-products derived from packing houses, processing plants, households and fresh fruit markets.

If citrus fruit by-products are discarded in the vicinity of citrus nurseries, commercial or abandoned citrus orchards, and susceptible citrus trees grown in private and public gardens and roadsides, the pathogen is likely to be transferred by natural means (rain or irrigation water, insects, birds, etc.) and infect susceptible plant tissues (leaves, twigs and fruit).

Based on the above, and in agreement with MacLeod et al. (2012), the intended use of the citrus fruit commodity is **moderately likely** to aid transfer of the pathogen to a suitable host, with **medium** uncertainty.

There are uncertainties concerning (i) the prevalence of *P. citricarpa* on infected citrus fruit imported into the pest risk assessment area, (ii) the frequency and quantity of infected citrus fruit by-products being discarded in close proximity to a host in the citrus-growing regions of the PRA area, and (iii) the time taken for discarded asymptomatic whole fruit or peel to produce pycnidiospores before decomposition by other organisms (MacLeod et al., 2012).

Based on these ratings, the Panel concludes that the transfer of *P. citricarpa* to a suitable host through the commercial fruit pathway is **moderately likely**, with **medium** uncertainty that is mainly due to the gaps in our knowledge listed above.

3.2.3. Entry pathway II: Tahiti lime fruit (*Citrus latifolia*) commercial trade (without leaves and peduncles)

A graphical pathway model illustrating the entry pathway of *P. citricarpa* with the commercial trade of Tahiti lime (*Citrus latifolia*) fruit, without leaves and peduncles, is shown in figure 15.

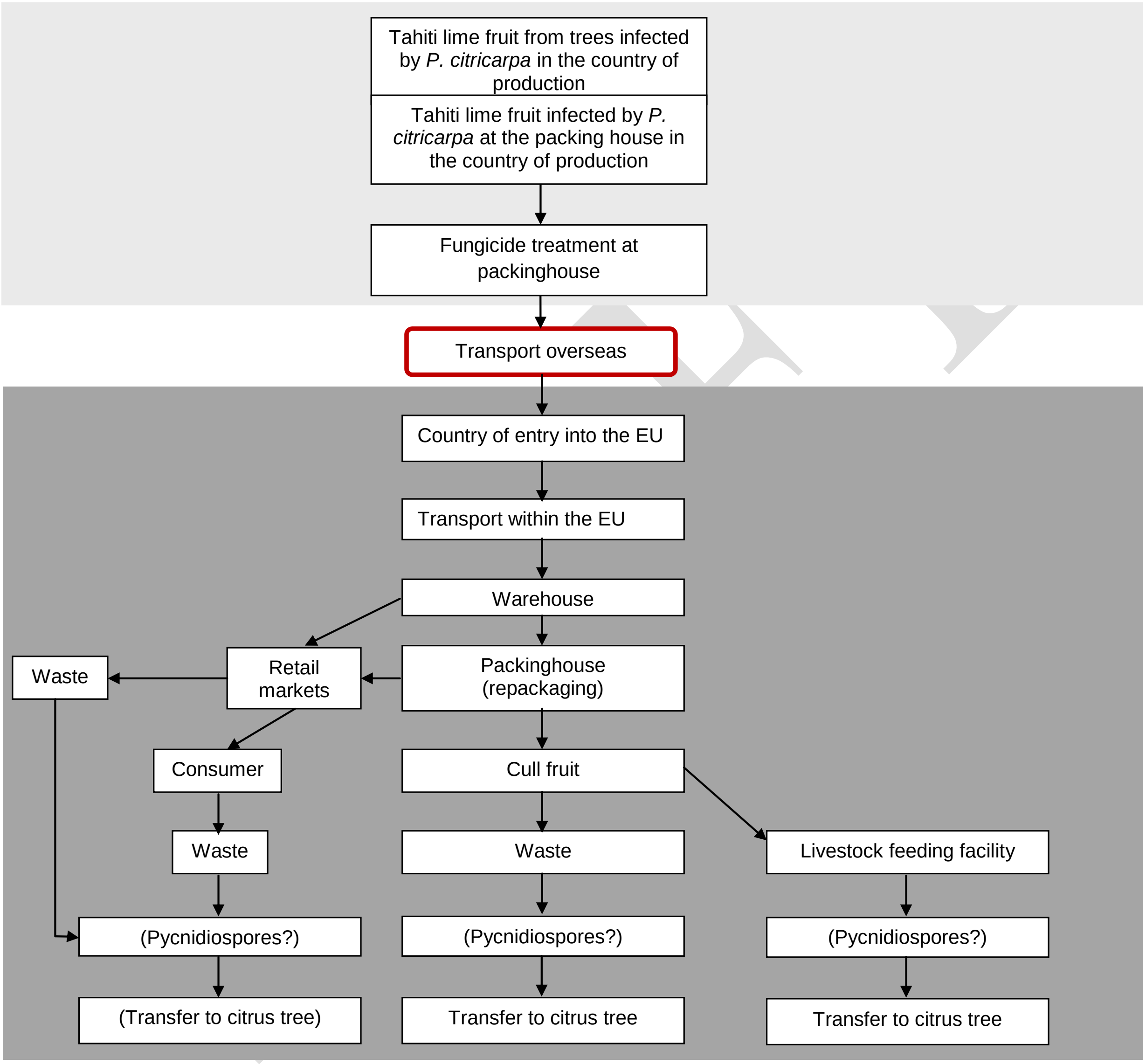


Figure 15: A graphical pathway model (pathway II) illustrating steps in the entry pathway of *Phyllosticta citricarpa* for the commercial trade in Tahiti lime (*Citrus latifolia*) fruit, without leaves and peduncles. The pathway starts in infested orchards in a country of origin outside the EU and ends with the transfer of spores of the pathogen to a host within the EU. The scheme is illustrative and departures from the depicted sequence may apply in some countries of origin and certain MSs of the EU depending upon the local characteristics of citrus production, trade, and processing. For instance, currently there is import inspection but in the scenarios considered in this opinion, there is no inspection specifically for CBS.

3.2.3.1. Probability of association with the pathway at origin

Although confirmatory long term and area-wide field surveys are not available, Tahiti lime (*Citrus latifolia*) fruit are reported not to develop CBS symptoms under field conditions in Brazil, even in areas with high inoculum pressure by *P. citricarpa* (Baldassari et al., 2008). However, in this study conducted in Conchal (Sao Paulo), two out of the 11 *Phyllosticta* isolates obtained from peel of fruit of Tahiti lime were identified as *P. citricarpa* and induced CBS symptoms when inoculated in sweet orange fruit. The study did not include inoculations in Tahiti lime fruit.

The major exporter of lime fruit into the EU is Brazil, with limited seasonality in the trade (Fig. 16). A total of ~ 435,000, ~ 2,600, and ~ 1,600 tonnes of fruits of Tahiti lime from Brazil, Argentina and South Africa, respectively, were imported into the EU territory between 2002 and 2011. Although *P. citricarpa* is present in these countries, no interceptions on Tahiti acid lime have been recorded in EU border inspections, confirming that no symptoms of CBS were detected on imported Tahiti lime fruit. However, since *P. citricarpa* is able to colonize Tahiti lime fruit under natural conditions, the probability of association of the pathogen with the pathway at origin is rated as **likely** with **high** uncertainty due to the limited amount of evidence available.

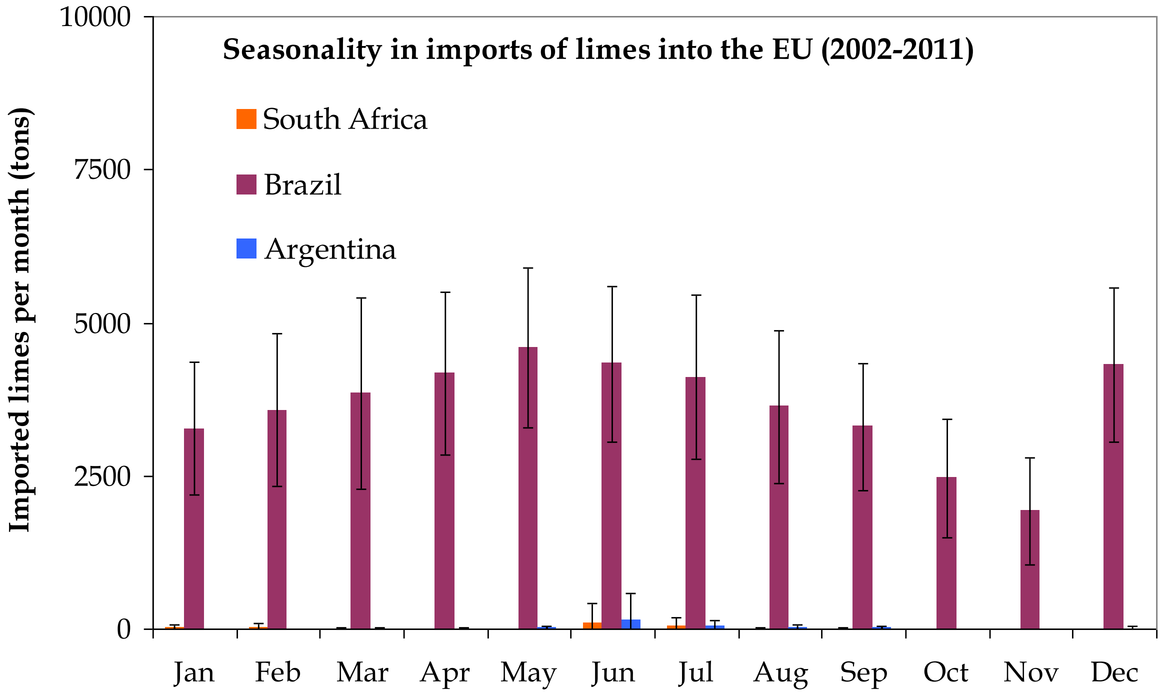


Figure 16: Seasonality in imports of limes into the EU from the three major exporting Third Countries (Brazil, South Africa and Argentina, 2002-2011). Error bars are standard deviations.

3.2.3.2. Probability of survival during transport or storage

Studies evaluating the survival of *P. citricarpa* in Tahiti lime fruit are not available. However, the same considerations made about the likely survival during transport or storage of *P. citricarpa* as latent mycelia present in asymptomatic fruits of other citrus species may also be applicable to Tahiti lime.

Therefore, the Panel considers that the probability of survival of the pathogen during transport or storage of Tahiti lime fruit is rated as **very likely**, with **high** uncertainty due to the lack of evidence.

3.2.3.3. Probability of survival to existing pest management procedures

Field observations in Brazil indicated that *P. citricarpa* does not induce symptoms in Tahiti lime (Baldassari et al., 2009; Wickert et al., 2009; 2012). Consequently, studies evaluating the efficacy of fungicide sprays, cultural measures or postharvest treatments for the control of CBS on this citrus species are not available. However, because *P. citricarpa* can survive as latent mycelia in asymptomatic fruits of other citrus species under existing CBS management procedures, it is also likely to survive in Tahiti lime fruit. Therefore, the pathogen is **very likely** to survive existing pest management procedures in Tahiti lime fruit. The level of uncertainty is **high**, due to the limited information available.

3.2.3.4. Probability of transfer to a suitable host

The pathogen can colonize Tahiti lime fruit under field conditions in Brazil (Baldassari et al., 2009), but there are no reports of symptom development or any reproduction of the pathogen on fruits of this citrus species. Nevertheless, it is not known whether CBS symptoms could develop or *P. citricarpa* could reproduce in harvested fruit of Tahiti lime after long storage periods or under waste disposal conditions outdoors. The pathogen could transfer to a suitable host only if it were able to sporulate on fruits or peel of Tahiti lime discarded in the vicinity of citrus trees in the PRA area, provided that environmental conditions are favourable for spore production, release, dissemination and subsequent infection (see section 3.3.2).

The Panel considers that the probability of transfer is rated as **very unlikely,** with **high** uncertainty due to the lack of studies on this issue.

3.2.4. Entry pathway III: citrus fruit import by passenger traffic

This is a pathway of lesser importance compared to the commercial fruit pathway, but could still result in pathogen entry. There is generally a lack of information on the volumes of citrus fruit imported by passengers, the probability of survival during transport and the likelihood of interception at points of entry if border inspection was in place. A graphical representation of this pathway is given in figure 17.

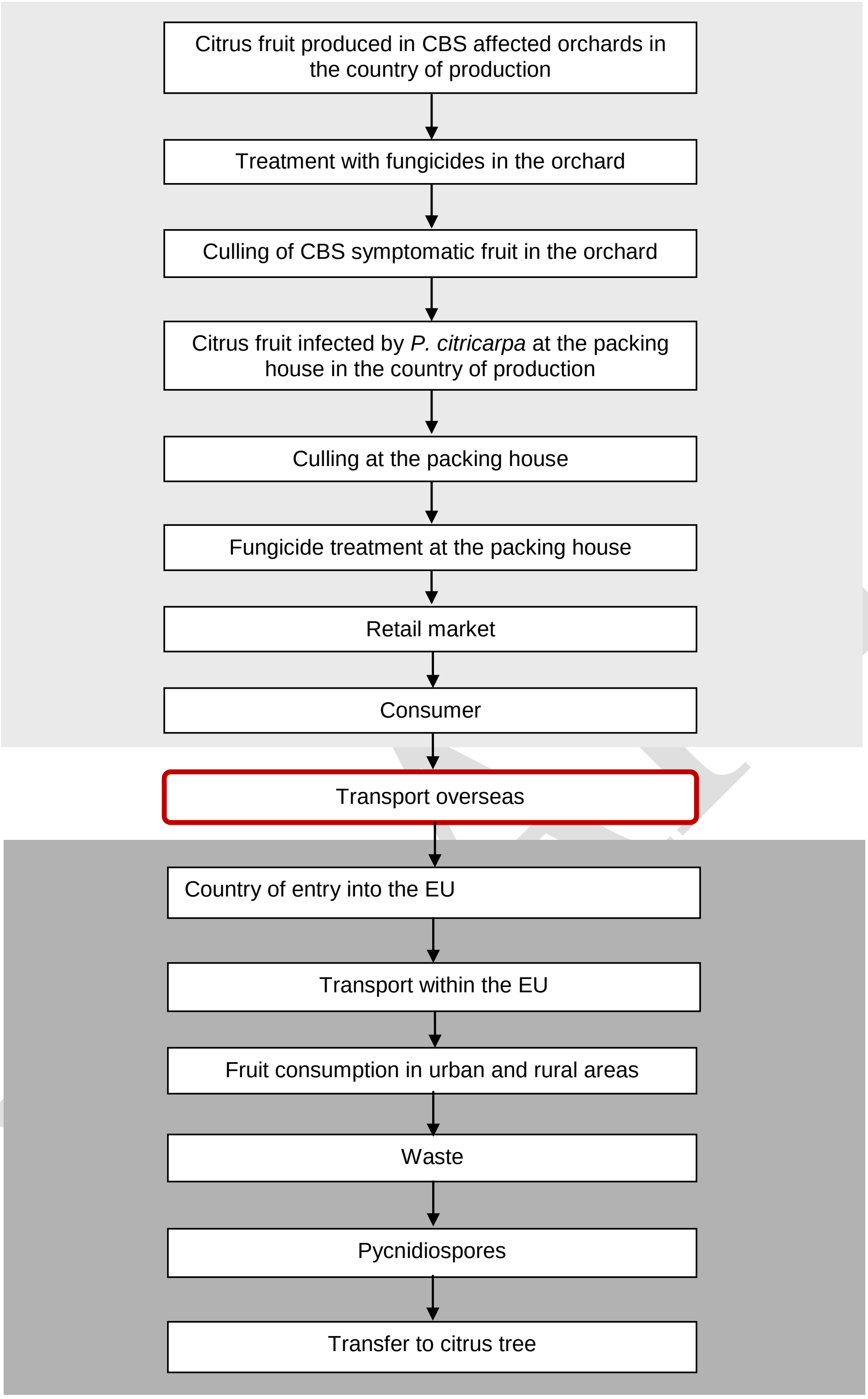


Figure 17: A graphical pathway model illustrating steps in the entry pathway of *Phyllosticta citricarpa* with citrus fruit imported by passengers. The pathway starts in CBS affected orchards in a country of origin outside the EU and ends with the transfer of spores of the pathogen to a host within the EU. The scheme is illustrative and departures from the depicted sequence may apply in some countries of origin or certain EU MSs due to local characteristics of citrus production, trade, and processing. For instance, in current practice, there is import inspection, but, in the scenarios considered in this opinion, there is no inspection specifically for CBS.

3.2.4.1. Probability of association with the pathway at origin

Citrus fruit brought into the EU by passengers can be infected by *P. citricarpa* if passengers arrive from the countries where the disease is present. In these countries, citrus fruit produced for the local market is likely to have a higher incidence of CBS than fruit produced for export markets. Therefore, citrus fruit bought by travelers into the EU is more likely to be infected with *P. citricarpa* than commercially imported fruit. The presence and severity of CBS in these countries is variable, and this variability will affect the probability of association with the pathway at the origin.

Based on the above considerations, the Panel concludes that the probability of association with the pathway at the origin is rated as **likely**, with **medium** uncertainty due to the lack of information on the volume and frequency of the movement along the pathway.

3.2.4.2. Probability of survival during transport or storage.

Experimental studies on *P. citricarpa* survival during transport by passengers appear to be lacking, but it can be assumed that if the pathogen can survive commercial transport and storage, it is very likely that it will survive the conditions of transport of individual passengers.

Based on this, the Panel considers that the probability of survival during transport or storage is rated as **very likely**, with **low** uncertainty despite the lack of information, by analogy with the commercial fruit pathway.

3.2.4.3. Probability of survival to existing pest management procedures

Inspections to see whether passengers carry citrus fruit with them when arriving at EU airports from countries where CBS is present are not systematic. There does not seem to be information available about how frequently passengers carry citrus fruit when arriving into the EU from CBS- infected Third Countries and how likely it is for such passengers to be identified, so that pest management procedures could be potentially applied.

Data on citrus fruit interceptions on individual international passengers are available from two regions of Australia (Central East Region: 8557 citrus fruit seized, Jan 2010-Mar 2011; South Eastern Region: (4892 citrus fruit seized, Jan 2010-Apr 2011; Australian Government, 2011). Considering that most international passengers arriving in Australia fly to these Central/South Eastern Regions, and since there are about 2 million international passengers per month (Australian statistics, this would roughly imply 1 million incoming passengers), a conservative estimate of about 1 passenger out of 1,000 carries one citrus fruit.

The figure can be considered as a low estimate if substantial numbers of international passengers fly to Australian airports from outside the Central/South Eastern regions and also taking account of the fact that some citrus fruit may not be noticed. However, only some this citrus fruit would be affected by CBS because not all passengers carrying fruit arrive from countries where *P. citricarpa* is present.

Based on this information, the Panel considers that the probability of surviving pest management procedures is rated as **very likely**, with **low** uncertainty despite the lack of information on this pathway, by analogy with the commercial fruit pathway.

3.2.4.4. Probability of transfer to a suitable host

The probability that CBS-affected citrus fruit imported by passengers may then transfer the pathogen to a suitable host is influenced by the proportion of passengers that:

- travel from an area where CBS is present to a citrus-producing EU country,
- carry CBS-affected citrus fruit bearing pycnidiospores, which can then be splash-dispersed onto citrus trees.

- discard citrus peel and fruit waste in proximity to citrus trees,
- arrive during a period with environmental conditions potentially conducive to infection (roughly speaking: April-May, and/or September-October),

The Panel considers that the probability of transfer of *P. citricarpa* to a suitable host in the risk assessment area by passengers discarding citrus fruit near fruit trees is rated as **unlikely**, with **medium** uncertainty due to the lack of information on the likelihood that the above mentioned events will take place.

3.2.5. Entry pathway IV: citrus fruit with leaves and peduncles in commercial trade

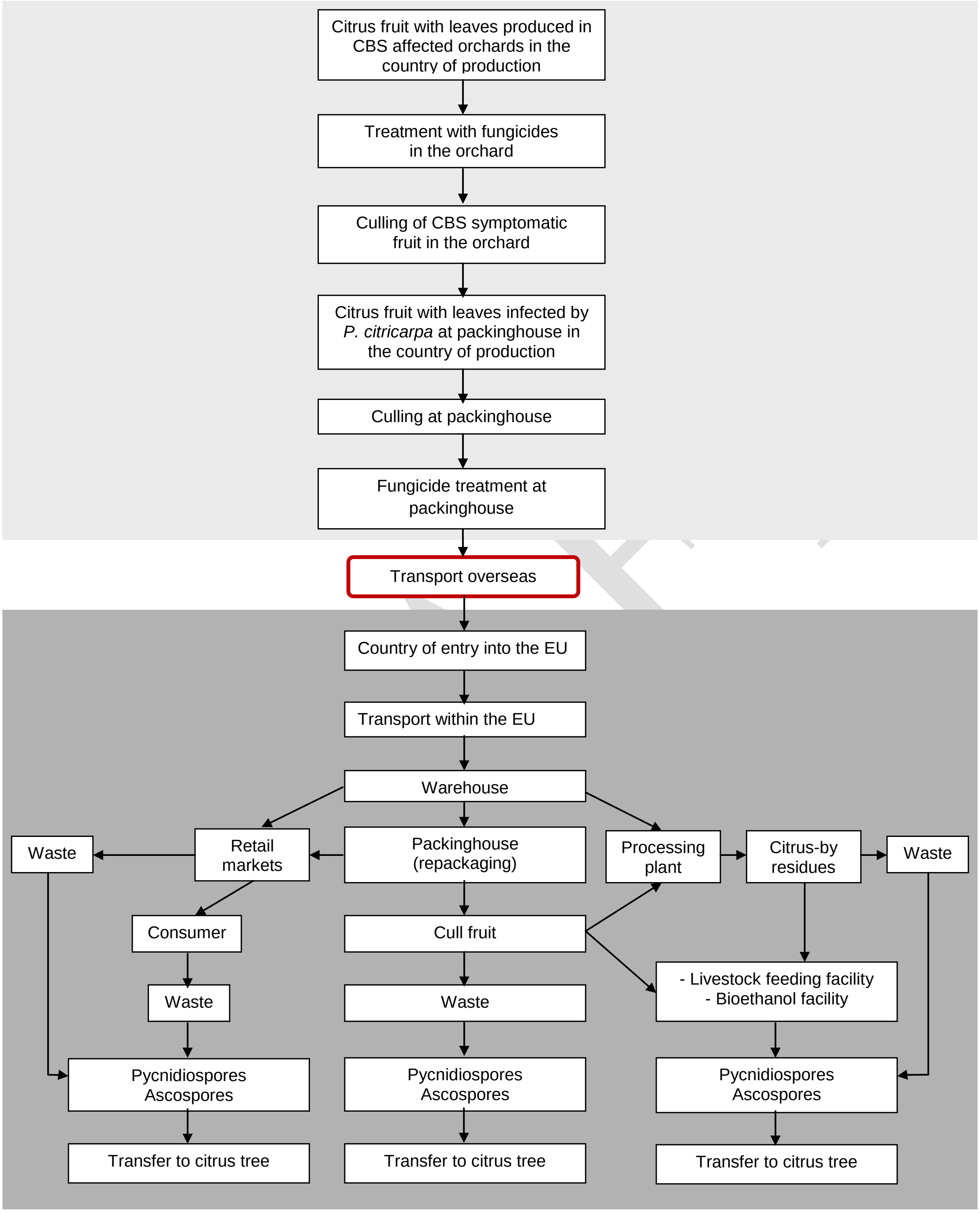


Figure 18: A graphical pathway model illustrating the entry pathway of *Phyllosticta citricarpa* with commercial trade in citrus fruit with leaves and peduncles. The pathway starts in CBS affected orchards in a country of origin outside the EU and ends with the transfer of spores of the pathogen to a host within the EU. The scheme is illustrative and departures from the depicted sequence may apply in specific countries of origin or specific MSs of the EU, depending upon local characteristics of citrus production, trade, and processing. For instance, in current practice, there is import inspection, but in the scenarios considered in this opinion, there is no inspection specifically for CBS.

A graphical representation of the pathway of commercial trade of citrus fruit with leaves and peduncles is shown in figure 18.

Although importation from Third Countries of citrus fruit with leaves is currently prohibited by EU legislation, there have been a number of interceptions made by EU countries of consignments with citrus fruit with leaves originating from Third Countries over the last years (Europhyt data). For example, citrus fruit with leaves were intercepted in consignments from Bangladesh to Denmark (2007); from Cameroon to Switzerland[12] (2012); from the Dominican Republic to the United Kingdom (2004); from Lebanon to Denmark (2000), France (2001) and the United Kingdom (no year given); from Morocco to the Netherlands (2000); from Pakistan to Germany (2009) and the United Kingdom (no year given); from Sri Lanka to Switzerland (2011); from Thailand to Denmark (no year given), Germany (2006 and 2010), the Netherlands (2000), the United Kingdom (2005 and 2006), Sweden (2000) and Switzerland (2011); from Turkey to Austria (2001); and from Vietnam to the Czech Republic (2009 and 2010), Germany (2006) and Switzerland (2011).

This number of interceptions of commercially traded citrus fruit with leaves should be considered as a conservative estimate because in many cases Europhyt interceptions of citrus “for other reasons including leaves” do not provide the specific reason for the interception, whereas the list above only includes interceptions that specifically mentioned citrus leaves. Moreover, four out of the 20 (20 %) above mentioned interceptions were made by Switzerland, a country whose imports of citrus fruit are much smaller than those of many EU MSs, but whose border controls may be stricter than for many other countries.

3.2.5.1. Probability of association with the pathway at origin

The probability of association with the pathway of citrus fruit with leaves (commercial trade) is similar to that for citrus plants for planting and citrus commercial fruit.

Therefore, taking into account the assessment of entry by these pathways, the pest is **likely** to be associated with the pathway at origin with **medium** uncertainty. In addition, there are the following considerations:

No trade data are available on the volume of citrus fruit with leaves and peduncles imported into the EU from countries where *P. citricarpa* is present . Nonetheless, the Panel considers that, owing to consumer preference for the consumption of citrus fruit still bearing fresh leaves (Li et al., 2013), there would be a non-negligible volume of citrus fruits with leaves, a fraction of which would be imported into the EU citrus-growing regions.

Uncertainties include: 1) the volume of citrus fruit with leaves that would be imported by EU citrus-growing countries (directly or indirectly through re-distribution from non-citrus-growing EU countries) in the absence of the current EU legislation forbidding such imports, 2) the number of imported citrus fruit with leaves with CBS infection, and 3) the effectiveness of any potential inpections at the EU points of entry to detect CBS infected citrus fruit with leaves.

3.2.5.2. Probability of survival during transport or storage

Since commercial citrus fruit with leaves is stored and transported under conditions that are not stressful or damaging for leaf tissues (so as to preserve citrus leaves in fresh conditions), the probability that *P. citricarpa* will survive transport and storage of citrus fruit with leaves, exported from countries where *P. citricarpa* is present into the EU, is rated as **very likely**, with a **low** level of uncertainty.

3.2.5.3. Probability of survival of existing pest management procedures

As noted for the plants for planting pathway:

[12] Switzerland is not a EU MS but it records its interceptions in the Europhyt database.

- the application of fungicides in citrus orchards can diminish disease incidence and severity, but does not eradicate infections;
- visual inspections are most likely to miss latently infected (asymptomatic) fruit and leaves;
- CBS symptoms on fruit are variable and they are rarely observed on leaves, with the exception of lemon leaves; in addition symptoms may be misidentified during visual inspection, as lesions are similar to those produced by other citrus pathogens;

Therefore, the Panel concludes that it is **very likely** that *P. citricarpa* will survive existing management procedures and remain undetected on commercial citrus fruit with leaves. The uncertainty is considered **medium** due to the lack of data on the volume of citrus fruit with leaves that could be potentially imported into the EU from infested Third Countries.

3.2.5.4. Probability of transfer to a suitable host

As noted above for the two main CBS pathways, discarded citrus fruit, peel or other citrus fruit waste with leaves and peduncles, derived from packinghouses, processing plants, fresh fruit markets, households, etc. and their management, would pose a risk of transfer of the pathogen to a suitable host. This is because:

- The long (2-12 months) quiescent period of CBS (McOnie, 1967; Kellerman and Kotzé, 1977; Kotzé, 1981; Aguiar et al., 2012), which in many cases would be longer than the time needed for transport of the commodity;
- CBS symptoms on citrus fruit are variable and they are rarely observed on leaves with the exception of lemon leaves; in addition, symptoms can be easily confused with those caused by other pathogens;
- Commercial citrus fruit with leaves and peduncles is likely to be distributed throughout the EU, including citrus-growing regions;
- The latent mycelium present in citrus leaves, if leaves are then improperly discarded, can then develop pycnidia with splash-dispersed pycnidiospores and pseudothecia with wind-disseminated ascospores that can enable the organism to enter new areas.

Thus, in the absence of the current legislation, the pathogen would be **likely** to be able to transfer by various means (wind, water (rain or irrigation), insects) to susceptible host plants, with a **medium** level of uncertainty deriving from the lack of data on the volume of the waste of citrus fruit and leaves that could potentially be disposed in the vicinity of susceptible hosts in the risk assessment area.

3.2.6. Entry pathway V: citrus plants for planting

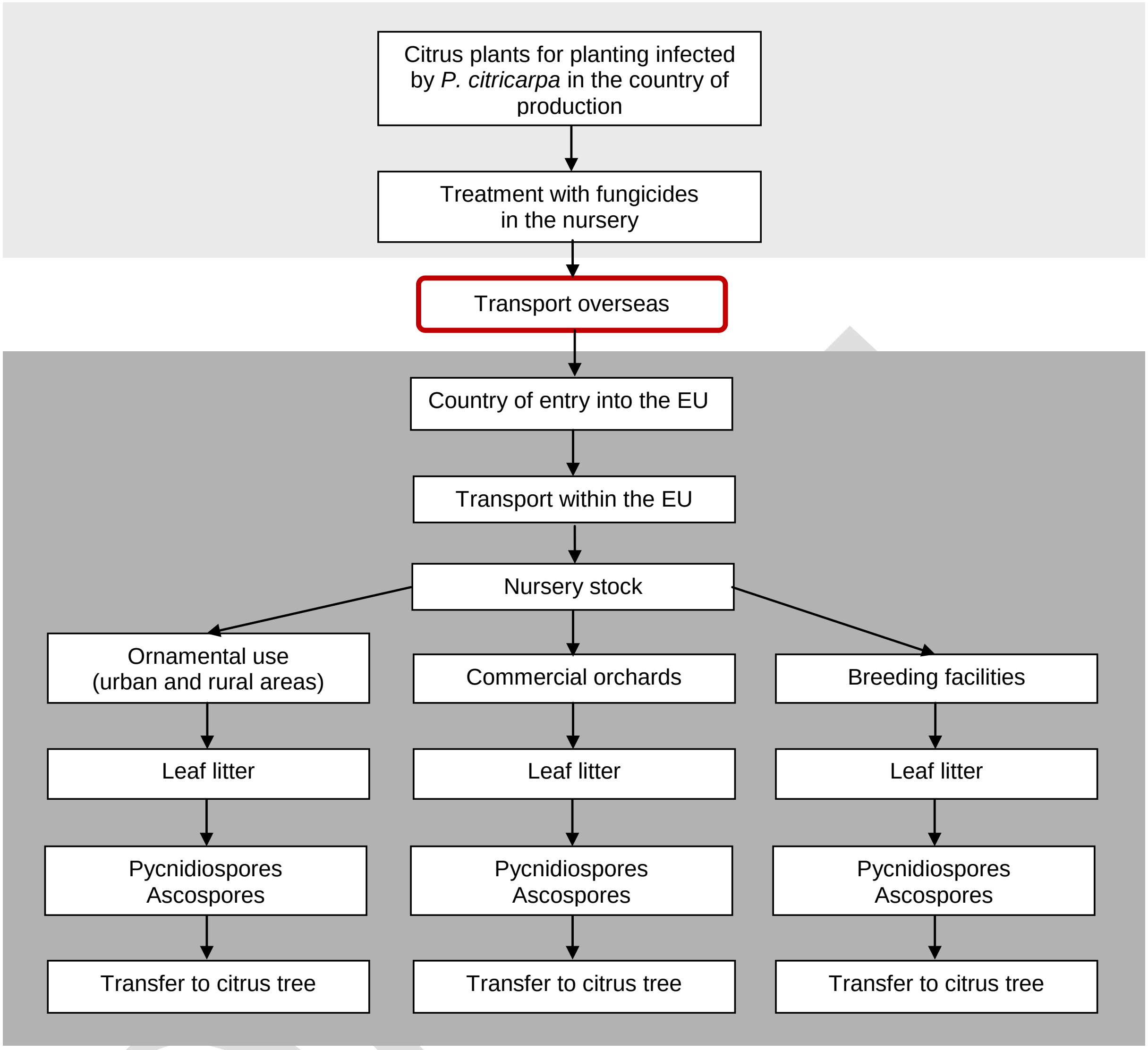


Figure 19: A graphical pathway model illustrating the entry of *Phyllosticta citricarpa* with citrus plants intended for planting. The pathway starts with infected plants in a country of origin outside the EU and ends with the transfer of spores of the pathogen to a host within the EU. The scheme is illustrative and departures from the depicted sequence may apply in specific countries of origin or specific MSs of the EU, depending upon local characteristics of citrus production. For instance, in current practice, there is import inspection, but, in the scenarios considered in this opinion, there is no inspection specifically for CBS.

The trade of citrus plants for planting (Fig. 19) is considered to be a very important potential pathway for the entry of *P. citricarpa* into new areas (Wager, 1949; Whiteside, 1965; Kotzé, 1981; Cortese et al., 2004; MacLeod et al., 2012). This is because citrus plants are normally propagated vegetatively by grafting onto rootstocks. Aerial parts of budwood, scions, rootstocks and nursery plants of citrus species in general may be infected with *P. citricarpa* without or with very few symptoms (see section 3.1.1.2).

3.2.6.1. Probability of association with the pathway at origin

P. citricarpa is most likely to be present in citrus propagating material from areas of its current distribution as mycelium in latently infected leaves. Under suitable conditions, *P. citricarpa* pseudothecia and pycnidia and, in turn, ascospores and pycnidiospores are likely to develop on shed infected citrus leaves, thus making the citrus plant for planting pathway the most effective means of spreading the disease to new areas (Kotzé, 1981).

There are no readily available data on the prevalence of *P. citricarpa* in citrus nurseries in countries where the pathogen is currently distributed. Similarly, there are no detailed data on the location of citrus nurseries in those countries. However, in agreement with MacLeod et al. (2012), the Panel considers that, particularly if citrus nurseries are located near to citrus orchards infected by *P. citricarpa*, then it is likely that there will be a high prevalence of the pathogen in citrus planting material for propagation purposes.

Foliar lesions of CBS are rare, especially in young vigorous plants (Kotzé, 1981). Therefore, culling in citrus nurseries in CBS-affected countries is not likely to lead to removal and destruction of seedlings with latent infections, as only symptomatic seedlings are likely to be detected.

The Panel considers it to be highly likely that infected citrus plant propagation material will be asymptomatic. This is because CBS does not generally appear on trees until they are over 10 years old and it has been known to remain latent for even longer periods (Whiteside, 1965; Kotzé, 1981). In addition, in most varieties, symptoms on leaves are generally absent or very limited, with the exception of lemon (Kotzé, 1981) (see sections 3.1.1.2 and 3.1.1.3 for more details).

As the import of citrus plants into the EU is forbidden, no trade data are available on the volume of citrus plant propagation material from countries where *P. citricarpa* is present to the EU. Nonetheless, in agreement with MacLeod et al. (2011b), the Panel considers that, owing to the large citrus-growing area in EU Southern MSs (table 6) and with a yearly rate of citrus tree renewal of 7.5% (Aubert and Vullin, 1997), in absence of such prohibition high volumes of citrus plant propagation material would be potentially imported in the EU.

Therefore, in agreement with MacLeod et al. (2012), the Panel considers that the pest is **likely** to be associated with the pathway at origin taking into account factors such as cultivation practices and the treatment of consignments with **medium** uncertainty, because of the lack of trade data of citrus planting material and on the structure of the trade network for citrus plants for planting in the EU.

3.2.6.2. Probability of survival during transport or storage

Considering that commercial citrus plant propagation material, as it happens with with all live plants, is stored and transported under conditions that are not stressful or damaging for plant tissues (and thus also not stressful to the latent mycelium of the pathogen). Therefore, and in agreement with MacLeod et al. (2012), the probability that *P. citricarpa* will survive transport and storage of citrus plant propagation material originated in infested Third Countries and imported into the EU, is assessed as **very likely**, with a **low** level of uncertainty.

3.2.6.3. Probability of survival existing pest management procedures

In agreement with MacLeod et al. (2012), the Panel considers that:

- The application of fungicides in citrus orchards can reduce disease incidence and severity, but it does not eradicate infections. The quiescent period of CBS in affected leaves is likely to be of sufficient duration to extend beyond the time in transit. Visual inspections are most likely to miss asymptomatic citrus plant propagating material infected by *P. citricarpa*;

- If CBS symptoms are present on leaves, they are likely to be relatively similar to those caused by other citrus pathogens (e.g., *Alternaria* spp., *Mycosphaerella citri* Whiteside, *Septoria* spp.) and thus might be misidentified during culling;

- Laboratory testing is needed to reliably detect and identify *P. citricarpa* on citrus plant propagating material (see section 3.1.1.3).

Therefore, the Panel, in agreement with MacLeod et al. (2012) concludes that it is **very likely** that *P. citricarpa* will survive existing management procedures and remain undetected on citrus plant propagating material. Because of the difficulties in identifying CBS symptoms, the uncertainty for this rating is considered **low** despite the lack of published studies on the application of fungicides to control CBS in nurseries where *P. citricarpa* is present.

3.2.6.4. Probability of transfer to a suitable host

With regard to the potential distribution of the imported citrus plants for planting throughout the risk assessment area the Panel considers that:

- Citrus species are extensively grown in EU Southern MSs in orchards (see Table 6), in nurseries for production of plant propagation material, as well as in private and public gardens and as ornamentals. In urban areas, citrus trees are also grown along streets and in squares;
- Lemon (*C. limon*), which is considered the citrus species most susceptible to *P. citricarpa* and usually the first to be affected when CBS outbreaks occur in new areas (Kotzé, 1981), is widely grown both in rural and urban regions, covering 63,000 ha - about one eighth of the total area cultivated with citrus in the EU;
- Citrus plant propagation material potentially imported into the EU would most probably be distributed first to nurseries for planting/grafting and subsequently to orchards, public and private gardens, in both rural and urban areas in the citrus growing EU MSs.

Therefore, in agreement with MacLeod et al. (2012), the Panel concluded that, if imported, citrus plant propagation material would be distributed **moderately widely** throughout the risk assessment area, with a **low** level of uncertainty.

With regard to the ability of the pathogen to be transferred from the imported plants for plantingl to susceptible hosts grown in the citrus-producing EU MSs, MacLeod et al. (2012) considered that:

- Although nurseries will tend to grow young citrus trees (after grafting or budding) for 1-3 years before selling and distributing them to customers, CBS has a long quiescent period (2-12 months) and infected leaves (with the exception of lemon leaves) rarely show symptoms during their life span (up to about 3 years);
- Despite the latent presence of the pathogen in citrus plant propagating material, nurseries provide favourable environmental conditions (high relative humidity and frequent wetting and drying of leaf litter due to overhead irrigation) for the pathogen to produce pycnidiospores and/or spores with which to transfer by natural means to susceptible host plants grown nearby;
- If nurseries use infected citrus rootstocks, budwood or scions as propagation material, the pathogen is very likely to be transferred by human assistance to (and infect) susceptible hosts grown at great distances from the nursery.

The Panel also agrees that the intended use of the commodity would **very likely** aid transfer to a suitable host or habitat, with a **low** level of uncertainty because:

- The intended use of citrus plant propagating material is planting (rootstocks) or grafting (scions, budwood);
- If citrus plant propagating material is infected by *P. citricarpa*, then there will be the opportunity for the pathogen either to infect directly the host plants (in case infected

budwood/scions are grafted onto citrus trees grown in the risk assessment area) or to be transferred by both natural means and human assistance from infected to susceptible host plants grown in citrus orchards, nurseries, private and public gardens;

- Spread of the pathogen is possible in various ways, naturally through wind and water-splash dispersal, but also with human assistance via infected scions and budwood;

- Improper management of leaf litter in CBS-affected nurseries may also result in transfer of the pathogen to healthy citrus hosts nearby, because the pathogen can produce ascospores and pycnidiospores on leaf litter, which can be spread by wind, rain or irrigation water.

The Panel therefore agrees with the conclusions by MacLeod et al. (2012) that the pest is **very likely** to be able to transfer from the pathway to a suitable host or habitat, with a **low** level of uncertainty.

3.2.7. Entry pathway VI: Tahiti lime (Citrus latifolia) plants for planting

A representation of the Tahiti lime plants for planting pathway is given in Fig. 20.

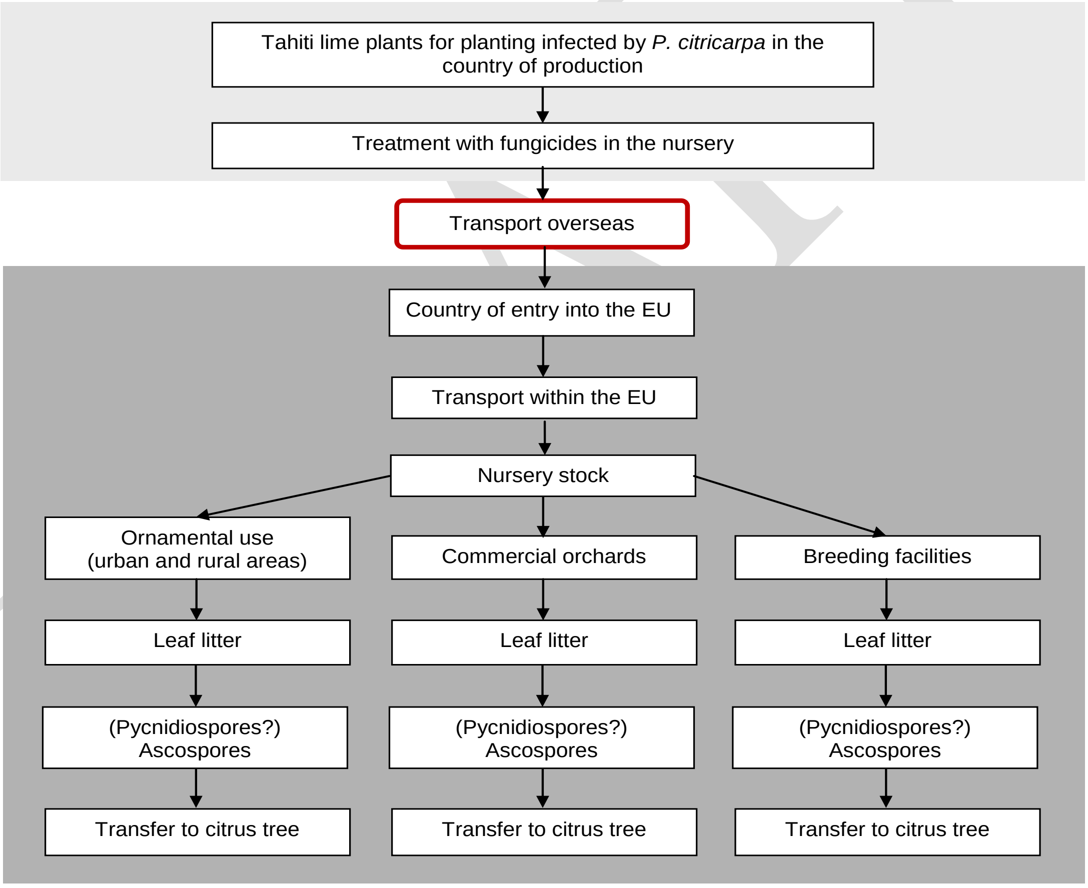


Figure 20: A graphical pathway model illustrating the entry pathway of *Phyllosticta citricarpa* with Tahiti lime (*Citrus latifolia*) plants intended for planting. The pathway starts with infected plants in a country of origin outside the EU and ends with the transfer of spores of the pathogen to a host within the EU. The scheme is illustrative and departures from the depicted sequence may apply in specific countries of origin or specific EU MSs, depending upon local characteristics of citrus production, trade, and processing. For instance, in current practice, there is import inspection, but in the scenarios considered in this opinion, there is no inspection for CBS.

3.2.7.1. Probability of association with the pathway at origin

In a study conducted in Conchal, Sao Paulo (Brazil), Baldassari et al. (2009) identified two isolates of *P. citricarpa* from a total of seven *Phyllosticta* isolates obtained from Tahiti lime leaves. In addition, ascospores of *P. citricarpa* formed in Tahiti lime leaves were captured using a wind tunnel. In other studies, the population genetics of *Phyllosticta* in Tahiti lime were characterized in two regions in Brazil, Estiva Gerbi/Conchal (Sao Paulo) and Itaborai (Rio de Janeiro) (Wickert et al., 2009; 2012). Leaves were collected from 24 different Tahiti lime trees in each region to obtain one *Phyllosticta* isolate per plant. In addition, 40 leaves per tree were collected from three different trees in each region to obtain 24 *Phyllosticta* isolates from the same plant. A total of 208 *Phyllostica* isolates were studied. All isolates from Itaborai were identified as *P. capitalensis*, but eight out of the 18 *Phyllosticta* isolates from Estiva Gerbi inoculated in sweet orange fruit induced CBS symptoms and were identified as *P. citricarpa* based on their morphological and molecular characteristics. Since these studies did not describe how the sampling was conducted and in particular from which plants and locations the eight *G. citricarpa* isolates were collected, it is not possible to determine precisely the prevalence of *P. citricarpa* in Tahiti acid lime leaves in Brazil. Despite the limited temporal and geographical range of these studies, these results clearly indicate that *P. citricarpa* can colonize and reproduce in Tahiti lime leaves.

Therefore, the pathogen is **likely** to be associated with the pathway at origin, with a **high** level of uncertainty because of the variation in disease prevalence in different regions and the lack of information on this pathway.

3.2.7.2. Probability of survival during transport or storage

Currently, there is no trade in citrus plants for planting imported from Third Countries into the EU, so the probability of survival of *P. citricarpa* in infected Tahiti lime plants cannot be quantified. However, since the pathogen can colonize Tahiti lime leaves and citrus plants for planting are sold with leaves, there is no reason to consider that the pathogen cannot survive during transport or storage.

This translates into a **very likely** survival during transport or storage, with a low uncertainty.

3.2.7.3. Probability of survival existing pest management procedures

Field trials for the control of *P. citricarpa* on Tahiti lime are not available. However, since *P. citricarpa* can survive under existing management procedures commonly applied to other citrus species, it is also likely to survive in Tahiti lime. Foliar symptoms of CBS are rare in most citrus species, and have been not reported in Tahiti lime. Thus, there is a **very high** probability of the pathogen remaining undetected as latent mycelia in asymptomatic Tahiti lime leaves during potential visual inspection, with **high** uncertainty due to the lack of studies.

3.2.7.4. Probability of transfer to a suitable host

The pathogen can colonize Tahiti lime leaves and reproduce on them forming wind-borne ascospores (Baldassari et al., 2009; Wickert et al. 2009; 2012). If Tahiti lime plants carrying leaves colonized by *P. citricarpa* were planted in the PRA area, ascospores may be formed on these leaves after falling onto the orchard floor. Once mature, ascospores may be released and disseminated relatively long distances, infecting leaves and fruits of nearby susceptible citrus trees in the area. However, this chain of events will occur only if environmental conditions in the PRA area would be conductive to pseudothecia production, ascospore maturation, release, dissemination and subsequent infection.

Nonetheless, by analogy with the citrus plants for planting pathway, the probability of transfer to a suitable host is assessed by the Panel as **very likely**, with a **high** uncertainty due to the lack of information on the above mentioned events.

3.2.8. Entry pathway VII: citrus plants for planting import by passenger traffic

As stated above for the pathway citrus plants for planting (commercial trade), infected citrus plants for planting can be a very important potential pathway for entry of *P. citricarpa* into new areas. If passengers imported scions to be used in the risk assessment area as rootstocks or grafting material (scions, budwood), and if such material is infected by *P. citricarpa*, there is the potential for the pathogen to enter the EU. A graphical presentation is given in Fig. 21.

3.2.8.1. Probability of association with the pathway at origin

The probability of association with the pathway at origin is similar to the citrus plants for planting pathway (commercial trade). The pest is thus **likely** to be associated with the pathway at origin, with **high** uncertainty related to the likelihood that passengers will decide to import citrus propagating material on their own without going through the commercial pathway.

3.2.8.2. Probability of survival during transport or storage

For the reasons described above in the pathway citrus plants for planting (commercial trade), the probability that *P. citricarpa* will survive transport and storage of citrus plant propagation material, exported from countries of *P. citricarpa* current distribution into the EU by passenger traffic, is **very likely**, with a **medium** level of uncertainty regarding the conditions under which citrus plant propagating material will be transported and stored by passengers.

3.2.8.3. Probability of survival existing pest management procedures

Similarly to the commercial pathway citrus plants for planting, it is **very likely** that *P. citricarpa* will survive currently existing management procedures and remain undetected on citrus plant propagating material imported by passengers. The uncertainty is considered **low** despite the lack of information on the application of fungicides to control CBS in orchards and nurseries where *P. citricarpa* is present and from which passengers may decide to take plant propagating material.

3.2.8.4. Probability of transfer to a suitable host

Provided that passengers manage to import infected plant propagating material to the PRA area and that they go on to use this material in private gardens or in commercial orchards in the pest risk assessment area, similarly to the commercial pathway citrus plants for planting, it is **very likely** that the pathogen will be able to transfer from the pathway of citrus plants for planting (passenger traffic) to a suitable host or habitat, with a **low** level of uncertainty, by analogy with the commercial plants for planting pathway.

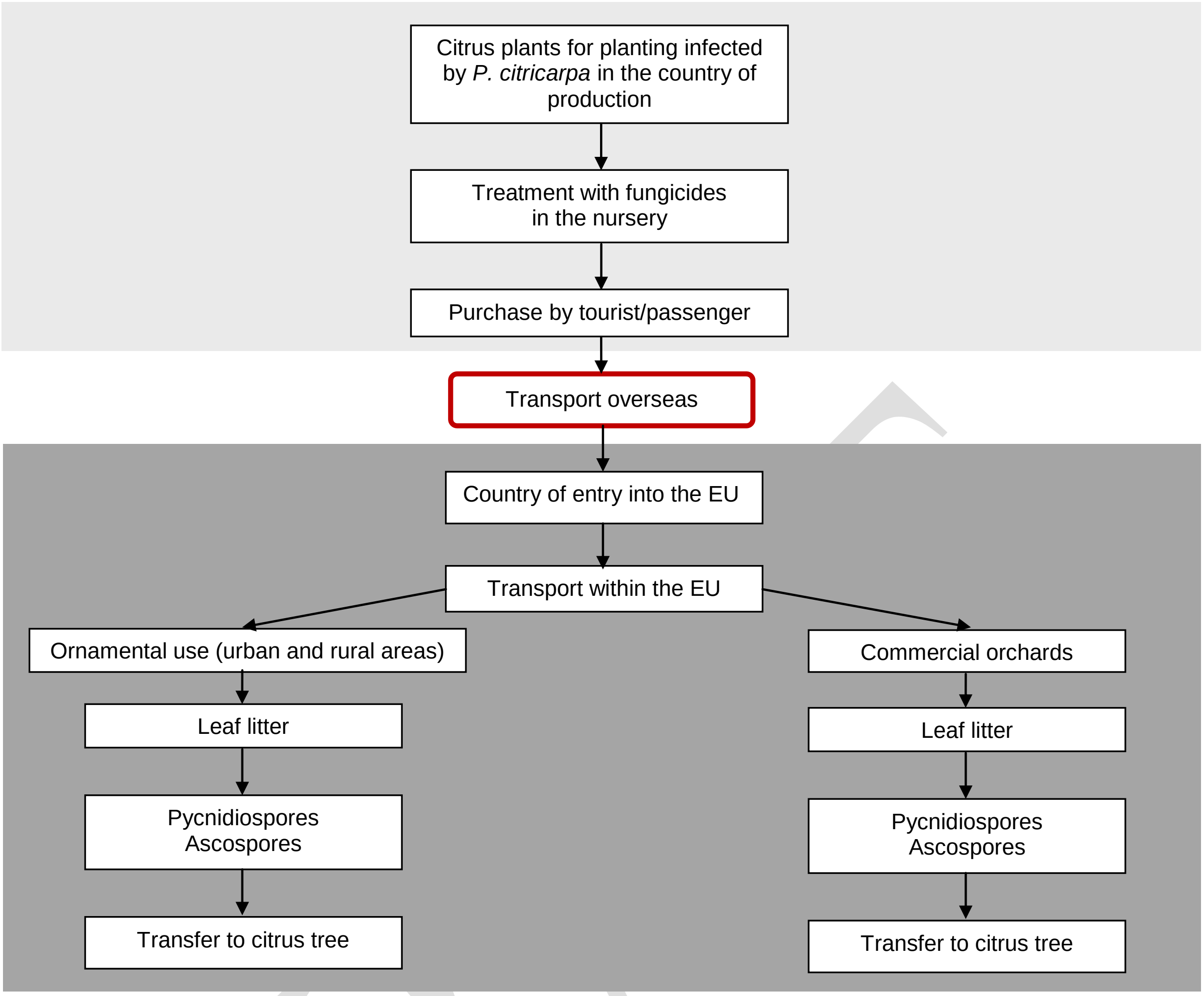


Figure 21: A graphical pathway model illustrating the entry pathway of *Phyllosticta citricarpa* with citrus plants intended for planting and imported by passengers. The pathway starts with infected plants in a country of origin outside the EU and ends with the transfer of spores of the pathogen to a host within the EU. The scheme is illustrative and departures from the depicted sequence may apply in specific countries of origin or specific EU MSs, depending upon local characteristics of citrus production. For instance, in current practice, there is import inspection, but in the scenarios considered in this opinion, there is no inspection specifically for CBS.

3.2.9. Entry pathway VIII: other citrus plant parts: leaves

Little information is available about the trade of citrus plant parts other than live plants and fruits. Limited quantities of citrus leaves are imported for flavouring food. Lemon (*C. limon*) and kaffir lime (*C. hystrix*) are the main species used for these purposes, although a variety of other exotic citrus species are also employed (Butryee et al., 2009). As stated in section 3.2.1, there is not considered to be a significant trade in leaves and branches for other purposes and so this has not been considered further in this opinion.

3.2.9.1. Probability of association with the pathway at origin

The probability of association with the pathway of leaves (commercial trade) of citrus species which are known to be hosts of *P. citricarpa* can be considered to be similar to that for citrus plants for planting and citrus commercial fruit with leaves. However, the status of *C. hystrix* and other exotic citrus species as hosts of *P. citricarpa* is unknown.

Therefore, taking into account the assessment of entry by these pathways, the pest is **likely** to be associated with the pathway at origin with **medium** uncertainty.

Uncertainties include: 1) the status of *C. hystrix* and other exotic citrus species as hosts of *P. citricarpa*, 2) the amount of citrus leaves imported by EU MSs, 3) the number of such imported consignments with *P. citricarpa* infection and the effectiveness of surveys operating at the EU points of entry in detecting *P. citricarpa* infection in leaves.

3.2.9.2. Probability of survival during transport or storage

As indicated in the case of citrus plants for planting and citrus commercial fruit with leaves, if the commercial transport of citrus leaves is carried out under conditions that are not limiting for *P. citricarpa* survival in these plant tissues (so as to preserve citrus leaves in fresh or dry conditions), then the probability that *P. citricarpa* will survive transport and storage in infected citrus leaves exported from countries where *P. citricarpa* is present into the EU, is rated as **likely**, with a **medium** level of uncertainty, given the lack of data on this pathway.

3.2.9.3. Probability of survival of existing pest management procedures

As noted above for the plants for planting and citrus commercial fruit with leaves pathways, the application of fungicides in citrus orchards can diminish *P. citricarpa* incidence and severity but does not eradicate *P. citricarpa* infections. In addition citrus leaves for flavouring or cooking might be produced in untreated or organic orchards to reduce the risk of pesticides residues. Moreover, culling at the country of origin can easily miss asymptomatic citrus leaves infected by *P. citricarpa*: CBS symptoms on leaves are rarely observed and may be misidentified as lesions are similar to those produced by other citrus pathogens.

Therefore, it is **very likely** that *P. citricarpa* will survive the current management procedures and remain undetected on traded citrus leaves. The uncertainty is considered **medium** due to the lack of data on this pathway.

3.2.9.4. Probability of transfer to a suitable host

As noted above for the citrus plants for planting and citrus commercial fruit with leaves pathways, discarded citrus leaves can pose a risk of transfer of the pathogen to a suitable host via airborn ascopores. This is because of: 1) the long quiescent period of *P. citricarpa*, 2) the difficulties in detecting CBS symptoms on citrus leaves, 3) the distribution of citrus leaves for flavouring or cooking throughout the EU, including citrus-growing regions, 4) the potential development of pycnidia with pycnidiospores and pseudothecia with ascospores on infected citrus leaves that might be discarded in the vicinity of citrus trees in the pest risk assessment area. However, the transfer from citrus leaves for flavouring or cooking is much less likely to occur because the majority of mycelium and spores will likely be destroyed by cooking.Moreover, the imported citrus leaves for flavouring or cooking are unlikely to be sorted and packed in packing houses near citrus orchards and any discards may remain in thei original packaging.

Thus, the pathogen would be **unlikely** to be able to transfer by various means (wind, water (rain or irrigation), insects) to susceptible host plants, with a **medium** level of uncertainty deriving from the lack of data on this pathway.

3.2.10. Conclusion on the probability of entry

The Panel has assessed the overall probability of entry by combining the ratings of the various steps for each pathway, following the rule that within each pathway the overall assessment should not be higher than the lowest probability. The ratings are presented in Table 3 and the justification for the overall ratings is summarised in Table 4.

Table 5: Ratings for the probability of entry and uncertainty for relevant entry pathways, under the scenario of absence of EU phytosanitary measures but with application of standard disease management practices in the country of origin, to comply with fruit quality standards.

Pathways	**Probability of association with the pathway at origin**		**Probability of survival during transport or storage**		**Probability of survival to existing pest management procedures**		**Probability of transfer to a suitable host**		**Overall probability of entry along the pathway**	
	Probability	Uncertainty	Probability	Uncertainty	Probability	Uncertainty	Probability	Uncertainty	Probability	Uncertainty
I,Citrus fruit trade	Likely	medium	Very likely	low	Very likely	low	Moderately likely	medium	Moderately likely	medium
II,Tahiti lime (*Citrus latifolia*) fruit trade	Likely	high	Very likely	high	Very likely	high	Very unlikely	high	Very unlikely	high
III,Citrus fruit import by passengers traffic	Likely	medium	Very likely	low	Very likely	low	Unlikely	medium	Unlikely	medium
IV,Citrus fruit with leaves trade	Likely	medium	Very likely	low	Very likely	medium	Likely	medium	Likely	medium
V.Citrus plants for planting trade	Likely	medium	Very likely	low	Very likely	low	Very likely	low	Likely	low
VI.Tahiti lime (*Citrus latifolia*) plants for planting trade	Likely	high	Very likely	Low	Very likely	high	Very likely	high	Likely	high
VII,Citrus plants for planting import by passengers traffic	Likely	high	Very likely	medium	Very likely	low	Very likely	low	Likely	medium
VIII.Citrus leaves for flavouring or cooking	Likely	medium	Likely	medium	Very likely	low	Unlikely	low	Unlikely	medium

1658 **Table 6:** Justification for ratings of probability of entry

Rating for entry	Justification
Citrus fruit trade **Moderately likely**	• Cultural practices and treatments applied in the current distribution areas of *P. citricarpa* may reduce the incidence and severity of CBS on citrus fruit imported into the PRA area, but they will not eliminate the pathogen, as also confirmed by the meta-analysis performed as part of this Opinion. • There is a high volume of citrus fruit imported every year into the EU from Third Countries where *P. citricarpa* is reported. The pathogen has been repeatedly intercepted at the EU borders on commercial citrus fruit imports over the last few years. • There is seasonality in citrus fruit imports, but the traditional period of arrival coincides in part with two periods of host susceptibility (European late spring and early autumn). • *P. citricarpa* is very likely to survive transport and storage in the form of (i) pycnidiospores within pycnidia in fruit lesions and/or (ii) latent mycelium present in asymptomatic fruit. • *P. citricarpa* is very likely to survive existing pest management procedures, particularly on latently infected (asymptomatic) fruit and fruit with low disease incidence and severity. • Although citrus fruit consignments are very widely distributed throughout the EU and they tend to arrive at a time of the year suitable for pest establishment, the intended use of the commodity (processing and human consumption) makes it moderately likely that the pathogen will transfer to a suitable host.
Tahiti lime (Citrus latifolia) *fruit trade* **Very unlikely**	• The probability of association of the pathogen with the pathway at origin is high as latent mycelia in asymptomatic fruits. • The likely survival during transport or storage of *P. citricarpa* as latent mycelia present in asymptomatic fruits is very high also on Tahiti lime. • Because *P. citricarpa* can survive as latent mycelia present in asymptomatic fruits of other citrus species under existing CBS management procedures, it is very likely to survive also in Tahiti lime fruit. • The transfer to a suitable host is the limiting factor for this pathway, as pathogen sporulation on whole fruits or peel of Tahiti lime has never been observed.
Citrus fruit import by passengers traffic **Unlikely**	• In countries where *P. citricarpa* is present citrus fruit produced for the local market is likely to have a higher incidence of *P. citricarpa* infection than fruit produced for export markets. • If the pathogen can survive commercial transport and storage, it is just as possible for it to be transported with citrus fruit carried by passengers. • Data on citrus fruit interceptions from Australia lead to a conservative and rough estimate of about 1 airplane passenger out of 1,000 carrying citrus fruit; given the sheer numbers of passengers flying into the EU, this would make it unlikely for control procedures to be able to stop the pathogen at the borders.

	• Since passengers are unlikely to discard fruit in the proximity of citrus orchards, , and due to the small number of citrus fruit potentially entering the EU on this pathway, the panel considers that the probability of transfer to a suitable host from this pathway is low.
Citrus fruit with leaves trade **Likely**	• The probability of association with the pathway of citrus fruit with leaves and peduncles (commercial trade) is similar to the citrus plants for planting and citrus commercial fruit pathways. • Although the importation from Third Countries of citrus fruit with leaves is prohibited by EU legislation, there have been a number of interceptions over the last few years. • Commercial citrus fruit with leaves is stored and transported under conditions that are not stressful or damaging for leaf tissues and thus to the pathogen. • Pest management procedures (pre- and post-harvest fungicide treatments, culling, physical treatments at packinghouses, etc) do not eliminate the pathogen; CBS symptoms can be misidentified or missed; latent infection is common. • If citrus fruit with leaves are improperly discarded, the latent mycelium present on them can develop pycnidia with pycnidiospores which can then go on to infect the host under suitable conditions. In addition, in the case of leaves, the pathogen can produce pseudothecia with wind-disseminated ascospores, which may spread the pathogen over long distances
Citrus plants for planting trade **Likely**	• Particularly if citrus nurseries at the place of origin are located close to infected citrus orchards, it is likely that there will be a high prevalence of the pathogen in citrus plant material for propagation purposes. • Citrus plant propagation material, as with all living plants, is stored and transported under conditions that are not stressful or damaging for plant tissues. The pathogen can survive those conditions. Cultural practices and fungicides applied in citrus nurseriesat the place of origin are unlikely to eradicate the pathogen from infected leaves; CBS symptoms on leaves are similar to those of other citrus diseases and latent infections are very common. • The pathogen is very likely to be able to transfer from the pathway to a suitable host in the RA area, because the intended use of plants for planting, including scions and budwood is very likely to aid such transfer.
Tahiti lime (Citrus latifolia) *plants for planting trade* **Likely**	• The ratings on this pathway were given by analogy with the citrus plants for planting trade pathway.
Citrus plants for planting import by passengers traffic **Likely**	• The ratings on this pathway were given by analogy with the citrus plants for planting trade pathway.

Citrus leaves for flavouring or cooking **unlikely**	• The transfer from citrus leaves for flavouring or cooking is much less likely to occur than from leaves of citrus plants for planting and citrus fruit with leaves because the majority of mycelium and spores will likely be destroyed by cooking. • Moreover, the imported citrus leaves for flavouring or cooking are unlikely to be sorted and packed in packinghouses close to citrus orchards and any discarded material is likely to remain in their original packaging

3.2.11. Uncertainties on the probability of entry

Table 7: Rating and justification for the uncertainty on the probability of entry

Rating for uncertainty	**Justification**
Citrus fruit trade **medium**	• The main uncertainties concerning this pathway include: • the prevalence of the pathogen in the various regions of CBS-infested Third Countries, • whether or not pomelo (*C. maxima*) is susceptible to *P. citricarpa*, • the frequency and quantity of infected fruit/peel or other citrus fruit by-products discarded in close proximity to susceptible hosts in the citrus-growing regions of the PRA area.
Tahiti lime (Citrus latifolia) *fruit trade* **high**	• There is a high uncertainty about all the stages of this pathway. • Most importantly, it is not known if *P. citricarpa* could develop symptoms and fruiting bodies in harvested fruits of Tahiti lime after long storage periods or under outdoor waste disposal conditions
Citrus fruit import by passengers traffic **medium**	• There is a lack of information concerning the volume and frequency of the movement of infected citrus fruit imported by passengers. • One key uncertainty is the probability that passengers will dispose citrus peel and whole fruit waste in the proximity of susceptible hosts in the RA area (citrus orchards, private gardens, nurseries, etc)..
Citrus fruit with leaves trade **medium**	• There is lack of data on the volume of citrus fruit with leaves that could be potentially imported into the RA area from infested Third Countries. • There is lack of data on the frequency and volume of citrus fruit with leaves that could potentially be discarded in proximity to citrus nurseries and orchards in the RA area.
Citrus plants for planting trade **low**	• There is a lack of data on the prevalence of *P. citricarpa* in citrus nurseries in countries with presence of CBS. • Uncertainty persists on compliance with reporting and quarantine regulations of plant nurseries, as well as on the likely structure of the trade network of citrus plants for planting.
Tahiti lime (Citrus latifolia) *plants for planting trade*	• Little is known about the prevalence of CBS on this pathway at origin. • Trade in citrus plants for planting imported from Third Countries into the EU is not allowed, so there is a lack of information on the survival of *P. citricarpa* in imported Tahiti lime plants.

high	• The chain of events that could lead to transfer of the pathogen to the host is also associated with high uncertainty, due to the general lack of studies.
Citrus plants for planting import by passengers traffic **medium**	• No data exists on the import of such material in the EU by passengers There is uncertainty concerning the conditions under which citrus plant propagating material will be transported and stored by passengers.
Citrus leaves for flavouring or cooking **medium**	• There is a general lack of data on this pathway

3.2.12. Comparison of entry conclusions with other PRAs

The entry ratings summarized above are in broad agreement with the assessment by USDA APHIS (2010a), when taking into account the fact that the APHIS ratings were assessed in the presence of regulations, whereas those of the Panel were made in the absence of regulations. That is why USDA APHIS (2010a) assessed the probability of entry of the pathogen through the citrus plants for planting pathway as low (the pathway is not permitted if US regulation is strictly enforced), whereas the Panel concluded that such probability was high (if current EU regulations were lifted).

There is a discrepancy for the non-commercial citrus fruit pathway: USDA APHIS (2010a) judged the probability of entry to be high (based on the many interceptions at US borders), whereas the Panel considered that this probability was low (based on the intended use of the commodity, which would not favour transfer to a suitable host).

There is a disagreement with the rating of the South African PRA (2000) concerning the incidence of the pathogen in exported fruit. The South African PRA judges this to be low due to pre-harvest control measures and inspections. Based on a meta-analysis of available data, the Panel concluded that the incidence of the pathogen at origin is high in the absence of control treatments, and non-negligible even in the presence of control treatments.

The survival of the pathogen during transport was judged by the South African PRA to be low due to the packing house treatments and shipping conditions. However, based on the literature reviewed, the Panel concluded that the pathogen is very likely to survive transport and storage of citrus fruit.

Similar points to the South African PRA were made by Cortese et al. (2004). Their PRA stressed the effectiveness of post-harvest treatments in reducing the viability of pycnidiospores present on infected fruit. The Panel concluded instead that such treatments do not completely eliminate the pathogen.

3.3. Probability of establishment

3.3.1. Availability of suitable hosts in the risk assessment area

Citrus is grown commercially for fruit production in all the countries of the EU with a Mediterranean climate: Croatia, Cyprus, France, Greece, Italy, Malta, Portugal and Spain. The cultivated area of orange, lemon and small fruited citrus varieties in the EU by countries and regions is given in Table 6. A total of 62 854 ha are cultivated with lemon, the citrus species most susceptible to *P. citricarpa* (Kotzé, 1981), covering about 13% of the citrus-growing area in the EU.

3.3.1.1. Periods of susceptibility of citrus leaves and fruits in the risk assessment area

Citrus leaves are susceptible to *P. citricarpa* for 8-10 months (Truter et al., 2004) and sweet orange fruits are susceptible for at least 6-7 months after fruit set (Reis et al., 2003; Baldassari et al., 2006; Brentu et al., 2012), although longer periods have not been evaluated. In countries of the EU with commercial citrus fruit production, citrus has three main leaf flushes per year and fruit set is concentrated in spring (Agustí, 2002; García-Marí et al., 2002). Susceptible leaves and fruits are therefore present in these parts of the risk assessment area in April-May and September-October. In the case of lemons, one or two additional flowering periods may occur in summer (July-September), so fruit at different growth stages are present at the same time (Cutuli et al., 1985; Agustí, 2002).

3.3.2. Suitability of environment

Climate is the key environmental factor that determines the potential for *P. citricarpa* establishment in the EU. The Panel has tackled this issue by:

- Summarising the role played by climatic factors in the life cycle of *P. citricarpa.*
- Reviewing the different methods (principally Paul et al. (2005), (EFSA Panel on Plant Health (PLH), 2008), Magarey et al. (2011), Prima phacie (2012), Yonow et al. (2012) and Fourie et al. (2013)) that have previously been used to assess, *inter alia*, the potential distribution of *P. citricarpa* in Europe. An evaluation of their advantages and disadvantages has been conducted in order to select the most appropriate method to employ in this pest risk assessment.
- Assessing the climatic suitability of *P. citricarpa* in Europe using the most suitable method identified.

3.3.2.1. Summary of the role played by climatic factors in the life cycle of *P. citricarpa*

Several environmental variables are associated with the biology of *P. citricarpa* and the epidemiology of CBS. As described in section 3.1.1.2, *P. citricarpa* has two infection cycles, with a primary cycle driven by ascospores produced by sexual fruiting bodies (pseudothecia) in the leaf litter, and a secondary cycle involving pycnidiospores produced by asexual fruiting bodies (pycnidia) on lesions in fruit, twigs and leaf litter. Warm temperatures and high soil moisture have been associated with rapid leaf litter decay, limiting further pseudothecia and ascospore development (Lee and Huang, 1973). The formation of pseudothecia in the leaf litter and the production and release of ascospores is influenced by the temperature and water regime. Pseudothecia develop from 23-180 days after leaf drop, depending on the frequency of wetting and drying as well as on the prevailing temperatures and the maturation of ascospores occurs almost simultaneously on infected leaves abscised throughout the year (Kotzé, 1963; 1981; McOnie, 1964c; Lee and Huang, 1973). According to Lee and Huang (1973), the optimum temperature for pseudothecia formation is 21-28 °C and no pseudothecia are produced below 7°C or above 35ºC. When mature asci within pseudothecia in the leaf litter are moistened with water, ascospores are ejected into the air and are disseminated by air currents (Kiely, 1948 and 1949; Wager, 1949; McOnie, 1964b; Huang and Chang, 1972; Kotzé, 1988). In the presence of water, ascospores are released when temperatures are between 5 and 25 ºC (Kotzé, 1963).

Table 8: The citrus production area (in hectares) in the EU in 2007 (including Croatia, EU MS since 2013). Data extracted from Eurostat (on line) on 21/02/2013.

Country /region	Orange varieties	Lemon varieties	Small-fruited citrus varieties	All citrus varieties (*)
EU (28 countries) (*)	279 048	62 854	151 510	493 413
Croatia	200	100	1 200	1 500
Cyprus	1 554	665	1 766	3 985
France	28	22	1 654	1 705

Provence-Alpes-Côte d'Azur	1	5	1	8
Corse	27	17	1 648	1 692
France, not allocated	0	0	3	4
Greece	32 439	5 180	6 631	44 252
Kentriki Ellada, Evvoia	6 531	1 969	0	8 500
Ipeiros	3 993	0	0	3 993
Peloponnisos	17 347	1 730	3 379	22 458
Nisia Aigaiou, Kriti	883	308	213	1 405
Kriti	3 410	277	356	4 044
Other Greek regions	266	885	2 598	3 750
Malta[a]	-	-	-	193
Italy	73 785	16 633	21 997	112 417
Piemonte	0	0	0	0
Liguria	7	17	3	28
Toscana (NUTS 2006)	6	0	0	6
Lazio (NUTS 2006)	399	82	178	660
Abruzzo	178	0	0	178
Molise	9	0	9	18
Campania	689	954	634	2 278
Puglia	3 462	146	4 059	7 668
Basilicata	4 640	39	2 093	6 774
Calabria	17 273	967	10 774	29 015
Sicilia	43 731	14 338	3 106	61 176
Sardegna	3 387	86	1 138	4 612
Portugal	12 416	494	3 235	16 145
Norte	734	52	133	920
Centro (PT) (NUTS95)	401	27	54	482
Lisboa e Vale do Tejo (NUTS95)	256	196	37	490
Alentejo (NUTS95)	1 585	11	247	1 844
Algarve	9 437	206	2 763	12 407
Spain	158 824	39 859	116 225	314 908
Principado de Asturias	0		0	1.00
Extremadura	278	0	38	317
Cataluña	2 080	20	10 777	12 877
Comunidad Valenciana	76 593	9 127	90 878	176 599
Illes Balears	660	397	98	1 156
Andalucía	64 158	5 646	9 999	79 804
Región de Murcia	14 514	24	4.433	43 509
Canarias (ES)	538	104	0	643

(*) = calculated. [a] Data for citrus production area for Malta are provided according to FAOSTAT (on line) for the year 2011.The detailed production structure is as follows: tangerins, mandarins, clementines (6 ha); grapefruit including pomelo (1 ha); lemons and limes (38 ha); oranges (95 ha); citrus fruit others (53 ha).

Pseudothecia formation and subsequent ascospore maturation and release in the Limpopo province of South Africa have been modelled by Fourie et al. (2013) using temperature sums and the moisture conditions in the leaf litter (resulting from rain, dew, or irrigation). Both ascospore germination and infection are driven by temperature and moisture conditions, where infection requires moisture in the specific form of a wet leaf surface for infection to occur (Kotzè, 1981). The requirements for ascospore germination on agar media varied between 15 and 29.5°C and 15 and 38 hours of wetness

(Kotzé, 1963). McOnie (1967) demonstrated that ascospores can infect when there have been at least 15 hours of continuous wetness, but no records of the temperatures were reported in this study. Timossi et al. (2003) evaluated the germination rate of ascospores of *Phyllosticta* spp. at different temperatures and incubation durations. The tested ascospores were produced on artificial media which, according to Baayen et al. (2002), are suitable only for ascospore production in *P. capitalensis* and not in *P. citricarpa*. No conclusive strain identification was provided by Timossi et al. (2003).

Temperature also influences the secondary infection cycle by determining the duration of the incubation period, symptom expression and consequently the formation of pycnidiospores on fruit lesions. Disease incidence and pycnidiospore production in naturally infected sweet orange fruit increased significantly at 27ºC compared to 20ºC. Light also augmented disease incidence and pycnidiospore production on fruit (Brodrick and Rabie, 1970). Field studies conducted in Brazil also showed that temperature was the main environmental factor affecting symptom expression (Ninin et al., 2013). Pycnidiospores are mainly disseminated by rain-splash (Whiteside, 1967) and are considered to be epidemiologically important in areas of Brazil (Sposito et al., 2007; 2008; 2011), where high rainfall frequently occurs during infection periods.

3.3.2.2. Review of the different methods used to assess the climatic suitability of the EU for *P. citricarpa*

Four methods have been employed, some in combination, when assessing climatic suitability of the EU for *P. citricarpa* establishment. This review gives a brief description of each method, lists the applications, describes the advantages and disadvantages and finally provides a conclusion concerning their applicability for the assessment of *P. citricarpa* climatic suitability in the EU.

(i) <u>Qualitative assessment based on the literature and expert judgement with or without model outputs</u>:

- Description of the method:
    - This has been the standard method of pest risk analysis since schemes were first developed in the early 1990s. It can be a general description of risk, e.g. EPPO (2007), or a detailed qualitative PRA scheme that requires a risk rating and an uncertainty score supported by a documented, referenced justification based on all the evidence including model outputs, e.g. EFSA Panel on Plant Health (PLH) (2010) and EPPO (1997; 2011). Risk ratings and uncertainty scores can be provided for each factor, e.g. climatic suitability, or just for each section, e.g. establishment.
- Applications:
    - The *P. citricarpa* datasheet in EPPO (1997) has a paragraph on phytosanitary risk to Europe based on a general review of the evidence without risk ratings and uncertainty scores.
    - The Prima phacie project (2011) assessed the risk posed by *P. citricarpa* to the EU based on the literature and the model evaluations and runs provided by the EFSA Panel on Plant Health (PLH) (2008) and answered the question: "How similar are the climatic conditions that would affect pest establishment, in the risk assessment area and in the current area of distribution? The risk was rated as moderately similar, with an uncertainty score of medium.
- Advantages:
    - it provides a clear written summary of risk and uncertainty that is based on the evidence presented and can be compared with other species
    - it integrates all the evidence available, not just the results from one model that will itself have uncertainties and often a range of plausible outputs
    - it is familiar to risk assessors and risk managers in the EU and elsewhere
    - it follows international guidelines (ISPM 11 by FAO, 2004) that do not stipulate that assessments should be quantitative

  - it follows the EFSA harmonised framework for pest risk assessment ((EFSA Panel on Plant Health (PLH), 2010).
- Disadvantages:
  - even if based on published data and model outputs, there are likely to be elements of subjectivity, e.g. due to inconsistencies between assessors in selecting appropriate risk ratings and uncertainty scores.
  - there can be a lack of transparency on how the different sources are combined and how risk ratings have been derived from the available information.
- Conclusions
  - this is a well recognised method for assessing risk that integrates model outputs and uncertainties with evidence from the literature
  - the results may depend on the assessor's subjective views
  - qualitative scores are often difficult to interpret

(ii) <u>Climate matching and correlative models</u>

- Description of the method
  - Climate matching methods, e.g. CLIMEX Match Climates, compare climates at one weather station or area with that in another using a variety of algorithms. Correlative models, e.g. MaxEnt (Elith et al., 2011) and BIOCLIM, use a wide variety of statistical methods or machine-learning techniques to assess climatic suitability. Classification rules are developed from the climatic variables at the locations where the pest is present and extrapolated to new areas.

- Applications
  - The CLIMEX Match Climates method (Sutherst et al., 2007) has been used for *P. citricarpa* by Paul (2006) evaluated by the EFSA Panel on Plant Health (PLH) (2008).
  - Climate response surfaces (Huntley et al., 1995)
    - Paul (2006) evaluated by the EFSA Panel on Plant Health (PLH) (2008).
- Advantages
  - Climatic matching methods are relatively simple to use and they provide preliminary indications of climatic similarity that can be used for further analysis.
  - The advantages of correlative methods are summarised by, e.g. Eyre et al. (2012). They are generally open access, relatively quick to use and the outputs are more likely to be consistent between different modellers.
- Disadvantages
  - The outputs of the climate matching methods expressed as climatic similarities, match indices etc are based on combinations of climatic variables and time periods that are unlikely to reflect the specific climate responses of the pest and the key periods during which they are important in the pest's life cycle.
  - Correlative methods greatly depend on: (a) the extent to which location data (for both presence and absence) are representative of the areas where the climate is suitable, (b) the climatic factors selected and (c) the methods for selecting thresholds for establishment (Dupin et al., 2011; Eyre et al.,2012).
  - The use of small presence/absence datasets may lead to inaccurate results (Dupin et al., 2011)
  - In both methods, the outputs are difficult to relate to pest biology and epidemiology.
  - The accuracy of the results of matching methods depends critically on the correctness of the assumption that physiological and ecological traits of organisms are identical between the area of origin and the area for which the potential for establishment is evaluated, and that these traits will remain unchanged over time. While this assumption of fixed traits is a valid null hypothesis to initiate the assessment, there are many examples of adaptation of invasive organisms to novel environments. The area for potential establishment will become larger than initial assessments would indicate

if an organism adapts to selective forces in a new environment. Therefore, in principle, matching methods have a fundamental weakness in demonstrating unsuitability of a geographic region for an organism, especially if a region is on the margin of suitability, posing oportunity for adaptation. In the case of *P. citricarpa*, there is very little information for diversity in ecophysiological traits, and its propensity for adaptation. Broadbent (1995) stated the following, indicating the risks of diversity and adaptation in the pathogen: "Black spot (caused by *Guignardia citricarpa* Kiely) causes serious losses in coastal orchards in New South Wales (Kiely 1948), but does not survive or cause symptoms in hot dry inland orchards (Barkley 1988). By contrast, black spot in South Africa was first reported in 1929 only in the cool misty areas of Natal, but in 1945 assumed more serious proportions when it spread to the hot dry subtropical East and North Transvaal (Wager 1952). Introduction to Australia of strains with a broader physiologic diversity could threaten export markets and reduce the viability of inland citrus".

- Conclusions
  - Climate matching methods are useful primarily as a preliminary guide and not for detailed analysis.
  - Given the paucity of representative location data and the complex relationship of the pest with climatic variables, it will be difficult to interpret the results of any correlative models applied to *P. citricarpa*.

(iii) Models combining correlative and deductive elements

- Description of the method
  - The CLIMEX Compare Locations model (Sutherst et al., 2007) can be parameterised by utilising a species' climate response data and by inference from its known distribution. The potential for establishment is based on the ecoclimatic index (EI) that combines a growth index, representing the suitability of the location for growth and development of the organism studied, and a stress index that is estimated according to the degree to which the climate is too wet, dry, hot, or cold. Once the parameters have been manipulated so that CLIMEX has satisfactorily emulated a pest's current distribution, EIs can be calculated from climatic data in the risk assessment area and mapped.

- Applications
  - CLIMEX Compare Locations
    - Paul et al (2005) evaluated by EFSA (2008)
    - Yonow et al. (2012) enhancing Paul et al (2005) and responding to EFSA (2008)
- Advantages
  - CLIMEX can integrate detailed climatic response data, e.g. temperature and soil moisture thresholds, with the climate in the area where the pest is present to mirror the current distribution that can then be projected onto the climate in the PRA area.
  - Yonow et al (2012) state that CLIMEX: "*is well suited to predicting the potential distribution of G. citricarpa because of the important influence of climate in the epidemiology of CBS*", but this argument is valid for any model taking into account climatic variables, not only for CLIMEX. This point is discussed further below.
  - Yonow et al (2012) also state that: "*CLIMEX has been successfully used to predict the potential distribution of other pathogens (Brasier and Scott, 1994; Venette and Cohen, 2006; Watt et al., 2011a,b; Yonow et al., 2004).*" However, as discussed below, the success of these predictions has not been systematically evaluated.
- Disadvantages

- EFSA (2008) noted that: "*it is difficult to reflect the relationship between pathogen infection and host phenology. All pest risk maps have to take into account the spatial presence of suitable hosts but, for many pathogens, temporal availability is also critical since infection may only occur if climatic conditions are suitable at specific host phenology stages. CLIMEX takes the whole year's climatic data into account so cannot readily be constrained to analyse just the period of suitable host phenology*". EFSA (2008) also noted "*the importance of complex variables, such as leaf wetness, that are not taken into account by CLIMEX and may act at a much shorter time scale (hours) than that utilised by CLIMEX (weeks for the moisture index)*". Yonow et al (2012) responded by stating that: "*EFSA (2008) argues that the climate during the period of host susceptibility alone should be considered, rather than the climate over the entire year. Whilst it is true that climatic conditions must be suitable at the appropriate time of host susceptibility for the presence of G. citricarpa spores to result in an infection, conversely, it is not true that a window of opportunity for host infection will necessarily lead to the permanent establishment of a population of G. citricarpa. An infection incident will not result in the establishment of a pathogen population unless the climate is suitable for the persistence of that population until the next infection incident can occur and a full life cycle can be completed. Suitably timed and repeated recurrence of such circumstances is required for there to be an opportunity for permanent establishment*". Yonow et al (2012) also state that "*it is true that CLIMEX does not consider the effects of a whole range of complex variables (which may or may not be driven by climate), such as leaf wetness, and it is true that the time scale at which a factor such as leaf wetness occurs is very short by comparison to the time scale at which CLIMEX operates. However, such issues are related to the first factor, where EFSA (2008) argues that only short periods of climate should be considered, and the counter-argument remains the same: short periods of suitability that may result in an infection incident will not necessarily result in the establishment and persistence of the pathogen*". The Panel agrees with Yonow et al. (2012) that modelling infection alone is insufficient. However, the climate (primarily temperature) not only also has to be suitable for development and spore production but the timing of spore release also has to coincide with key stages in host phenology. For successful establishment, suitable hourly temperature and leaf wetness conditions required for infection to take place need to coincide with the availability of inoculum (i.e. spore presence) and host phenology (i.e. citrus hosts in a susceptible phenological stage). This complex combination of climatic factors and host phenologies requires models such as those proposed by Fourie et al (2013) and Magarey et al (2005) that, unlike CLIMEX, can operate at a high temporal resolution related to the timing of key epidemiological events, utilise parameters such as leaf wetness and can be constrained to interact with host phenology.
- EFSA (2008) noted that there are "*discrepancies between the pathogen and host's climatic responses. The pathogen's climatic responses may be much greater than the range suitable for the host*" Yonow et al (2012) state that "*a pathogen and its host may indeed have differing climatic responses. In the case of G. citricarpa and citrus, there is evidence in both South Africa and Australia that despite the extended absence of restrictions on the movement of citrus propagation material from CBS-infected areas into CBS-free areas, the disease has never established in these areas. These areas are thus evidently climatically suitable for citrus, but unsuitable for G. citricarpa. The current CLIMEX model predicts correctly that several citrus regions are unsuitable for the long-term persistence of G. citricarpa and it also predicts potential climatic suitability for G. citricarpa in some parts of the world that are not suitable for citrus production. Our model therefore appropriately provides for differentiation between potential distribution of the host and pathogen*". The ecoclimatic index calculated by Yonow et al. (2012) for *P. citricarpa* is highest in areas of Europe, e.g. southern Romania, where the winters are too cold for

commercial outdoor citrus production. The Panel accepts that species distribution models may predict potential establishment based on climate in areas that are not climatically suitable for their host. Such discrepancies highlight the importance of taking host distribution into account when assessing the area of potential establishment.

- the successful use of CLIMEX in predicting the potential distribution of pathogens is subjective and has never been properly analysed. The "success" of a model in projecting the distribution of any organism, whether or not it is a pathogen, depends on many factors, e.g. the complexity of the life cycle, the extent to which distribution is dependent on climate and whether the key climatic factors are represented in CLIMEX and are available at an appropriate spatial and temporal resolution. In addition, the volume, quality and spatial distribution of locations where the pest is known to be present (Eyre et al., 2012) and the extent to which the pests is known to have high/low incidence at these locations are also important. Moreover, the extent to which CLIMEX has been successful in predicting the potential distribution of the pathogens may be difficult to evaluate due to limited evidence. The paper by Brasier and Scott (1994) is particularly difficult to assess because they modelled a root pathogen (*Phytophthora cinnamomi* Rands) that lives in an edaphic microclimate that is very different from that measured by weather stations and did not provide the model parameters and justification for their selection. Model parameterisation and outputs are strongly influenced, not only by the availability of reliable climatic response data and representative presence data, but also by the likelihood of continuing spread and disjunct distributions. The distribution of citrus and therefore CBS in South Africa and Australia is highly disjunct and is also affected by major geographical features (principally the sea) and irrigation. This makes it difficult to determine with confidence the factors that are critical in setting the limits to the distribution of *P. citricarpa*.
- Although Yonow et al (2012) state that: "*Climatic suitability can be broadly categorised as follows: EI = 0 (unsuitable), 1 ≤ EI ≤ 4 (marginal), 5 ≤ EI ≤ 9 (suitable), 10 ≤ EI ≤ 29 (highly suitable), and 30 ≤ EI (optimal)*", classifying outputs into marginal, suitable and optimal is difficult and species specific. Stephens et al (2007) stated that: "*The assignment of classifications to EI values is usually an arbitrary process, as the resulting patterns are species-specific*". Sutherst et al. (2004) provide some suggested guidelines: "a*n EI = 0–0.49 indicates that the climate is unsuitable; the species cannot persist in an area under average environmental conditions, an EI of 0.50–9.99 indicates marginal conditions, an EI of 10– 19.99 indicates suitable conditions and an EI of 20+ indicated optimal conditions. An EI of 100 indicates that conditions are perfect all year round, and there are few environments that are stable enough to provide perfect habitat year round*". Baker et al. (2011) stated that the ecoclimatic index "*can be classified by looking at where the pest is: (a) present but with very low populations, (b) present but not abundant and (c) generally abundant and if (a), (b) and (c) are clearly primarily influenced by climate and not other factors they can be used to classify the EIs. EI values close to zero can be considered marginal, and we would generally expect that a species distribution in climatically marginal habitats would be patchy, and restricted to more climatically favourable sites. In this zone, we would also expect that a species presence would be patchy in time, and metapopulation dynamics might play a strong role in maintaining its presence on a regional basis. If the EI, which is scaled from 0-100, is greater than 30, the climate can generally be considered to be very favourable for establishment (Sutherst et al, 2007; Pinkard et al., 2010). However, the maximum climate suitability that a species can experience under field conditions depends upon the interplay between the seasonality of temperature and moisture variables and the individual species' climatic niche. In climatic terms, it is possible to have too much of a good thing. As noted by Brown (1998), biotic factors tend to define a species range where*

resources are abundant. These factors underline why the climate suitability classification needs to be considered on a species-specific basis".

- Fitting the distribution simulated by CLIMEX to the actual distribution of the organism by the iterative adjustment of parameters can be difficult and can lead to difficulties of interpretation if the values selected are significantly different from those in the literature. As noted above, a key advantage of CLIMEX, compared to other species distribution models, is that it can be parameterised with climatic response data that have been published on the species of interest. For example, the minimum temperature threshold for development is available for many species (Jarosik et al., 2012) including some data on certain life cycle stages of *P. citricarpa* (Kotzé, 1963; 1981). All parameters, both those that have been obtained from the literature and those, such as the stress indices, that are inferred from the species distribution can be modified by a process of iteration to match the distribution simulated by CLIMEX with the known distribution. Where there are no published data, the modification of parameters has few constraints. Departing from published climate response thresholds is justified when there is considerable uncertainty, experimental data vary or there is evidence that data obtained from lab experiments do not accurately represent field conditions. Since the literature on the minimum temperature threshold for development of *P. citricarpa* as summarised by Yonow et al. (2013) does not provide one clear value there is considerable scope for parameter variation. Nevertheless, the published literature all point to a threshold at or below 15°C (though one unpublished South African report states that subsequent infection has not been observed at these temperatures). However, Yonow et al (2013) have selected a threshold of 20°C justifying the much higher temperature solely on the basis that this was the only way they could find of excluding the simulated distribution of *P. citricarpa* from the Western Cape Province of South Africa where the disease is absent. The decision to select a minimum temperature threshold for development that is considerably outside the published range makes their model results very difficult to interpret.

- Conclusions
  - CLIMEX Compare Locations can provide misleading results for this species because of the lack of data from sites where the pest is marginal, the difficulty of addressing key events in the life cycle of the pathogen and their relation to host phenology together with the short time scale over which some key events in the life cycle operate..

(iv) <u>Deductive models (generic infection, leaf wetness and temperature models)</u>

- Description of the method
  - These models focus on the key processes in the life cycle that determine whether the life cycle can be completed and perpetuated. Phenology models, based on degree days, are often used to determine whether there is sufficient temperature above the minimum threshold to complete development. For foliar fungal pathogens typically moisture, in addition to temperature, is modelled to determine whether conditions are suitable for spore development, release and germination.
- Applications
  - generic infection (temperature and leaf wetness) models
    - Magarey & Borchert (2003) using the generic infection model
    - EFSA (2008) using the generic infection model (Magarey et al., 2005)
    - Magarey et al. (2011) using the generic infection model (Magarey et al., 2005)
  - inoculum production and release models (combined temperature and moisture models: degree day models with or without moisture restriction to predict the release of ascospores)
    - Fourie et al. (2013) contradicting EFSA (2008)

- Advantages
  - The models directly simulate key processes in the pathogen life cycle on which establishment depends
- Disadvantages
  - The models need very high temporal resolution climatic data. Leaf wetness (required for the generic infection model by Magarey et al., 2005) is not commonly measured at meteorological stations.
  - The models are difficult to parameterise because they need experimental data to estimate the minimum, optimum and maximum temperatures for successful infection, the minimum and maximum wetness durations for successful infection and the tolerance to short dry periods. Only limited experimental data are available to estimate the parameters of the Magarey et al. (2005) model for *P. citricarpa*.

    The timing of life cycle events must be closely related to host phenology to help predict the likelihood of establishment.
- Conclusions

It was concluded that a combination of model based assessments could give a better insight into the risk of establishment of *P.citricarpa* in the EU territory. Three models have been used: (1) a model by Fourie et al. (2013) describing the timing of pseudotheticia maturation in *P. citricarpa*; (2) a model by Fourie et al. (2013) describing the seasonal time course of ascospore release; (3) the model by Magarey et al. (2005) describing when environmental conditions (temperature, humidity) are suitable for infection. The results of these three models have been combined with records and expert knowledge on the phenology of susceptible host tissues. The overall conclusions have been based on a qualitative assessment of the establishment potential following the EFSA guidance document (EFSA Panel on Plant Health, 2010).

3.3.2.3. Analyses of climate suitability done by the Panel

The suitability of the environment was analyzed by the Panel mainly using two different types of model simulations:

- Simulations of pseudothecium maturation and ascospore release with the models of Fourie et al. (2013) (3.3.2.4)

- Infection simulations with the generic infection model of Magarey et al. (2005) (3.3.2.5)

Environment suitability was evaluated from these simulations and from the periods of susceptibility of citrus leaves and fruits derived from the scientific literature and from technical documents (see section 3.3.1.1).

In addition, the Panel undertook a limited investigation of the CLIMEX model parameterization for *P. citricarpa* done by Yonow et al. (2013) (3.3.2.6).

3.3.2.4. Simulations of pseudothecium maturation and ascospore release

Fourie et al. (2013) parameterized models to predict pseudothecium maturation and the onset and seasonal course of ascospore discharge of *Phyllosticta* spp. (*P. citricarpa* and *P. capitalensis*). These models were previously developed for the pear scab pathogen, *Venturia pyrina* Aderh., by Rossi et al. (2009). The models of Fourie et al. (2013) were fitted to ascospore trap data collected in the Limpopo province of South Africa. The authors compared several variants of their models and finally recommended two models:

- A model based on a Gompertz equation predicting the onset of ascospore release as a function of degree-day accumulation from daily weather data using mid-winter (i.e. January 1[st] in the northern hemisphere and July 1[st] in the southern hemisphere) as the biofix and 10°C as the base temperature (further referred to as **Model 1**). Time of onset is defined in this model as the moment at which the probability of spore discharge on days that are suitable for such discharge (3-day cumulative rainfall >0.2mm or vapour pressure deficit <5hPa) pass a predefined threshold. Fourie et al. (2013) recommend thresholds of 0.5 and 0.7. The capture of spores on days that are suitable for spore release is thus used as evidence that the pseudothecia are mature.

- A model based on a Gompertz equation predicting the cumulative proportion of ascospores trapped per season as a function of degree-day accumulation only on days with measurable rainfall (>0.1mm) or vapour pressure deficit <5hPa) (further referred to as **Model 2**).

- Model 1 was run by the authors for three localities in Europe (Valencia SP, Messina IT, Pontecagnano IT) using average monthly climatic data. According to Fig.1 of Fourie et al. (2013), the onset of ascospore release would occur between May and June in Valencia and in Messina, and between June and July in Pontecagnano, based on the probability thresholds of 0.5 or 0.7. However, the between-year variability in the onset of ascopore release was not investigated and the uncertainty of the model prediction was not analysed by the authors. Fourie et al. (2013) concluded that the bulk of ascospores in Mediterranean-type climates would most likely be released during the dry summer months, but did not run Model 2 to predict the dynamics of ascospore release for any European location.

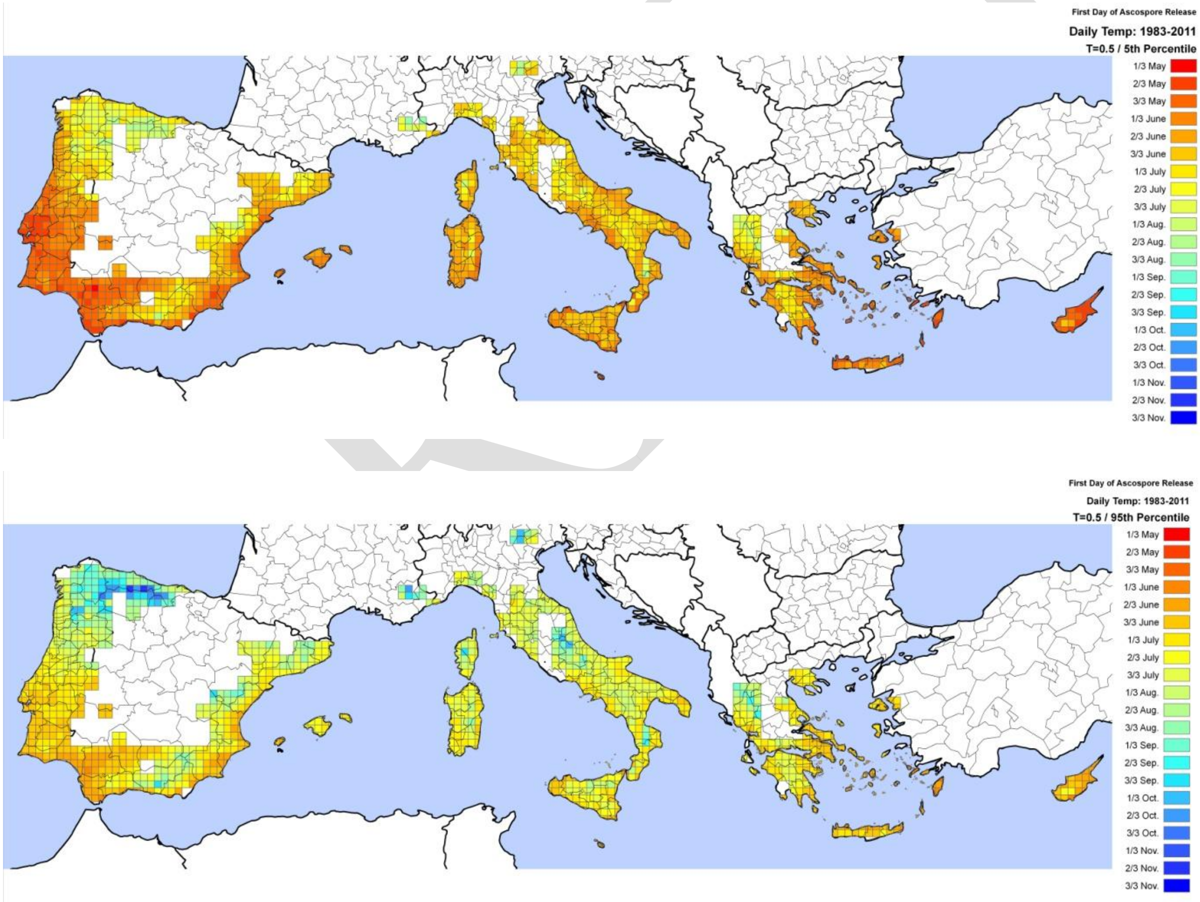


Figure 22: Onset of ascospore release predicted by Model 1 (Fourie et al., 2013) for a 25km-grid interpolated climatic data for the EU citrus-growing areas from 1983 to 2011 (Probability threshold set to 0.5 and the upper and lower map shows respectively the 5th and 95th percentiles of the results for the 29 years).

Model 1 from Fourie et al. (2013) was run by the Panel with daily weather data interpolated to a 25km-grid for the EU citrus-growing areas to predict the potential onset of ascospore release in these locations. The 0.5 and 0.7 thresholds were evaluated using a weather dataset consisting of daily data from 29 consecutive years (1983-2011). The results of the simulations using the 0.5 probability threshold are shown in Fig. 21, other outputs are included in the appendix. The model predicted the onset of ascospore release from the beginning of May to the end of June, depending on locations and years. In general, coastal citrus-growing regions showed an earlier onset of ascospore release compared to inland areas. The areas predicted to have May as the dominant period (50 percentile) for the onset of ascospore release are Cyprus, Malta, some of the islands in Greece and some areas in sourthern Spain. Model 1 was also run for eight agrometeorological stations located in citrus-growing regions in Italy (Caronia Buzza, Lentini, Mineo, Misilmeri, Paterno, Ribera, Riposto and Siracusa) to obtain the biofix to run Model 2 and predict the subsequent dynamics of ascospore release.

3.3.2.5. Infection simulations with the generic infection model of Magarey et al. (2005)

This model requires estimates of the three cardinal temperatures (T_{max}, T_{min}, T_{opt}), of two wetness duration thresholds (W_{max}, W_{min}), and a parameter describing tolerance to dry interruptions (D_{50}). It computes the leaf surface wetness duration requirement for infection. The parameter values were estimated by EFSA (2008; table 3, page 36) based on published experiments on germination or infection by pycnidiospores and ascospores separately. Studies on the infection efficiency of *P. citricarpa* ascospores under different combinations of temperature and wetness durations are not available. Thus, parameters values were mainly obtained from published data on ascospore germination and mycelial growth. A sensitivity analysis carried out by EFSA (2008) indicated that model uncertainty was mainly due to the parameters D_{50} and $T_{min.}$ EFSA (2008) set the T_{min} to 15ºC, based on the studies by Kotzé (1963) who reported germination of *P. citricarpa* ascopores at this temperatures. However, lower temperatures were not tested in this experiment and the possibility of infection below 15ºC cannot therefore be excluded. The value of T_{max} was set at 35 ºC, as indicated by Magarey et al. (2005) when there is no information on the upper temperature limit for infection, as is the case for *P. citricarpa*. With regard to T_{opt}, Kotzé (1963) obtained the highest germination rate at 29.5 ºC, which was also the highest temperature tested. The optimal temperature for the growth of *P. citricarpa* on liquid basal synthetic medium is 27 ºC (Kotzè, 1981) and the optimal temperature for hyphal growth is 25-28ºC (Chiu, 1955). Therefore, EFSA (2008) used a value of 27 ºC for $T_{opt.}$ McOnie (1967) demonstrated that ascospores can infect with at least 15 hours of continuous wetness. This value is supported by Kotzé (1963), who obtained 15.7% germination of ascospores after 15 hours of incubation at 29.5 ºC, showing consistency between germination and infection data. Thus, EFSA (2008) set the value of W_{min} to 15 hours. A W_{max} value of 38h was selected by EFSA (2008) according to the results of Kotzé (1963). No information was found in the literature on the sensitivity of *P. citricarpa* to dry interruptions during infection, so D_{50} was set to 3 hours as a value which is often found in the literature as being a generally acceptable period of leaf wetness interruption (Xu and Butt, 1993; Rossi et al., 2007). The parameters of EFSA (2008) were later validated by Magarey et al. (2011). The model was then run by EFSA (2008) with climatic data interpolated to a 50km grid for the EU citrus growing areas with simulated wetness data (Bregaglio et al., 2010, 2011) and with agro-meteorological station data (14 Spanish stations and 10 Italian stations) equipped with on-site wetness sensors. The model used by EFSA (2008) predicted numerous pycnidiospore and ascospore infection events over a ten year period (1998-2007) at agro-meteorological stations and 50 km grids. With the gridded data, almost no infection events were predicted in summer (June-August) but significant numbers of events occurred at many locations in the spring and autumn (see Fig. 23 for ascospore infection). In general, data from agro-meteorological stations followed the same pattern, although with a somewhat longer infection period reflecting microclimate variability.

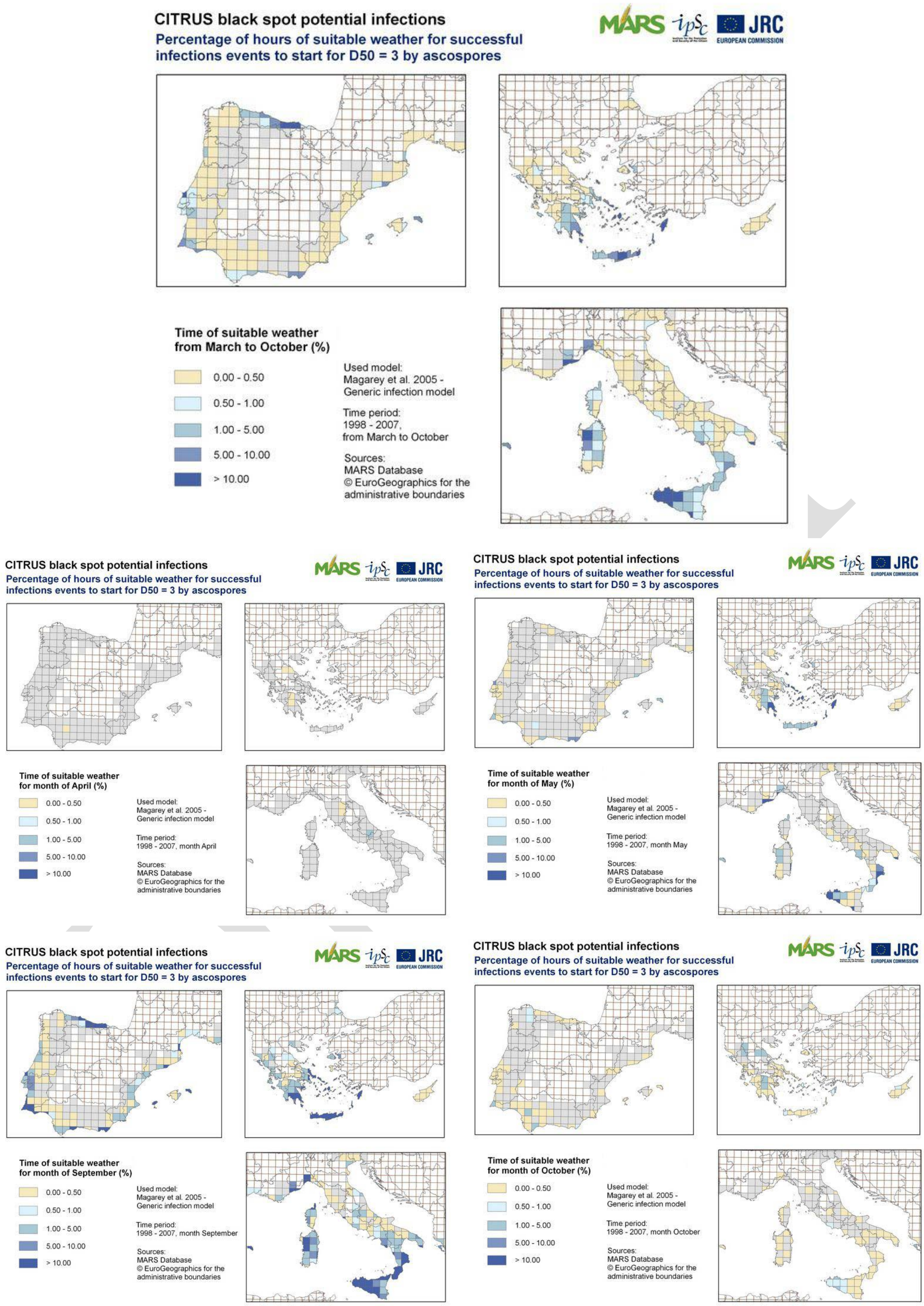


Figure 23: Percentage of hours with weather conditions suitable for successful infection events by ascospores in April-October, April, May, September and October (JRC 2008, EFSA Journal (2008) 925,1-108)

For all studies where weather or climate data are used to assess the suitability of environment for a pest in a new area, it is a prerequisite that the weather data should be recorded under standard meteorological conditions in order to be comparable. Standard meteorological conditions mean that measurement equipment is placed in an open field with a standard cover of a grass lawn kept cut short on a regular basis. The sensors measuring air conditions such as air temperature, wind speed and relative humidity should be placed at the standard height of 2 metres above ground. When these data are used as inputs to model based simulations, in which the key parameter values originate from laboratory experiments where the measurements underlying the estimates of environmental conditions are not recorded under standard meteorological conditions, this can cause error and introduce additional uncertainty into the model results. Taking air temperature as an example, the air temperature close to citrus leaf litter lying on the ground, or close to the surface of a living citrus leaf, can differ by several degrees to the air temperature recorded at the same time at a nearby meteorological station where the air temperature is recorded 2 metres above ground in an open field. Ribeiro et al. (2005) evaluated daily and seasonal changes of leaf temperature in relation to the variation of meteorological elements (global radiation and air temperature) and air vapour pressure deficit in field-grown citrus plants and recorded differences between leaf and air temperatures up to 8°C.

3.3.2.6. Conclusions derived from the models by Fourie et al 2013. (Model 1 and 2) and by (EFSA 2008) results from applying the model by Magarey et al. (2005).

After the successful transfer of *P. citricarpa* to susceptible citrus leaves and fruit in the PRA area, the pathogen may then reproduce through ascospores in infected leaves and pycnidiospores in infected fruit, twigs and leaves. Since ascospores can be disseminated at relatively long distances by air currents (Kotzé, 1981), a potential epidemic development of CBS in the EU citrus-growing areas would mainly be driven by the duration and efficiency of ascospore infection, their reproduction and dissemination rate and the host availability and environmental conditions that allow symptoms to develop on the fruit.

Nevertheless, the importance of pycnidiospores has recently been shown in CBS epidemics in Brazil (Spósito et al., 2007; 2008; 2011). Pycnidiospores were also considered important during the early stages of CBS epidemics in Zimbabwe and Argentina, where no ascospores were detected in the leaf litter and the disease was limited to a low number of foci with symptoms occurring each year on the fruit from the same trees and even on fruits from the same part of of individual trees (Whiteside, 1967; Garrán, 1996). Therefore, the potential role of pycnidiospores in CBS epidemics in the risk assessment area cannot be completely excluded. Warm temperatures and high soil moistures have been shown to enhance citrus leaf litter decay in South Africa and Taiwan (Kotzé, 1963; Lee and Huang, 1973), limiting further pseudothecia and ascospore development. Specific experiments on citrus leaf litter decomposition under semi-arid conditions are not available, but general studies on leaf litter decomposition indicate that the decomposition rate increases with mean annual temperature and precipitation, mainly due to the enhanced activity of the decomposer organisms (Zhang et al., 2008). Pseudothecia and ascospores are produced in the leaf litter after periods of alternate wetting and drying (Kiely, 1948; Lee and Huang, 1973; McOnie, 1964). The extensive use of irrigation in the EU citrus-growing areas (Section 3.3.3.1) will add to the suitability of the environment since it lengthens the periods of leaf wetness aiding infection.

Figure 23 and figures 48-55 in the Appendix C shows, for 8 Italian stations and 6 years, preliminary results on the monthly dynamics of ascospore release predicted for each station with the two Fourie et al. (2013) models, together with the average proportion of hours with environmental conditions favorable for ascospore infection predicted by the Magarey et al. (2005) model as described in EFSA (2008) with D_{50}= 3. Data on potential ascospore release were obtained on a daily basis whereas hourly estimates of the weather conditions for infection were produced. Monthly summaries of the outputs from both models have been presented for clarity.

When running the ascospore maturation and release model (Fourie 2) it was observed that a minor proportion of the spores would not mature within one growing season, and would not be released until the following season. This might be a consequence of extrapolating the models to a markedly different climatic region. Although there are no published studies about the decomposition time of citrus leaf litter under semi-arid conditions, data from other climatic regions indicate that it is unlikely that fallen leaves will maintain their integrity as a substrate for inoculum production for such a long period (Lee and Huang, 1973; Mondal and Timmer, 2002; Mondal et al., 2003). Therefore, only predictions for the first year are shown in Figure 23 and figures 48-55 in the Appendix C.

The graphs indicate that there is generally an overlap between potential ascospore release and the weather conducive to infection with peaks in September and October when susceptible leaves and fruits are widely available in the risk assessment area (Section 3.3.1.1). From these results, it can be concluded that the climate in the risk assessment area would sustain the reproduction, dissemination and infection of *P. citricarpa*, at least at some European locations.

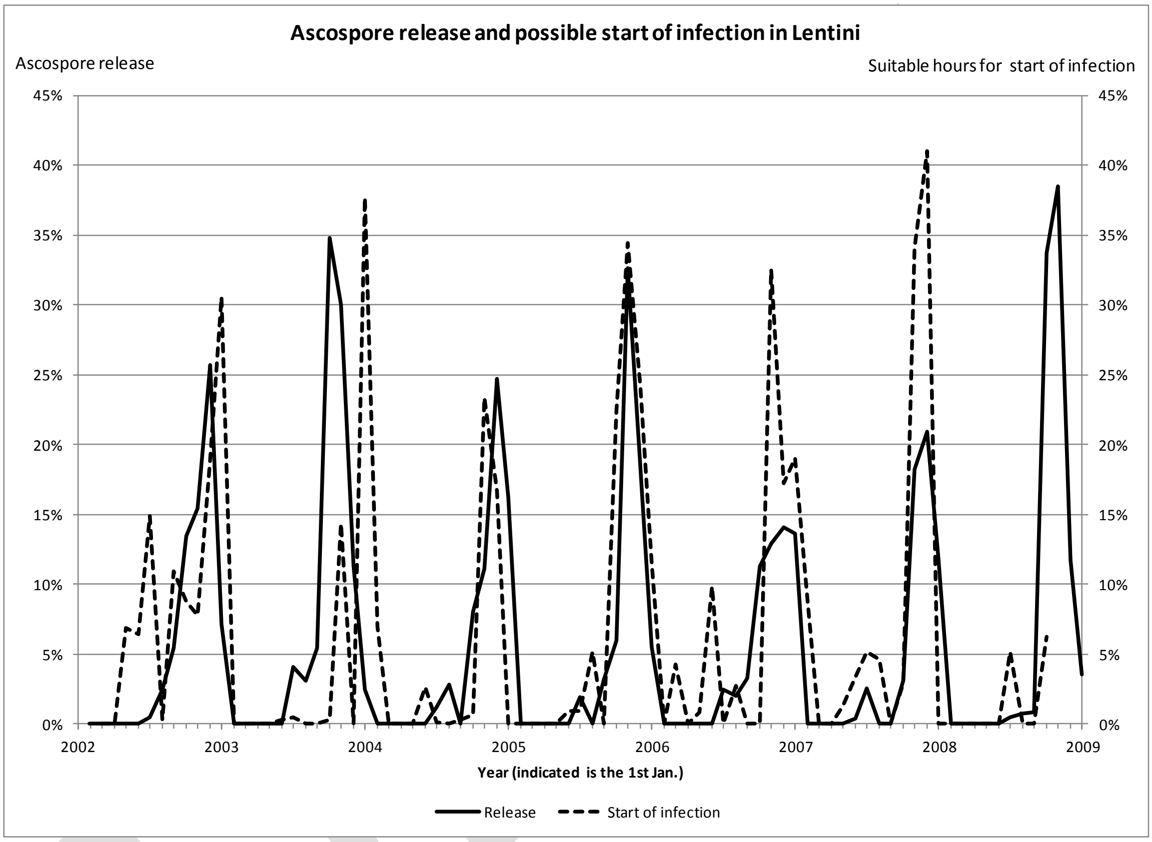
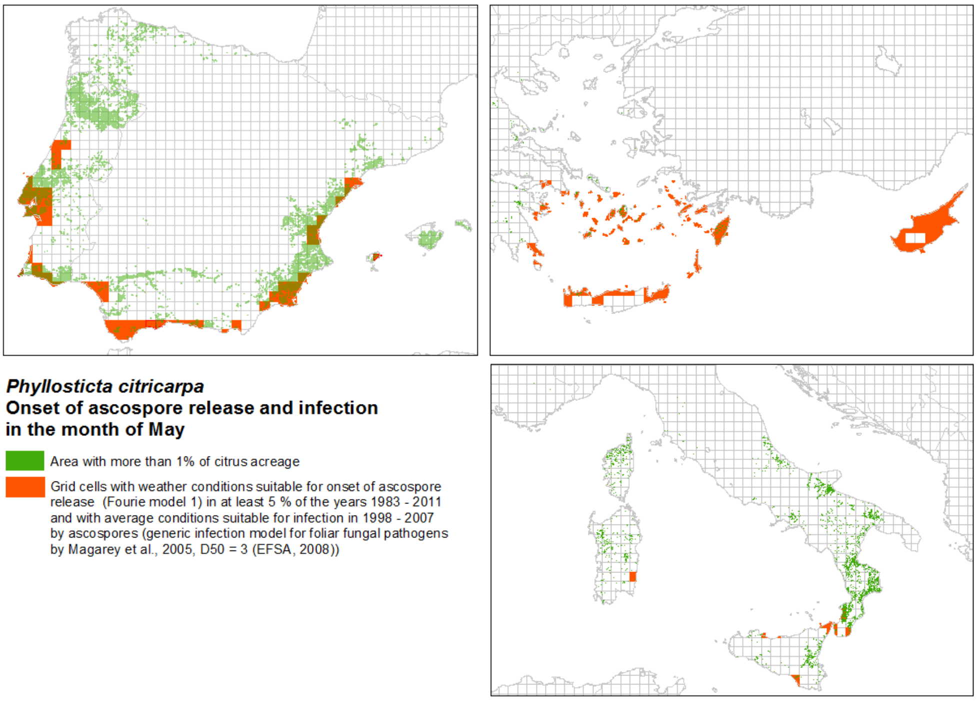


Figure 24: Comparison of the dynamics of ascospore release (2002-2007) and average percentage of hours suitable for start of a successful infection by *P. citricarpa* ascospores (2002-2008) predicted for the station of Lentini in Italy.

A.



B.

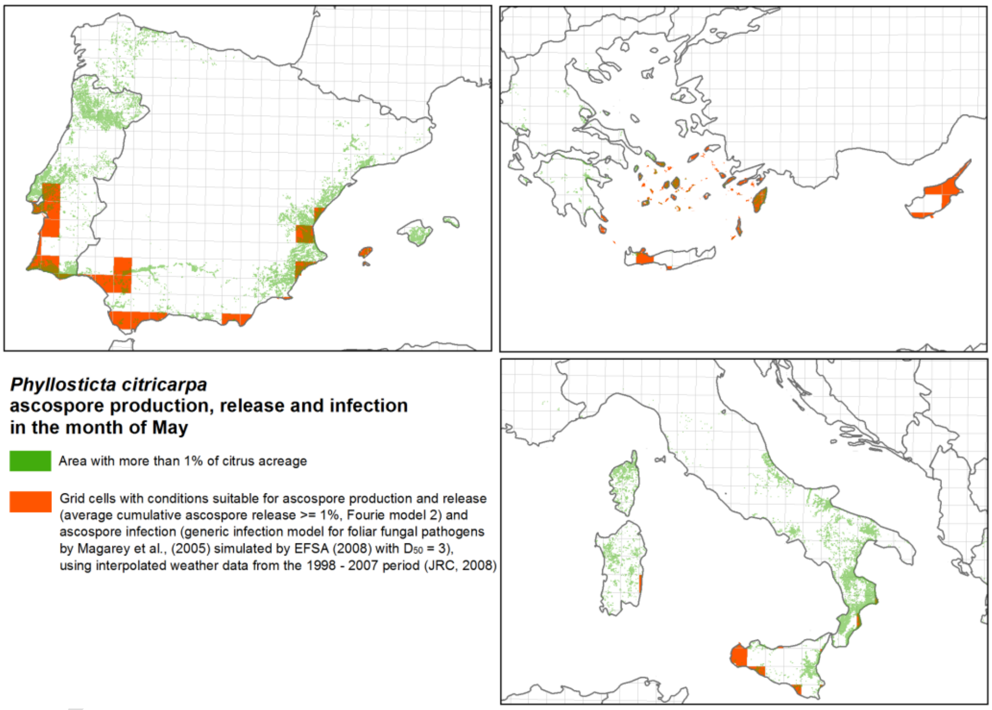


Figure 25: Areas predicted to be at risk of *P. citricarpa* epidemics to begin in spring (May) due to weather conditions conducive for inoculum production (Fourie 1 and Fourie 2 models) and infection (EFSA, 2008). Figure 25A shows (in red) where the onset of ascospore release could occur in May according to the Fourie 1 model (5th percentile of the years 1983 - 2012) based on interpolated weather data to a 25 km x 25 km grid and where the climatic conditions are also suitable for infection (number of hours suitable for infection >0) according to the EFSA (2008) simulation results applying the "Generic infection model for foliar fungal pathogens" (Magarey et al., 2005). In Figure 25B, the red areas correspond to areas having periods suitable for infection according to the simulations from the generic infection model fitted for *P. citricarpa* (Magarey et al., 2005), and where inoculum is

available by the release of ascospores equal or greater than 1% of total spore release during the year according to the Fourie 2 model (average for the period 1998-2007).

These results are confirmed by the maps displayed in Figures 25 and 26 where the outputs of the Magarey et al. (2005) model and of the two Fourie models were superimposed with the principal European citrus growing areas. Figure 25A shows the European areas (in red) where the onset of ascospore release could occur in May according to the Fourie 1 model and where the climatic conditions are suitable for infection according to the Magarey model (number of hours suitable for infection >0). Figure 25A shows that infection may be able to start in several areas of Europe with a Mediterranean climate. However, Figure 25B shows that the percentage of ascospore release is likely to be lower than 1% in most of these areas, with only a few exceptions in Portugal, Spain, Sicily and Crete when it can happen in some years. Overall, these results show that, in May, infection could probably occur only at a limited number of locations, due to the production of limited quantities of ascospores in most parts of the EU.

The risk of infection is much higher in September and October according to the results displayed in Figure 26. In September, the percentage of ascospore release simulated by the Fourie 2 model was found to be higher than 1% in most areas where the percentage of hours with suitable climatic conditions was greater than zero. Conditions were slightly less favourable for infection in October but, overall, the percentage of ascospore release was also higher than 1% in most areas with climatic conditions suitable for infection during this month.

Although the scenario considered in Figures 25 and 26 is a worst-case scenario (with a low threshold for the onset of ascospore release), the results of the Magarey et al. (2005) and Fourie et al. models show that infection cannot be excluded in Europe especially in September-October.

Uncertainty: The results of the Fourie et al. (2013) models should be interpreted with caution for several reasons and the Panel considers that there is a high uncertainty related to these predictions because:

- The models were developed and evaluated in the Limpopo province of South Africa, a region characterized by a summer rainfall pattern. The capability of the models to predict ascospore release in other areas has not been investigated.

- Although the standard deviations of the parameter estimations are reported in Table 5 by Fourie et al. (2013), the consequences of the uncertainty concerning the parameter values on model predictions were not analysed by the authors.

- The results presented in Figure 1 of Fourie et al. (2013) are based on average monthly climatic data. The authors do not report on the between-year variability of ascospore release.

- Model 2 was not used by the authors to predict the proportion of ascospore release in Europe.

- Rossi et al. (2009) used a base temperature of 0 ºC to calculate degree days but Fourie et al. (2013) chose 10 ºC. With this higher base temperature, negative values are obtained for some days in many Mediterranean locations. The Panel adopted the general practice of considering negative values as zero values in the degree days calculation (De Wit, C.T. & J. Goudriaan, 1978).

- The models were fitted to ascospore trap data consisting of a mixture of two species, *P. citricarpa* and *P. capitalensis*, in unknown proportions.

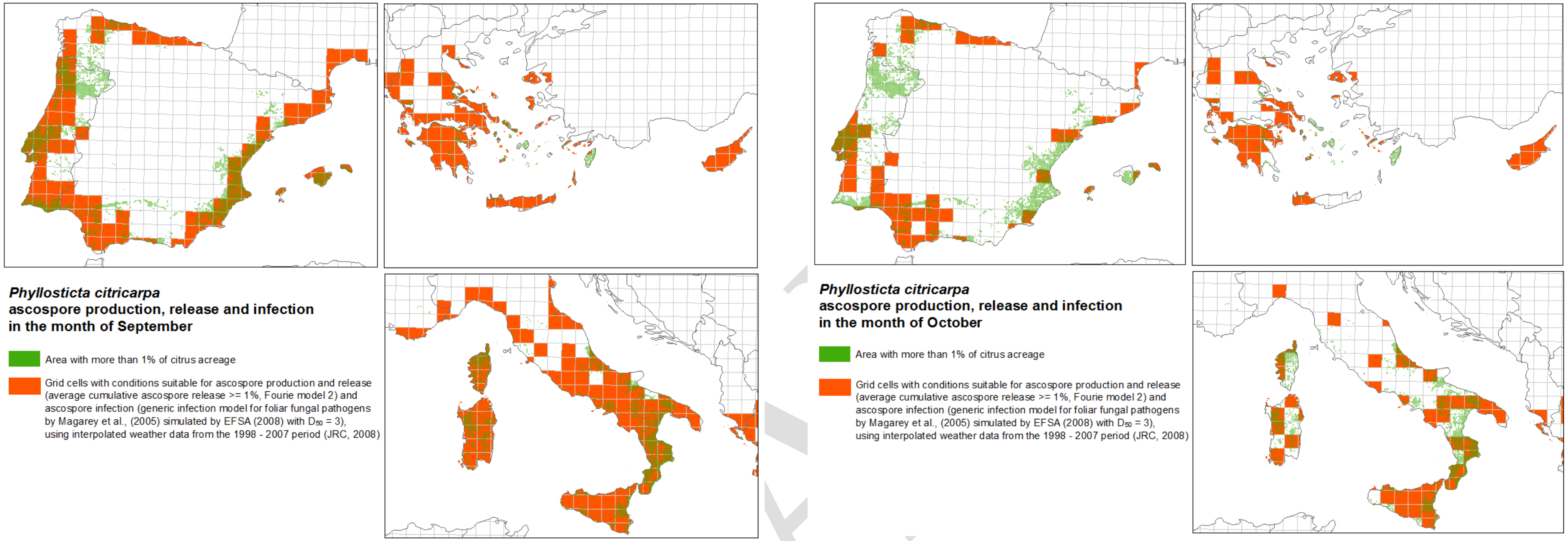


Figure 26: The risk of *P. citricarpa* infection in September (left panel) and in October (right panel) according to the infection model results and model predictions of inoculum availability (Fourie 2). The red areas correspond to areas where there are both periods suitable for infection according to the simulations from the generic infection model fitted for *P. citricarpa* (Magarey et al., 2005), and inoculum available by the release of ascospores equal or greater than 1% on average for the period 1998-2007 according to the Fourie 2 model.

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Experiments carried out to determine the temperature and wetness duration requirements of *P. citricarpa* were reviewed by EFSA (2008) showing that, due to the scarcity of experimental data available, there is a high uncertainty concerning the values of the parameters describing the climatic requirements for infection. The minimum and optimum temperatures for infection and the degree of tolerance to dry periods were considered as highly uncertain by the PLH Panel (see section 2.3.2 of EFSA, 2008). The uncertainty analysis performed by EFSA (2008) for the Magarey et al. (2005) generic infection model of using agro-meteorological station data showed that the simulations of infection were highly uncertain (see 2.3.5.3 of EFSA, 2008). As no new experimental study has been performed to estimate the parameters of the infection model since 2008, the Panel considers that the level of uncertainty concerning these aspects is unchanged.

The Panel concluded that the only way to further reduce the uncertainty concerning the climatic requirements of *P. citricarpa* would be to conduct new experiments in order to determine more precisely the temperature and wetness duration requirements of this fungus. Based on the sensitivity analysis presented in EFSA (2008), the parameters that have the strongest influence on the wetness requirements calculated by the wetness model of Magarey are the minimum temperature requirement for infection and the degree of tolerance of the dry period (see Table 6 in section 2.3.5.3 of EFSA, 2008) are. It would thus be useful to carry out experiments to estimate these parameters more accurately.

3.3.2.7. CLIMEX model parameterized to model the potential global distribution of the citrus black spot disease by Yonow et al. (2013)

In 2008, the EFSA Panel on plant health evaluated a pest risk assessment for *Guignardia citricarpa* conducted by South Africa (Hattingh et al., 2000) and additional supporting material (Paul et al., 2005; Paul 2006). In its evaluation the EFSA (2008) scientific opinion expressed concerns about the appropriateness of applying the CLIMEX modelling approach underpinning the pest risk assessment for *Guignardia citricarpa* conducted by South Africa. These concerns expressed by EFSA (2008) were recently challenged by Yonow et al. (2013) who published a new set of CLIMEX parameters for *P. citricarpa* in order to model the potential global distribution of the pathogen with a particular focus on the risk posed to Europe.

In the preparation of this scientific opinion the Panel explored the basis for the arguments raised by Yonow et al. (2013) (see section 3.3.2.2). In addition, the Panel analysed the sensitivity of the CLIMEX model outputs for Europe to climate data inputs at different spatial resolutions and time periods (see Figure 27).

To display the results of their CLIMEX model, Yonow et al. (2013) used a 0.5° latitude x 0.5° longitude grid with interpolated monthly 1961-1990 climatic data. When this is substituted by a higher spatial resolution (0.1° latitude x 0.1° longitude) 1961-1990 climatology (New et al., 2002) (see Figure 27c) a larger area of citrus production in Europe is suitable for *P. citricarpa* based on the classification utilised by Yonow et al. (2013). When the Yonow et al (2013) CLIMEX model is run with more recent climate data for 1998-2007 (JRC, 2008) at a different spatial resolution (25 km x 25 km), a larger area is predicted as being suitable (Figure 27d) and some areas have a higher EI than that predicted for the period 1961-90. According to the classification of the EI by Yonow et al (2013), one area is even predicted to be highly suitable. This corresponds to the area of the Ebro delta in eastern Spain where the northernmost commercial citrus production in the country takes place. Overall, it can be concluded from these analyses that the suitability of climate for *P. citricarpa* as predicted by the CLIMEX model parameterised by Yonow et al (2013) is very sensitive to the spatial resolution and time period of the climate data inputs.

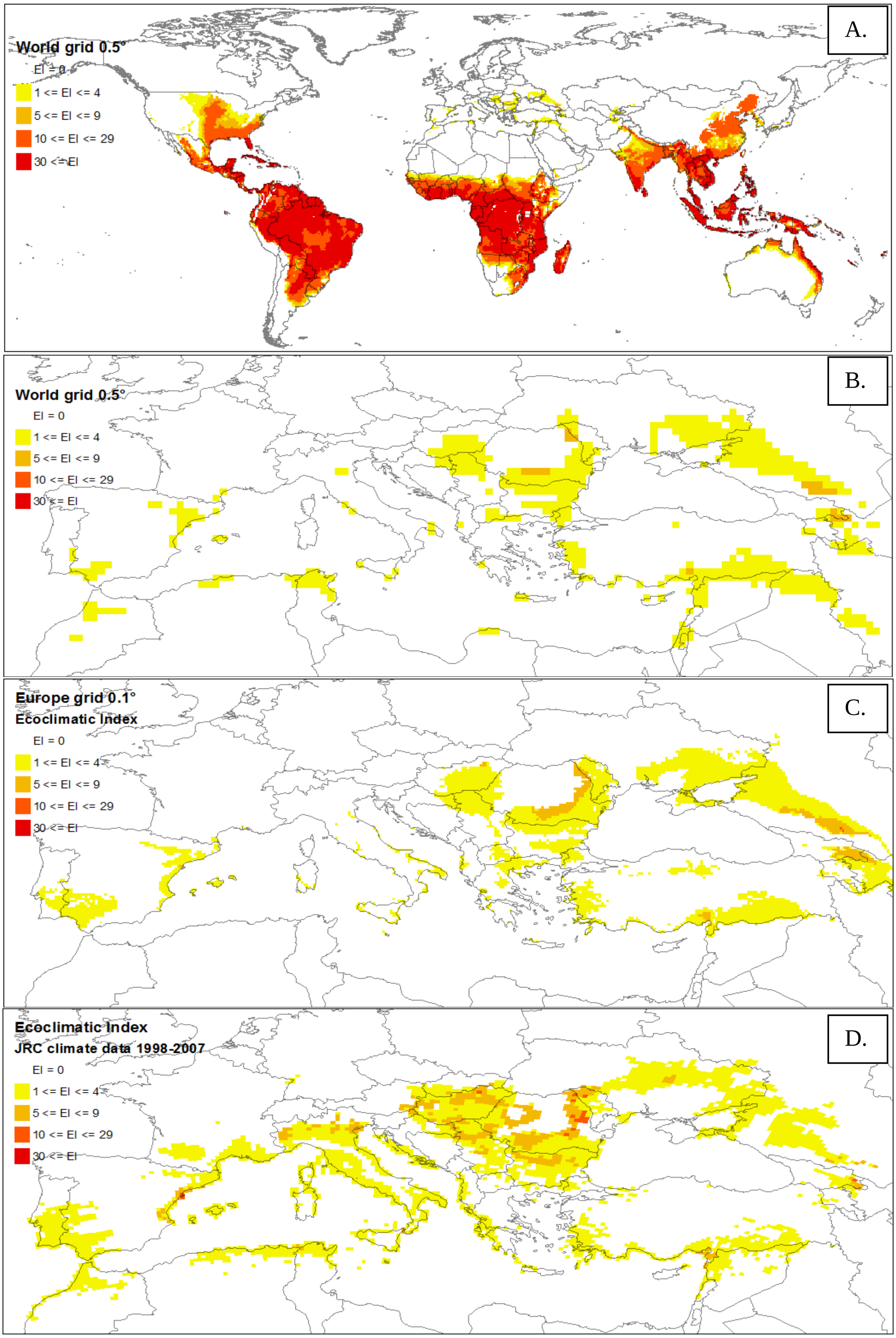

Figure 27: Prediction of the potential global distribution of *P. citricarpa* according to the CLIMEX model developed by Yonow et al. (2013) based on: a) 0.5° latitude x 0.5° longitude global spatial resolution 1961-90 average climate data, b) the latter zoomed to Europe, c) 0.1° latitude x 0.1°

longitude spatial resolution 1961-90 climate data and d) 1998-2007 JRC climatic data at 25 km resolution.

A detailed examination of the output from the Yonow et al (2013) CLIMEX model in EU citrus growing areas shows that positive EIs occur where sufficiently high temperatures and moisture for pathogen development coincide. With the exception of a small accumulation of cold-wet stress, the outputs for all the other stress indices included in the model do not exceed zero indicating that no stresses accumulated during the season that is unfavourable for development.

In Figure 28, the results for the two key factors promoting growth in CLIMEX, namely the Temperature Index and the Moisture Index, and the product of these two, the Growth index, have been plotted at the weekly resolution on which CLIMEX operates at one location in the Ebro delta. The Panel concludes from these results that the CLIMEX parameterisation by Yonow et al (2013) appears to emulate the temperature and moisture requirements of the fungus in a similar way to the modelling approach for spore maturation and infection adopted by the Panel, but with a lower level of detail. The accumulation of the Growth Index for *P. citricarpa* occurred during weeks 15 – 22 in spring and weeks 37 – 43 in autumn (Figure 28). More details from these analyses can be found in Appendix D.

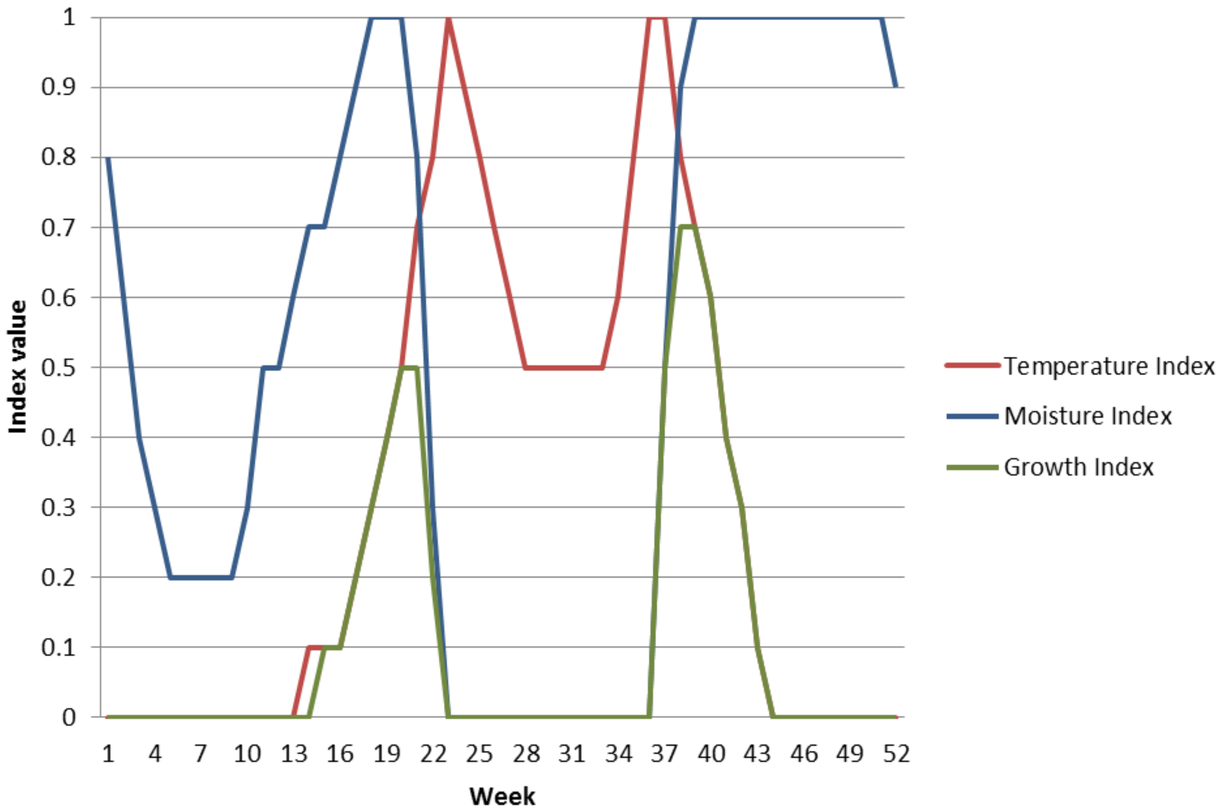

Figure 28: Weekly CLIMEX Temperature, Moisture and Growth Index values from the Ebro delta region in eastern Spain for 1961-90 at 0.1° latitude x longitude spatial resolution.

Yonow et al. 2013 (drew) the conclusion from their CLIMEX modelling of the potential global distribution for citrus black spot disease that *"Within European citrus producing regions, suitable areas are highly constrained, never more than marginally suitable, and all have lower levels of suitability than any area in South Africa and Australia where G. citricarpa is known to occur"*. However, the results presented here from a limited analysis of this new CLIMEX model supports the Panel's concerns about the extent to which the CLIMEX Compare Locations procedure can provide reliable results for this species. This is because the CLIMEX model for *P. citricarpa* parameterised by Yonow et al (2013):

- is shown to be highly sensitive to the spatial resolution and time period of the climate data inputs with regard to whether the EU citrus growing areas are suitable for establishment of *P. citricarpa*. Thus for some of the EU citrus growing areas, the climatic suitability classification varies from “marginally suitable”, through “suitable” and even to “highly suitable” based on the classification of the EI by Yonow et al. (2013) when changing either the spatial resolution or the temporal period of the climate data inputs.

- For the EU citrus growing areas, the outputs from the Yonow et al (2013) CLIMEX model mainly show where high temperatures and moisture coincide, as very little stress is accumulated in these areas

- The summary of high temperature and moisture coincidences provided by CLIMEX cannot be used to draw reliable conclusions about the extent to which EU citrus growing areas have a suitable climate for *P. citricarpa*.

3.3.3. Cultural practices and control measures

3.3.3.1. Irrigation

Practically all the commercial citrus orchards existing in the EU are irrigated nowadays (Carr 2012). However, the type of irrigation system employed is not uniform throughout the EU citrus orchards. This is important because the irrigation system employed and its management can influence the incidence of *P. citricarpa* primarily by affecting ascospore production and release due to changes in the leaf microclimate, the tree leaf ageing rate and the leaf decomposition on the ground(Dewdney et al., 2011).

The irrigation systems used in the EU citrus orchards are: surface irrigation, sprinkler irrigation and micro irrigation (see Stewart et al., 1990 for more details about each method). While 40 years ago most of the citrus plantations were surface irrigated, the general trend is to replace surface irrigation methods with pressurized systems (sprinkler and micro-irrigation) reducing soil evaporation, increasing the overall orchard irrigation efficiency and minimising the volume of water applied.

<u>Surface irrigation</u>

In these irrigation systems, the irrigation water is applied at one edge of an orchard and flows across the soil surface by gravity. As water moves over the soil, water infiltrates into the rootzone. Irrigation water applications are generally applied every 13-25 days watering the soil to a depth of 40 to 80 mm at each irrigation event. Two main types of surface irrigation methods are applied in Europe: 1) flooding, where the entire orchard floor is irrigated, and 2) the graded furrow, where prior to the first irrigation application, furrows to convey the water across the field are ploughed between the crop rows. In the furrow irrigation method, the proportion of the soil orchard that is wetted might vary from 30 to 70% of the entire citrus orchard floor. In this case, most of the ground shaded by the tree, where most of the fallen citrus leaves will remain, will be wetted. Even with the furrow method most of the citrus leaves on the ground will be wetted.

<u>Sprinkler irrigation</u>

In these systems water is supplied in a pressurized network and emitted from sprinkler heads mounted on either fixed or moving supports. In European citrus orchards, only set sprinkler irrigation systems are found. Set systems are those in which the sprinklers are placed in a fixed grid or spacing. The entire orchard floor is wetted and the water applications are applied over the tree canopy, so the irrigation water completely wets the tree canopy in the same way as rainfall. Sprinkler irrigation is generally applied every 7-20 days giving an amount of water equivalent to 20 to 60 mm of rainfall.

In addition to irrigation water applications, set sprinkler systems can also be used for frost protection. Sometimes citrus orchards use another type of irrigation system employing set sprinkler irrigation only for frost protection.

Micro irrigation

Micro irrigation includes methods that are more commonly known as drip irrigation and other low pressure systems. Water is generally distributed in plastic conduits and emitted by trough drippers, tricklers, foggers, micro-sprinklers or sprayers. In European citrus orchards, two main types of micro irrigation systems are found.

Drip irrigation, where water is allowed to drip slowly to the soil through an emitter with a low discharge rate (0.8 to 8 liters per hour). The main feature of this type of irrigation technology is that: 1) only a small proportion of the entire orchard floor is wetted by the irrigation system (15 to 35 % of the soil beneath each tree), and 2) frequent applications of water are applied (generally 1-4 mm per irrigation event, with a daily application during the summer months). Another subtype of drip irrigation is sub-surface drip irrigation, where the pipelines transporting water and the emitters are located in the sub-soil at a depth of 30 cm, and where the water does not reach the soil surface. However, sub-surface irrigation systems are rarely used in citrus orchards because their installation is expensive and they are complex to maintain.

Micro-sprinkler is another type of micro irrigation system where water is applied by sprayers located underneath the tree canopy, 45-70 cm above the soil orchard. This wets 30-70% of the entire orchard floor and some of the lower part of the tree is also directly wetted by the irrigation system.

3.3.3.2. Regional differences in citrus irrigation

Spain

The Spanish citrus irrigation orchards are mostly irrigated either by flood or drip irrigation using low pressure operating emitters located at the soil surface. In the Valencia region, according to Pons (2008), 67% of the citrus orchards are irrigated using drip systems, while 32% is under flood irrigation. Micro-sprinkler irrigation is only used in the remaining 1% of the Valencia citrus orchard plantations, where they are employed to provide some frost protection. However, this sprinkler system is not overhead and only wets the lower part of the tree canopy.

In the southern citrus growing areas of Spain (Andalucía and Murcia), where citrus orchards plantations are generally younger (particularly in Andalucía), drip irrigation systems are more common with 81% of the citrus orchards using drip systems and the remaining 19% using flooding irrigation (MAGRAMA 2013).

Italy

In Sicily the dominant irrigation system is a type of micro-sprinkler irrigation which uses low pressure sprayers that often wet most of the orchard floor (Liberati 2008). Irrigation is applied every 8 to 25 days and applications range from 20 to 60 mm per session. Drip irrigation is applied in the remaining 10% of the citrus irrigated area. Overhead sprinkler systems are used in some areas of Sicily and particularly in the regions of Calabria and Campania but the percentage of the citrus irrigated area with overhead sprinkler systems in these two regions is only 6% (Consoli, 2010).

Portugal

In Portugal, most of the commercial irrigated citrus orchards are located in the Algarve region. According to Norberto (2011), in this region, 88% of the citrus orchard are irrigated by drip irrigation, 8% by micro-sprinklers applied below the tree canopy at about 100 cm above the soil surface, and 4% by flood irrigation.

<u>Greece</u>

According to a recent review by Shirgure (2012), micro and flood irrigation are the two main types of irrigation systems used in the citrus growing areas of Greece. In the Argolis county of South-Eastern Peloponnese (Prefecture of Argolida, Subject: Data on irrigation systems of citrus in the prefecture of Argolida, 28/11/2012), with a total citrus area of 12,500 ha: 1,000 ha have flood irrigation (8%), 300 ha have drip irrigation (2.4%) and 11,200 ha use low pressure micro-sprinkler sprayers (89.6%). In the low pressure system, the sprayers are located at a height of 40 cm above the orchard floor with one sprayer per tree at a distance of 40-80 cm from the trunk. This means that the water drops are ejected up to a height of 60 cm wetting most of the lower parts of the tree canopies. During the winter months, sprayers are used for the protection of citrus trees from frost in an area of 2,000-3,000 ha.

<u>Cyprus</u>

In Cyprus, traditionally farmers have used the flooding method to irrigate citrus orchards. However, with modernization, 26% of the orchards are now drip irrigated. In the remaining 74% of the irrigated citrus orchards, flood irrigation that wets the entire orchard floor is applied (Mehmet and Ali Biçak 2002).

<u>Malta</u>

In Malta the most reliable source of information comes from the study by Attard and Azzopardi (2005). They reviewed the irrigation systems used and water use efficiency in irrigated Maltese agriculture. Drip irrigation use has steadily increased in recent years and 46% of the citrus is drip irrigated (National Statistics Office, Malta 2010). However, 52% of the irrigated citrus orchards are still flood irrigated. The remaining 2% of the orchards are irrigated according to other systems apart from flood and drip irrigation.

<u>France</u>

In the French citrus orchards mainly located in the Corsica island 43% of the plantations are under sprinkler overhead irrigation, while drip and micro-sprinkler irrigation are used in 28 and 29% of the citrus orchards, respectively (Dr. Jean Bouffin, INRA, personal communication).

<u>Summary of the irrigation practices in European citrus orchards</u>

In summary, it is clear that the trend is to move away from the irrigation systems, e.g. flood and sprinkler irrigation that use large amounts of water, wet the soil surface of the whole orchard and are likely to have a major influence on the microclimate within the orchard. However, while most of the micro irrigation systems use much less water and are likely to have a minor effect on microclimate, micro-sprinkler irrigation uses spray jets located under the tree canopy that not only wet the soil but also the lower canopy of the tree significantly increasing leaf wetness and relative humidity. Micro-sprinkler irrigation is particularly common in Sicily and Greece. Even though some of the information on irrigation practices are not from recent publications, micro sprinkler irrigation together with flood and sprinkler irrigation systems are still considered to be widely used in most citrus producing EU countries. This is likely to enhance the likelihood of *P. citricarpa* establishment in EU citrus orchards by providing greater opportunities for completing the life cycle than predicted by the Magarey et al (2005) and Fourie et al (2013) models.

3.3.3.3. Citrus growing habits and other cultural practices in the EU

The range of variation in tree growth habits exhibited by the citrus trees as a whole is very wide: from the straggly, shrub-like citron to the large, highly symmetrical trees of most of the sweet oranges and grapefruits and some of the mandarins.

Citrus trees are generally pruned to a central leader or a modified central leader shape, and pruning operations are conducted annually, reducing the apical dominancy of the natural tree branches. A full canopy of leaves is normally maintained in order to protect the bark of the trunk and branches from direct sun and potential sunburn. Trees often have branches close to the ground (a full skirt) in order to maximize photosynthesis and therefore tree productivity (Agustí 2003). In addition, since practically all the commercial citrus orchards in the EU are manually harvested and the labour costs are expensive, it is important to maintain orchards that can be easily harvested by hand operators. Because of this, in general, the European citrus orchards tend to be restricted to small trees with a height that is often less than 3 m (Vacante and Calabrese 2009). In addition, modern plantations now use citrus scions grafted onto semi-dwarfing rootstocks which limits tree height to less than 2.5 m height (Legua et al., 2011). Under these situations, the weight of developing fruit generally pulls some branches down very close or even in direct contact with the orchard floor (Fake 2012).

3.3.3.4. Citrus disease management in the EU

It is considered that current fungicide spray schedules in EU citrus-growing areas generally will not prevent the establishment of *P. citricarpa*. Some late maturing sweet orange cultivars are sprayed in autumn with fungicides such as fosetyl-Al for the control of brown rot caused by *Phytophthora* spp. These fungicides are specific for oomycetes and are ineffective against fungi like *P. citricarpa* (Tuset, 1987). Some late maturing mandarin hybrids, such as 'Fortune', 'Nova' and 'Murcott', are routinely sprayed in spring and autumn with copper or mancozeb for the control of Alternaria Brown Spot (Vicent et al., 2009; Vicent et al., 2007). Although these chemicals are not among the most effective for the control of *P. citricarpa* (see section 3.6.1.1), they could to some extent prevent possible infections of *P. citricarpa*. However, the areas grown with cultivars susceptible to Alternaria Brown Spot represent a very minor proportion of the EU citrus-growing area, whereas in the most of the EU citrus-growing areas no fungicide sprays are usually applied.

3.3.4. Other characteristics of the pest affecting the probability of establishment

Very little is known of the rate of inoculum build-up from small initial populations of *P. citricarpa*. *P. citricarpa* may be present as latent mycelia in asymptomatic citrus fruit and leaves with a long lag phase between the first establishment and subsequent epidemic development (Kotzé, 1981; for more details see section 3.4.3).

3.3.5. Conclusions on the probability of establishment

A summary regarding the assessment of the components of the probability of establishment is presented in Table 7 below.

Assessment of the components of the probability of establishment and uncertainty

Rating for establishment	**Justification**
Availability of suitable host(s) **Widely** available	• Citrus is grown in southern areas of the EU with a sufficiently warm climate that is only rarely exposed to frosts • Within the citrus growing regions, the host plants are grown in commercial citrus orchards and nurseries, as well as in streets and private and public gardens, both in urban and rural areas

	• Citrus leaves are susceptible to *P. citricarpa* for 8-10 months and citrus trees have three main flushes in spring, summer and autumn. Sweet orange fruits are susceptible for at least 6-7 months after fruit set in spring.
Suitability of environment - *Similarity of climatic conditions in the current area of distribution* **Slightly similar**	• This rating is limited to the citrus growing areas of the EU. *P. citricarpa* mainly occurs in subtropical citrus-growing regions characterized by a summer rainfall pattern and high annual precipitation. However, the disease is also present in drier areas, such as the Eastern Cape province in South Africa, with an annual rainfall of approximately 400 mm that is comparable to the rainfall in some EU citrus growing areas. Based on simulation results, conditions in the EU citrus growing areas during September and October are generally suitable for ascospore development and infection whereas spring infection is limited to specific locations, e.g. in southern Spain, Malta, Cyprus and the Greek islands.
Cultural practices and control measures - *To what extent is the managed environment in the risk assessment area favourable for establishment?* **Highly favourable** - *How likely is it that existing pest management practice will fail to prevent establishment of the pest?* **Likely**	• For several reasons, EU citrus orchards are designed and maintained as small trees and their height is often lower than 3 m. The weight of developing fruit generally pulls some branches down very close or even in direct contact with the orchard floor and this will aid splash dispersal of pycnidiospores. • Irrigation techniques wetting the leaves and fruit are still in use in parts of the EU citrus growing areas, thus creating a micro-environment favourable to the establishment of the disease
Other characteristics of the pest affecting the probability of establishment **Likely**	• Small populations are likely to become established as there is evidence that shows that it may take decades from initial introduction until epidemics reach damaging levels of impact.
Overall probability for establishment **Moderately likely**	• Based on the modelling of ascopore maturation and release, the Panel found additional evidence that part of the risk assessment area has climate conditions favourable for inoculum production for *P. citricarpa.* • The results from these simulations based on both the gridded and station weather data show that there are locations where the period of host susceptibility, inoculum availability and suitable environmental conditions for infection overlap. • The likelihood of establishment is assessed as **moderately likely:** owing to the simultaneous occurrence of host susceptibility and weather conditions suitable for ascospore production and infection

	(primary infection cycle).

3.3.6. Uncertainties on the probability of establishment

Overall, uncertainty on the probability of establishment is rated as high, mainly due to lack of knowledge on how *P. citricarpa* will respond under the EU climatic conditions. Although the environmental factors that are important in the various stages of the life cycle are known, there is insufficient scientific evidence to determine the thresholds of the values the organism requires, e.g. for the temperature and wetness levels and durations (table 8). Further validation of the models applied by the Panel, especially for marginal areas within the current distribution of the citrus black spot disease would be needed to reduce the uncertainty on the probability of establishment of *P. citricarpa* in the EU.

Uncertainties concerning the probability of establishment

Level of uncertainty	Justification
Availability of suitable host(s) **Low**	• The citrus varieties grown in the EU are known to be susceptible to *P. citricarpa*
Suitability of environment - *Similarity of climatic conditions in the risk assessment area and in the current area of distribution* **High**	• Only limited data are available to precisely define some of the climatic requirements of the pathogen (temperature and wetness duration thresholds). The previous EFSA opinion showed that the outputs of the epidemiological model of Magarey et al. were highly sensitive to some parameters (D50 and Tmin). • As the relationship between the proportion of ascospore release and infection efficiency has not been studied, there is a high level of uncertainty concerning the threshold of percentage of ascospore release that must be set for the Fourie 2 model to determine whether ascospore release occurs or not (it has been set to 1% in the opinion) • There is limited information on pathogen presence/absence, as well as disease development data, from marginal areas within the current area of distribution, e.g. the Eastern Cape Province of South Africa. If marginal areas within the current distribution could be defined, detailed weather data from such marginal areas could reduce uncertainty about suitability of climate in areas outside the current distribution
Cultural practices and control measures - *Managed environment favour establishment* **Low** - *Existing management practices will fail to*	• The cultural practices in use are relatively well known and there are also to some extent available quantitative data on the cultivation practices in use in the EU citrus production area • The meta-analysis of fungicide trials show that most fungicide treatments have limited efficacy in eliminating the pathogen. It

prevent establishment - **Low**	could therefore be assumed that there is low uncertainty about the likely failure of existing non-targeted management practices to prevent pathogen establishment
Other characteristics of the pest affecting the probability of establishment **High**	• Very little is known on the rate of inoculum build-up from small initial populations of *P. citricarpa*
Overall probability for establishment **High**	• The uncertainty is high, mainly due to: i) the uncertainty on key biological parameters of the pathogen, ii) the need for more experimental data covering a wider range of climatic and citrus growing conditions to model the establishment potential, iii) the lack of knowledge on the relationship between ascospore proportion and infection efficiency and iv) the lack of knowledge about the rate of inoculum build-up for this pathogen.

3.4. Probability of spread after establishment

3.4.1. Spread by natural means

Natural spread of *P. citricarpa* occurs by ascospores and pycnidiospores, the former by wind dispersal and the latter primarily by splash dispersal. The pycnidiospores are formed into asexual fruiting bodies (pycnidia) on lesions in fruit, twigs and leaf litter. Pycnidiospores are splash-dispersed or washed-off by rain for relatively short distances, infecting susceptible leaves and fruit. On fruit and twigs only pycnidiospores are formed, while on citrus leaf litter both ascospores and pycnidiospores are formed. By removing the leaf litter, Spósito et al (2011) studied the relative importance of inoculum sources of ascospores and pycnidiospores in the spread of CBS under natural conditions in Brazil. The recorded distance of disease spread was less than 80 cm from these inoculum sources.

The removal of fallen leaves was not sufficient to completely suppress the disease because of the presence of pycnidiospores in fruit and dead twigs. Therefore, Spósito et al (2011) concluded that the reduction of pycnidiospores sources should be considered in CBS management in Brazil.

Pazoti et al. (2005), referring to a paper by Goes (2002), stated that “ascospores are spread not only on short, but also on long distances: the wind can spread it, infecting orchards at kilometres of distance”. However, no data on ascospore dispersal are provided by Goes (2002).

3.4.2. Spread by human assistance: fruit trade

The citrus fruit trade networks shown in this section were created using Gephi, an open-source and free software for network visualization and analysis (https://gephi.org/). Networks are sets of ‘nodes’ (in this case, EU MS) connected by links (in this case, consignments of citrus fruit during 2011). For the trade of citrus fruit, the networks are directed, because export of a certain amount of citrus fruit from country A to country B in year Y does not imply that the same amount (and type) of citrus fruit is exported from country B to country A in the same year Y, so that it is important to keep track of the directionality of trade flows.

The network based on the intra-EU trade data for oranges in 2011 (Eurostat, online) is shown in Fig. 29. Croatia is included because this country joined the EU in July 2013. The network has 28 nodes and 320 links (320 incoming and 320 outgoing), and thus a connectance ($C = L/N^2$) of 0.41. This means that 41% of the potential links are realized. The total amount of sweet oranges traded in 2011 was about 2 million tons.

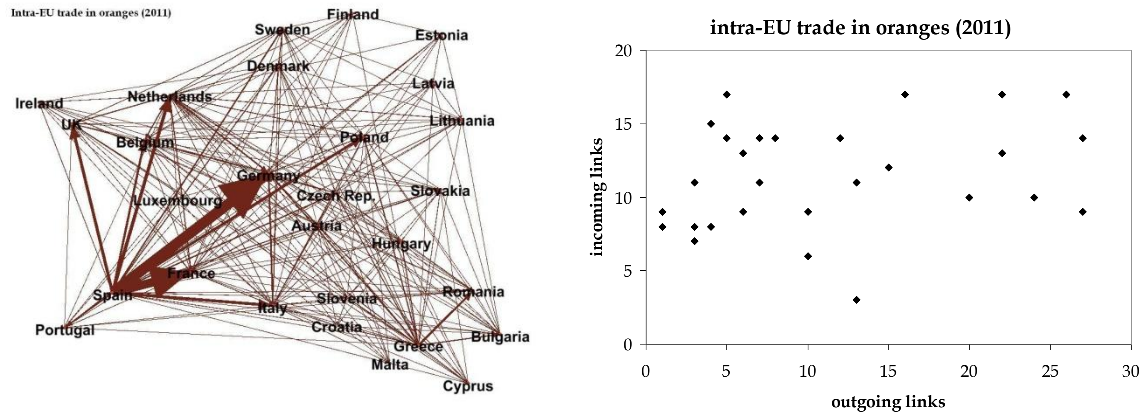

Figure 29: The intra-EU trade in sweet oranges (2011): (left-hand panel) network visualization (the weight of the links is proportional to trade volume), and (right-hand panel) correlation between the number of incoming links (countries from which sweet oranges were imported) compared to the number of outgoing links (countries to which sweet oranges were exported).

Seven countries export sweet oranges to at least 20 other countries (Spain and the Netherlands (27), Italy (26), Greece (24), Germany and France (22), and Belgium (20)). This is not the case for imports: the maximum number of countries from which sweet oranges are imported is 17 (this is true for Denmark, Germany, Italy and Poland). Some countries are more connected than others, as shown by Fig. 29.

There is no correlation between the number of incoming and outgoing links. Such a correlation, at least in theory, and other things being equal, would make it easier for a pathogen to spread (as reviewed by Moslonka-Lefebvre et al., 2011). Nonetheless, there are some countries that import sweet oranges from many countries and also exporting them to many countries (e.g., the Netherlands, Italy, Germany and Poland). These countries are more likely to contribute to disease spread than countries importing from many countries but not exporting to many countries (e.g. Denmark, Slovenia, Slovakia and Romania). With the exception of Cyprus (3), Portugal (6) and Luxembourg (7), all EU countries import sweet oranges from at least eight different countries.

There is considerable variability in the weight of the connections, with just two links (from Spain to Germany and to France) making up about one third of the whole sweet orange trade between European countries. On its own, Spain is responsible for nearly two thirds of the intra-EU trade in sweet oranges (Fig. 30). About 80% of the Spanish export goes to just six countries (in decreasing order of imported sweet oranges: Germany, France, the Netherlands, the UK, Italy and Poland). Sweet orange imports are also uneven between countries, but less than exports. Germany imports about 24% of the total intra-EU trade, France 18% and a further 20% is imported by the Netherlands (7%), Poland (7%) and the UK (6%). The network from the point of view of the Netherlands is shown in Fig. 30.

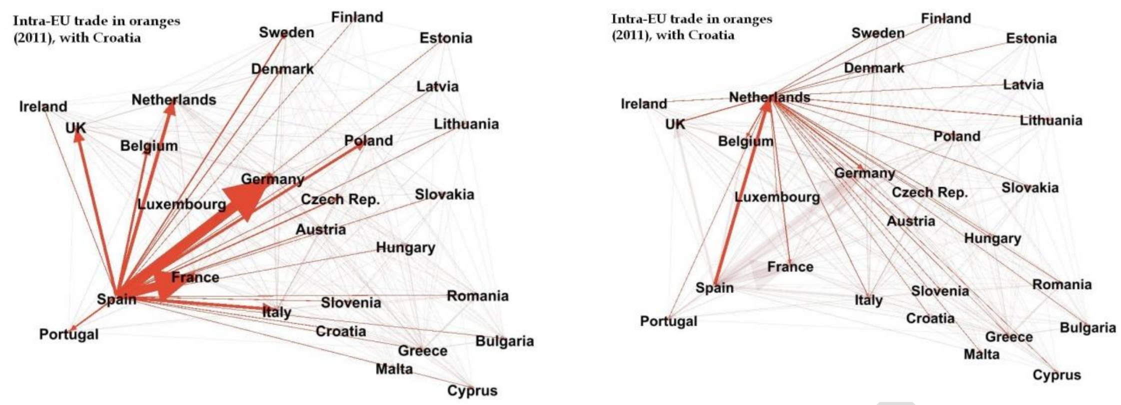

Figure 30: Trade of sweet oranges with other EU countries in 2011: (left-hand panel) by Spain, and (right-hand panel) by the Netherlands, a country that exports more sweet oranges to EU countries than it imports.

In 2011, the Netherlands imported more sweet oranges from South Africa than from the rest of the EU. South Africa is well connected to the EU sweet orange trade network, as shown in Fig. 31. In 2011, South Africa exported about 0.3 million tons of sweet oranges to 22 EU countries, with nearly 50% of South African sweet oranges being imported by the Netherlands. Argentina is another major exporter of sweet oranges into the EU (Fig. 31). In 2011, like South Africa, Argentina exported sweet oranges to all the citrus-producing countries of the EU.

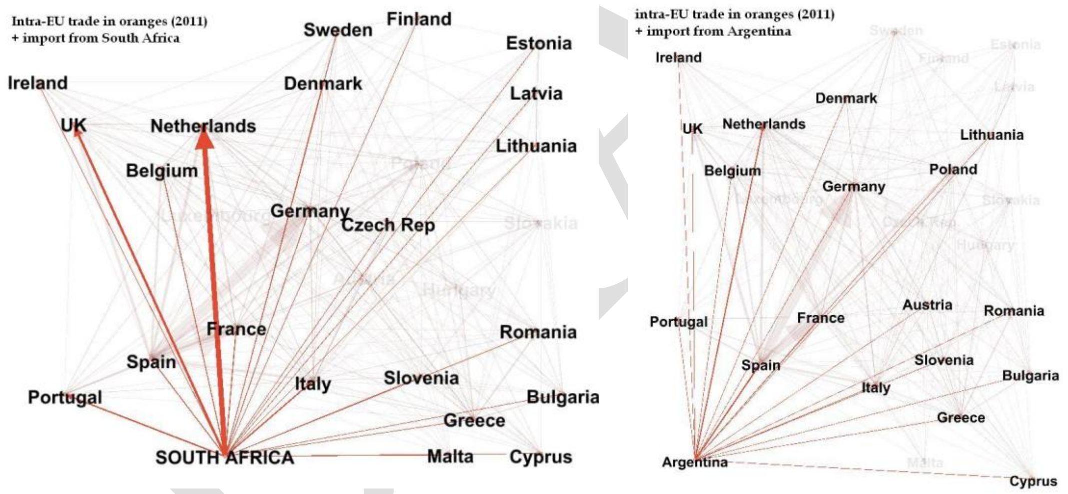

Figure 31: Imports of sweet oranges by EU countries (2011): (left-hand panel) from South Africa and (right-hand panel) from Argentina.

3.4.2.1. Mandarins

The network of the intra-EU trade in mandarins (2011) is shown in Fig. 32 (with the addition of Croatia). There are fewer trade links than for sweet oranges (300 instead of 320) and hence a slightly lower connectance level (0.38 instead of 0.41). Also the amount of traded mandarins is lower than for sweet oranges (~ 1.6 vs. 2 million tons). There are six countries exporting mandarins to at least 20 EU countries: the Netherlands (to 27 countries), Spain and Italy (26), Germany and France (22), and Greece (21). There is a weak positive correlation between the number of countries from which mandarins are imported and the number of countries to which mandarins are exported (Fig. 32). No EU country imports mandarins from 20 or more EU countries, with Italy importing them from 17 countries and Spain and Poland from 16.

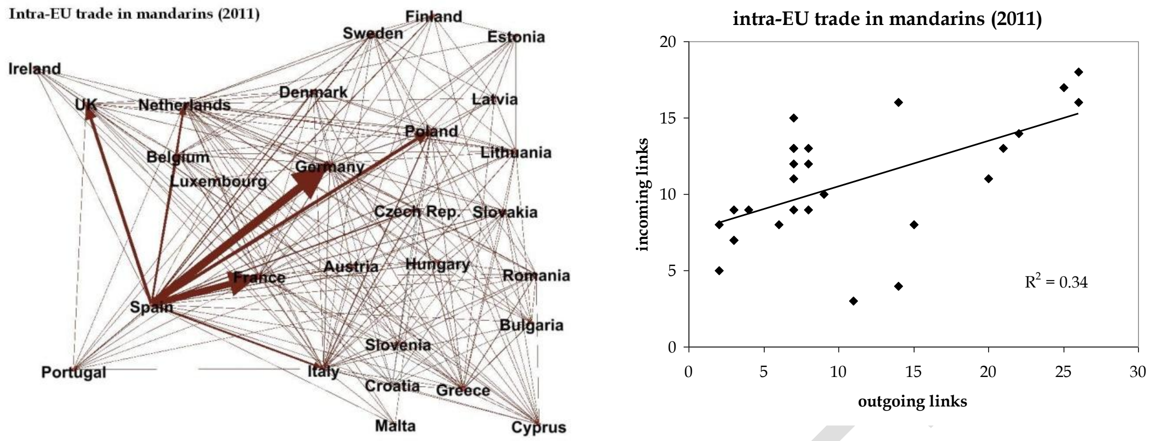

Figure 32: The intra-EU trade in mandarins (2011): (left-hand panel) network visualization, and (right-hand panel) correlation between the number of countries to which mandarins were exported and from which mandarins were imported.

Just as for sweet oranges, Spain is the major EU mandarin exporter (three quarters of exports), whereas France and Germany are the main EU importers (together, 42% of imports). About three fourths of exported EU mandarins come from Spain. Approximately half of the Spanish export to EU countries goes to Germany and France (Fig. 33).

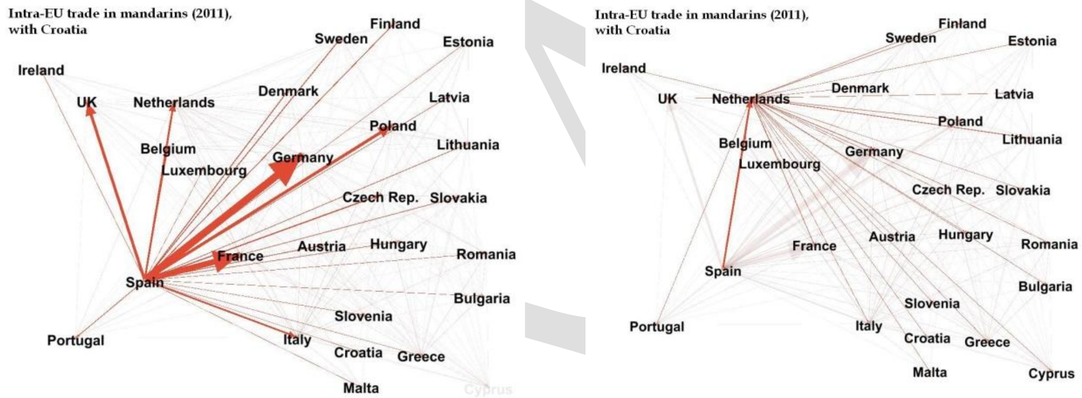

Figure 33: The intra-EU mandarin trade network (2011) from the point of view of Spain (left-hand panel), and the Netherlands (right-hand panel).

Unlike with sweet oranges, the Netherlands import fewer mandarins from EU countries than this country exports to them, but the Netherlands are still a major re-exporter (to all other EU countries, including Spain) (Fig. 33).

The South African exports of mandarins (0.05 million tons) are smaller than for sweet oranges (one sixth of that figure) and go to 12 countries (mostly the UK and the Netherlands) (Fig. 34). The exports of mandarins to EU countries from Argentina for the same year are shown as a comparison in Fig. 34.

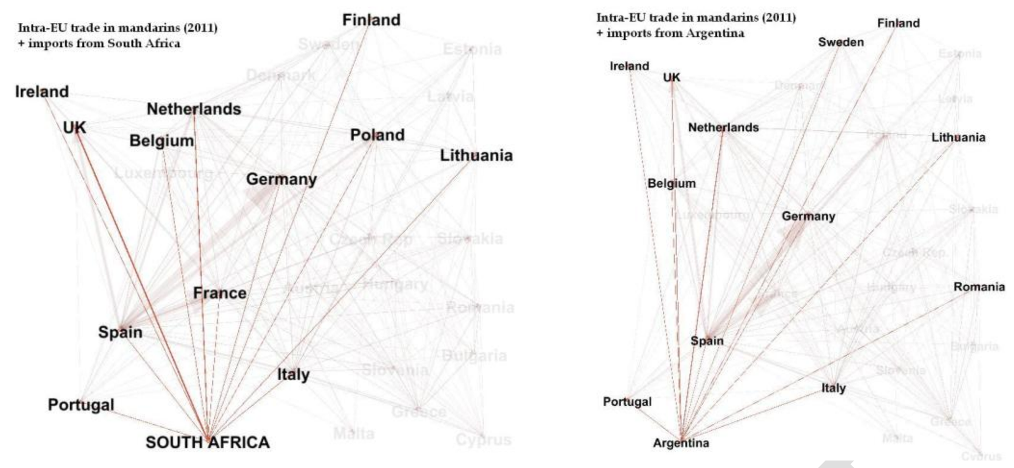

Figure 34: Import of mandarins (2011) by EU countries: (left-hand panel) from South Africa, and (right-hand panel) from Argentina.

3.4.2.2. Lemons

The intra-EU trade in lemons (2011) is shown in Fig. 35 (with the addition of Croatia). The network is slightly less connected than for mandarins (269 instead of 290 links with a connectance level of 0.36. The amount of traded lemons is also lower than for mandarins (~ 0.5 vs. 1.6 million tons).

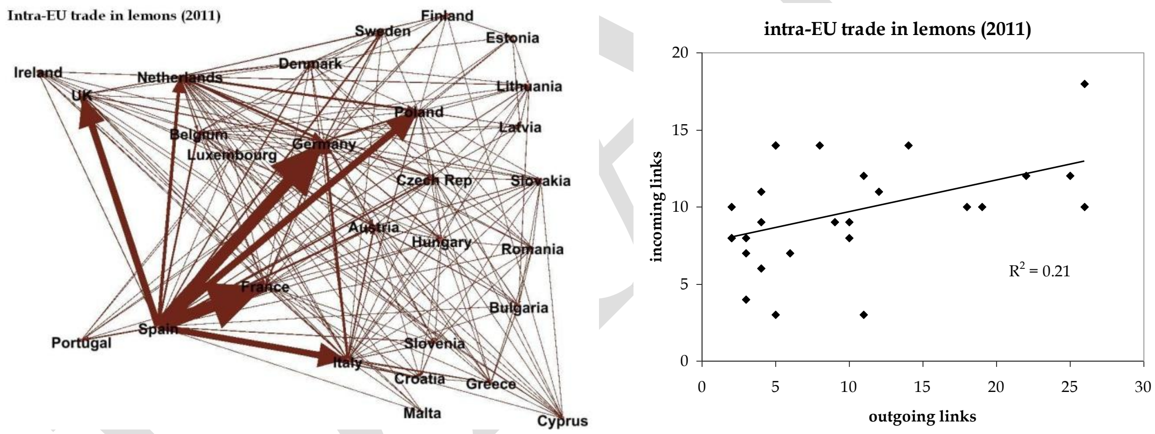

Figure 35: The intra-EU trade in lemons (2011): (left-hand panel) network visualization, and (right-hand panel) correlation between the number of EU countries from which lemons were imported and the number of EU countries to which lemons were exported.

In 2011, only four EU countries exported lemons to at least 20 EU countries: Spain and the Netherlands (26), Italy (25), and Germany (22). Import sources are less diverse, with Poland importing lemons from 18 countries, and Denmark, Estonia, Portugal, Slovenia from 14 (Fig. 35).

Spain is the major EU exporter of lemons (Fig. 36). About one third of imported EU lemons go to Germany and France. The Netherlands import few lemons from EU countries (Fig. 36) (fewer than, e.g., Austria imports), but the Netherlands export to EU countries more than twice as many lemons as they import from EU countries.

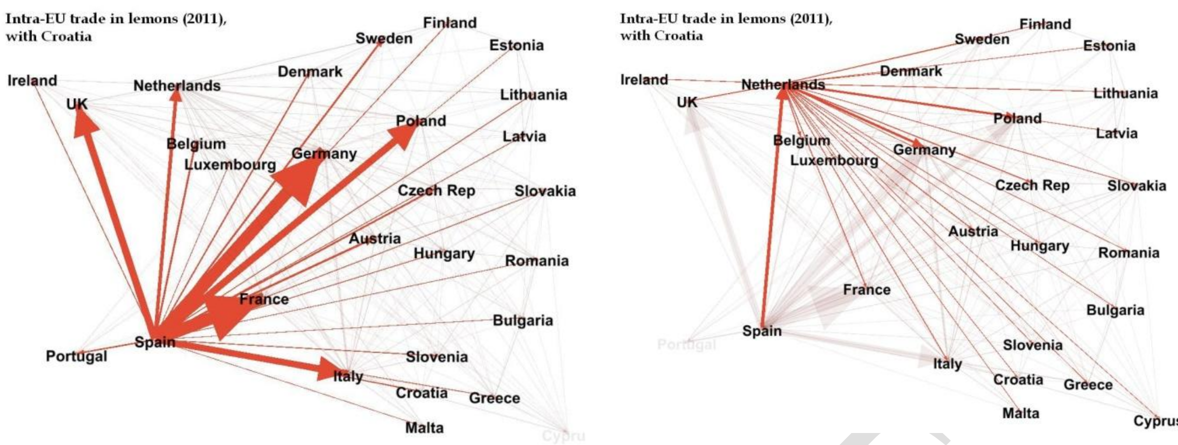

Figure 36: The intra-EU network in lemons (2011) from the point of view of (left-hand panel) Spain and (right-hand panel) the Netherlands.

South Africa exported about 0.04 million tons of lemons to EU countries in 2011 (Fig. 37). This is more than the lemons exported by Italy to EU countries in 2011. South African lemons are directly imported by 16 EU countries, including Greece, Italy, Spain and Portugal. Most South African lemons exported to the EU went to the UK and the Netherlands. Argentina is a more important exporter of lemons to the EU than South Africa, with most lemons exported to the Netherlands, Spain and Italy, as shown by Fig. 37.

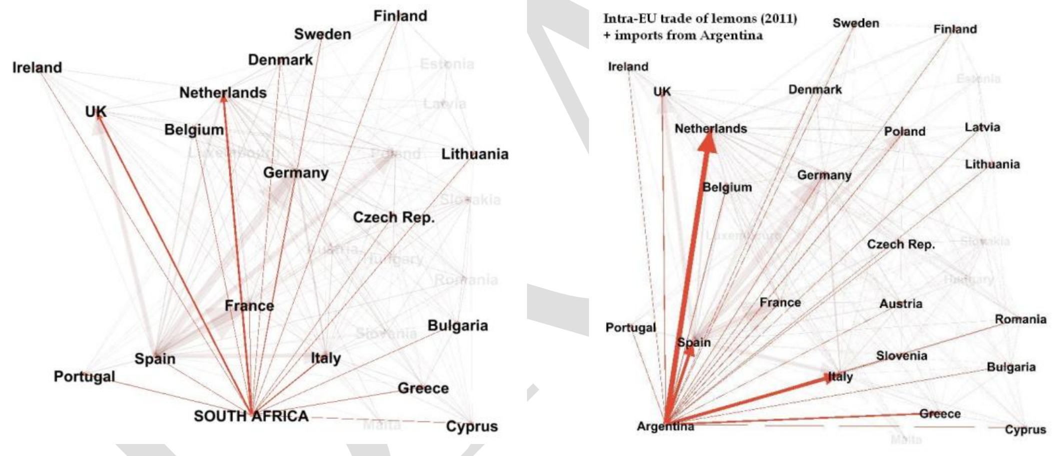

Figure 37: Import of lemons (2011) by EU countries: (left-hand panel) from South Africa, and (right-hand panel) from Argentina.

3.4.2.3. Conclusions on spread by human assistance: trade of citrus fruit

1. On the whole, the citrus trade data for 2011 show that the EU market for sweet oranges, mandarins and lemons is closely integrated. On average, each EU MS imports from (or exports to) 9, 10 and 11 other EU countries (for lemons, mandarins and sweet oranges, respectively).

2. There are strong variations in connectivity between countries, both in terms of the number of links and traded volumes. Heterogeneities in the contact structure have been shown to reduce thresholds for disease spread and persistence in networks (Jeger et al., 2007), although for directed networks this is only the case in the presence of a positive correlation between incoming and outgoing links (Moslonka-Lefebvre et al., 2009). This is weak here, despite the presence of the Netherlands that plays a strong role as importer and re-exporter not only from European countries.

3. The trade data indicate that sweet oranges, mandarins and lemons from countries where CBS is present (e.g. Argentina and South Africa) can reach citrus-growing EU countries both directly and via the Netherlands (or, potentially, through other re-exporting EU countries). However, the available Eurostat data only provide information on direct links between EU countries and thus do not allow examining how likely it is that imports of citrus fruit by EU MS will be re-exported to citrus growing EU MS.

3.4.3. Spread by human assistance: trade in citrus plants for planting

Infected plants for planting are considered to be the main pathway of introduction of CBS into new areas (Kotzé, 1981). However, documented and traceable reports of introductions by means of propagating plant material have not been found in the literature. Once introduced in an area, trees from nurseries located in the affected area may also spread the disease to new locations. Due to their microscopic nature, the movement of *P. citricarpa* ascospores is unlikely to be prevented by screening. Leaf wetness, which is necessary for spore germination and infection, can be reduced in greenhouses by avoiding overhead sprinkler irrigation. However, condensation of water on the plants as a result of soil water evaporation and the cooling of leaves below the dewpoint cannot be completed prevented (Jarvis, 1992, Wei et al., 1995). Foliar lesions of CBS are rare, especially in young vigorous plants (Kotzé, 1981). Since plants for planting produced in an affected area may carry latent infections, the efficacy of visual inspections in the nurseries is very low.

3.4.4. Containment of the pest within the risk assessment area

CBS has never disappeared or declined after the epidemic stage has been reached (Kotzé, 1981), and successful disease eradication has not been achieved anywhere. Whiteside (1967) proposed drastic pruning, involving the removal and destruction of all wood and leaves leaving only the frame work limbs of the tree, as a method to eradicate CBS in affected orchards in Zimbabwe. However, this method has not been put in practice and no reports of its efficacy are available. Field surveillance for CBS eradication is challenging, considering that *P. citricarpa* may be present as latent mycelia in asymptomatic citrus fruit and leaves and there is a long lag phase between the first establishment and subsequent epidemic development (Kotzé, 1981).

In 2010, CBS was first detected in U.S.A. in commercial citrus-growing areas in the Collier and Hendry Counties of south Florida (USDA APHIS, 2010b and c). After this first outbreak, a programme for the effective eradication of CBS was set up according to the recommendations of the Florida Citrus Health Response Program Working Group. This is possibly the only documented attempt to eradicate CBS anywhere. The programme included measures to suppress CBS in affected orchards as well as contiguous areas by monthly applications of fungicides and inoculum suppression by enhancing leaf litter decomposition with irrigation and the application of urea, dolomite lime or ammonium nitrate. Other measures such as avoiding off-season blooms, increasing air flow in the orchards, planting clean nursery stock and maintaining appropriate tree nutrient status were also recommended. However, all the recommended measures include removal and destruction of trees and/or systematic plant debris elimination in affected orchards in the quarantined area. In addition the recommended practices for disease suppression, regulatory measures have also been implemented in Florida. The movement of citrus plant material from quarantine areas is currently regulated (USDA APHIS, 2012b). Citrus plants may not be moved to other States from the quarantine areas. Fruit may be moved to other states of the U.S.A. only if they have been treated with specific postharvest treatments including sodium hypochlorite, sodium o-phenyl phenate, peroxyacetic acid, imazalil or thiabendazole. In the case of intra-state movement, fruit should be transported in vehicles properly covered by a screen mesh or tarpaulin to prevent the loss of fruit, leaves, or plant debris while in transit. After being emptied and cleaned of plant debris, trailers, field boxes and bins must be disinfested using sodium hypochlorite, quaternary ammonium chloride or peroxyacetic acid. Plant debris cleaned from trailers must be treated at 82ºC for at least 1 hour, incinerated, buried at an approved disposal site or used as livestock feed. Processors and packers receiving fruit from quarantine areas must follow also specific sanitation measures.

Nevertheless, despite all the measures described above, the disease expanded to new locations in south Florida in 2011 and 2012, and spread to Polk County in central Florida in 2013 (USDA APHIS, 2013).

3.4.5. Conclusion on the probability of spread

Rating for spread	Justification
Moderately likely	Natural spread of *P. citricarpa* is known to mainly happen by airborne ascospores. The distances it can spread by natural means is poorly known., The pathogen is very likely to move with human assistance along commercial fruit and plants for planting pathways. However, because spread is defined as the expansion of the geographical distribution of a pest within an area, the rate of spread depends not only on the rapidity of movement and the number of spread pathways but also the likelihood of finding a suitable environment for establishment. When the proportion of the citrus growing areas identified as potentially suitable for *P. citricarpa* is taken into account, the Panel considered that a rating of moderately likely is most appropriate for spread. The managed citrus orchards as well as home gardens, parks and abandoned citrus cultivations along all the EU citrus growing MSs will likely provide a continuum for the spread of CBS. The intra-European trade in citrus fruit is closely integrated, with an average of nine, ten and eleven trade connections (for lemons, mandarins and sweet oranges, respectively) from each EU MS. The Netherlands play a key role in re-exporting citrus fruit from citrus-producing countries (both within the EU and elsewhere) to other citrus-producing and non-citrus producing EU MSs..In the absence of regulation, infected plants for planting are the main pathway of CBS spread. This is because plants for planting produced in a CBS-infected area can carry latent infections. As a consequence, the efficacy of visual inspections in the nurseries, wholesale traders and retailers is limited.

3.4.6. Uncertainties on the probability of spread

Uncertainty on the probability of spread is rated as low.

Rating for uncertainty	**Justification**
low	• There is uncertainty about the potential natural spread of ascospores carried by wind over long distances, but this uncertainty does not concern the two main pathways of spread (intra-European trade of commercial fruit and plants for planting). There is uncertainty about the structure of the EU intra-trade network for citrus plants for planting owing to lack of data, however this does not influence the conclusions above.

3.5. Conclusion regarding endangered areas

The risk assessment has identified parts of the EU where host plants are present and where, based on simulation results, the climatic conditions are suitable for ascospore maturation and release followed by infection (see figures 25 and 26).

Conclusions from simulations of the release of ascospores based on gridded interpolated climate data of the EU citrus producing areas show that, in almost all years (for the 95 percentile), ascospore release in areas of Cyprus, Crete, Southern Greece, Italy, Spain and Portugal will start early enough to coincide with climatic conditions that are conducive to infection in September and October as simulated by EFSA (2008). However the same simulations indicate that the onset of ascospore release in most areas will start too late to coincide with the climatic conditions conducive to infection in April-May. Therefore, early maturing orange varieties might generally be infected in autumn only, which is when the availability of inoculum coincides with suitable conditions for infection. Due to the long incubation time, fruits from these early varieties will be harvested before symptoms appear. The late maturing oranges varieties and lemons are expected under such scenario to show CBS symptoms.

There are some areas however, such as locations in Cyprus, Greek islands, Malta and Southern Spain where onset of ascospores is expected also in May in half of the years simulated. In those years, it is expected that symptoms can develop on the fruit before harvest, and therefore have an impact on the fruit quality.

The results from the simulations on interpolated (grid-based) weather data are consistent with the simulations run on weather data measured by agrometereological stations.

The uncertainty is high as indicated in the establishment section.

3.6. Assessment of consequences

3.6.1. Direct pest effects

In most of the the area of its current distribution, *P. citricarpa* is reported to cause severe quality and yield losses to citrus production. Because of its quarantine status in some countries, *P. citricarpa* is not specifically listed in the International Standards for citrus fruit of the OECD. However, for other diseases, such as Alternaria brown spot with fruit symptoms similar to some of CBS, the presence of more than one lesion per fruit is considered detrimental to quality and fruits with more than six lesions are considered out of grade (OECD, 2010). In Sao Paulo, Brazil, fruits with more than three CBS lesions are considered unacceptable for the fresh market (Goes de, 2002). Premature fruit drop due to CBS causes significant yield loss in Brazil, and probably in other citrus regions of the world (Reis et al., 2006).

In order to obtain quantitative estimates of CBS impact in its current area of distribution, the Panel undertook a meta-analysis of recorded disease incidence in control (untreated) plots from CBS experiments on fungicide evaluation trials. A total of 46 experimental plots (site-years) from 16 papers were included in the dataset (table 9). Fungicide evaluations trials are generally optimized towards displaying treatment effects and it can be assumed that the experimental plots will be located in orchards severely affected by CBS. Because of a generally higher disease pressure than the average for the region, the disease incidence in untreated plots should be interpreted as the estimates of the highest potential loss (losses occurring in absence of control measures) and the incidence in fungicide treated plots as estimates of the highest primary loss (direct crop losses in presence of control measures) (Zadoks and Schein, 1979).

Directive 2000/29/CE (Annex IV Part A Section I 16:4) lists several requirements for the introduction of citrus fruit into the EU territory. One of these requirements is that the fruits originate in a place of production subjected to appropriate treatments against *P. citricarpa*. Fungicide schedules currently applied for CBS control are mainly based on the results obtained from field trials conducted in affected areas. Therefore, a meta-analysis study of these experiments may help to evaluate the efficacy of the different spray programs worldwide. Moreover, it may determine what level of disease control may be achieved by implementing the appropriate treatments required in the Directive 2000/26/CE and would help to devise the most appropriate fungicides to be used if an outbreak of CBS occurred in the EU.

Table 9: CBS fungicides control trials considered in the meta-analysis

Country	Citrus species	Number of experiments	Reference
Argentina	lemon	2	Fogliata et al., 2001
Argentina	lemon	1	Agostini et al., 2006
Argentina	sweet orange	1	Agostini et al., 2006
Argentina	sweet orange	1	Agostini et al., 2006
Argentina	sweet orange	3	Rodriguez et al., 2010
Australia	mandarin	1	Miles et al., 2004
Australia	sweet orange	2	Miles et al., 2004
Brazil	sweet orange	2	Kupper et al., 2006
Brazil	sweet orange	3	Goes, 2002
Brazil	sweet orange	2	Bernardo and Bettiol, 2010
Brazil	sweet orange	1	Goes et al., 2000
South Africa	sweet orange	4	Schutte et al., 2003
South Africa	sweet orange	2	Schutte et al., 2012
South Africa	sweet orange	1	Schutte, 2002
South Africa	sweet orange	4	Schutte, 2006
South Africa	sweet orange	1	Schutte et al., 1997
South Africa	sweet orange	4	Kellerman and Kotzé, 1977
Taiwan	mandarin	5	Tsai, 1981
Taiwan	sweet orange	3	Tsai, 1981
United States	sweet orange	1	Hendricks et al., 2013

3.6.1.1. Pest effects on citrus crop in the areas of current distribution

Disease incidences in untreated plots

The proportions of diseased fruits are shown in Figure 38 for several untreated plots (site-years) located in six different countries (Argentina, Australia, Brazil, South Africa, United States, and Taiwan. Mean proportion of diseased fruits ranged from 0.46 (Taïwan) to 0.98 (Brazil) (Table 10). The proportion of diseased fruits varied strongly between plots in a given country. For example, in South Africa, the minimum proportion of diseased fruits was equal to 0.14 and the maximum proportion was 0.98. Statistical tests performed with a generalized linear mixed-effect model showed significant differences between countries; compared to Argentina, mean proportion of diseased fruits were significantly higher in Brazil ($p<0.01$). It is important to note that the untreated plots reported in the literature are likely to correspond to heavily infected citrus orchards because these plots were primarily selected to test the effectiveness of fungicide treatments. For this reason, the values of disease incidence reported in Figure 38 and Table 10 should not be considered as representative of average situations, but rather as worst cases. However, these values reveal that very high disease incidence levels can be reached in countries where CBS is present.

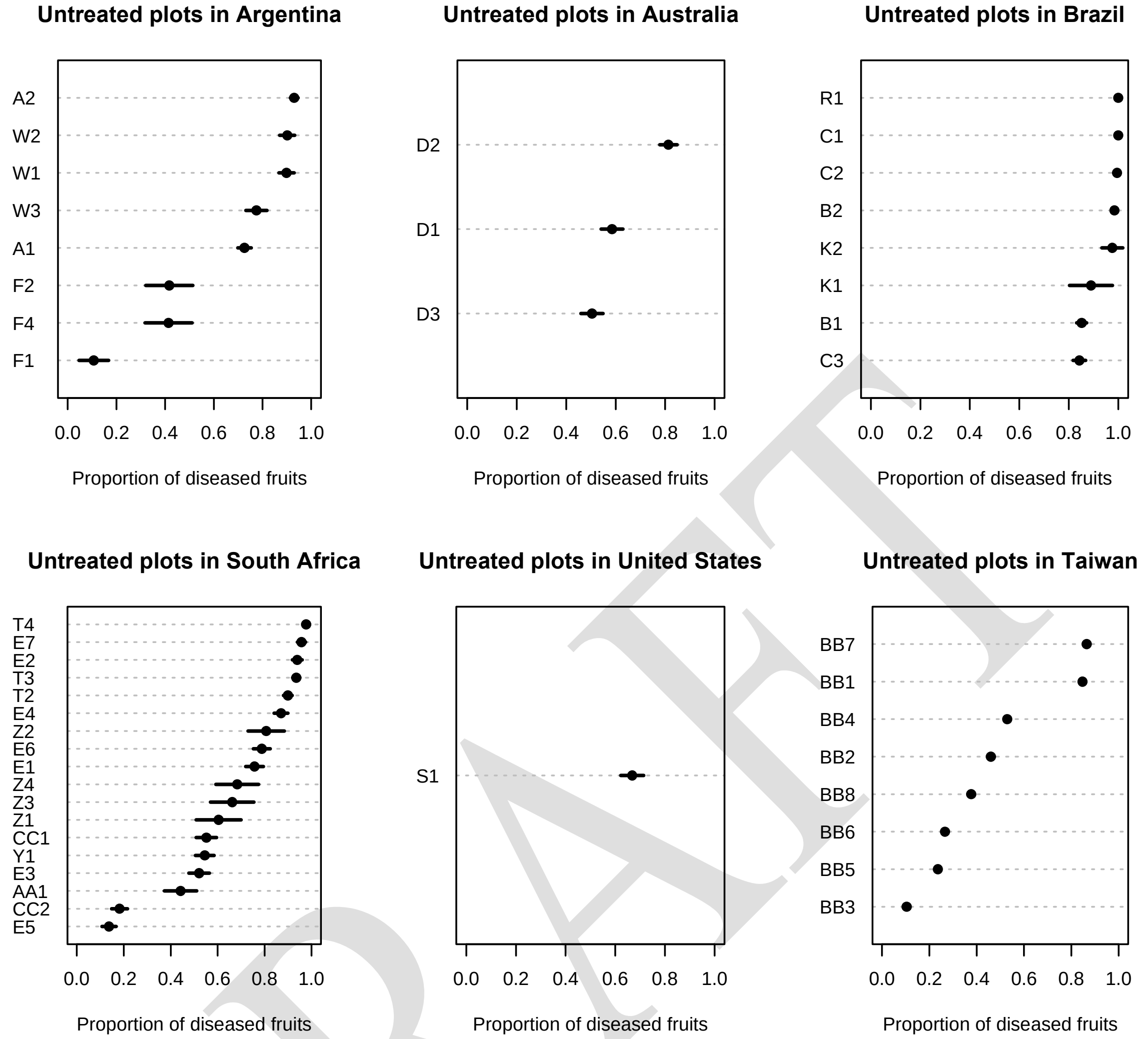

Figure 38: Proportion of CBS-affected fruits in untreated plots in Argentina, Australia, Brazil, South Africa, USA, and Taiwan. Plot names are given in the y-axis. Bars indicate 95% confidence intervals (missing for Taïwan).

Table 10: Proportions of CBS-affected fruits in untreated plots. Estimated median proportion, odd (ratio of the proportion of diseased fruits to the proportion of healthy fruits), and 95% confidence intervals between brackets (missing for Taïwan). Country effect was statistically significant ($p<0.01$).

Country	Number of plots	Estimated proportion of diseased fruits	Odds
Argentina	8	0.69 (0.44, 0.87)	2.26 (0.8, 6.42)
Australia	3	0.65 (0.48, 0.79)	1.84 (0.91, 3.7)
Brazil	8	0.98 (0.93, 0.99)	63.99 (13.04, 314)
South Africa	18	0.74 (0.59, 0.85)	2.87 (1.45, 5.68)
USA	1	0.67 (0.62, 0.72)	2.01 (1.63, 2.47)
Taïwan	8	0.46 (-)	0.85 (-)

Effects of fungicide treatments on disease incidence

Fungicide schedules were classified according to the chemical groups of the products evaluated and their combinations (FRAC, 2013): copper (“cu”), dithiocarbamates ("dit"), quinone outside inhibitors or strobilurines (“qoi”) and benzimidazoles (“ben”). The proportions of diseased fruits in plots treated with fungicide and in untreated plots are compared in Figure 39 and Table 11, for six different types of fungicide. Results show that fungicide treatments were systematically able to reduce the proportion of diseased fruits. However, the effectiveness of fungicide treatment varied a markedly between plots (Figure 39). In some plots, disease incidence was only slightly reduced by fungicide treatment, while the proportion of diseased fruits was reduced to zero in other plots. We tried to explain part of the between-plot variability of the fungicide effect by the number of sprays of fungicide using generalized linear mixed-effect model including the number of sprays as covariable (Figure 40). However, this model did not perform better (in terms of AIC and BIC) than a model including a fungicide effect but no covariable related to the number of sprays. The between-plot variability of the fungicide effect is thus probably not related to the number of sprays.

Odds ratios reported in Table 11 show that some types of fungicide were more efficient than others. The type of fungicide with the highest odd ratio (i.e., the least efficient fungicide type) was copper-based compounds (“cu”) while the types of fungicide showing the lowest odd ratios (i.e., the most efficient) were dithiocarbamates (“dit”), dithiocarbamates+strobilurines (“dit+qoi”), and copper+dithiocarbamates+benzimidazoles+strobilurines (“cu+dit+ben+qoi”). Fungicide treatments were all able to significantly reduce average diseased incidence ($p<0.001$) (Table 11), and the mean proportion of diseased fruits in treated plots ranged from 2.2% to 23% depending on the fungicide type (Table 11).

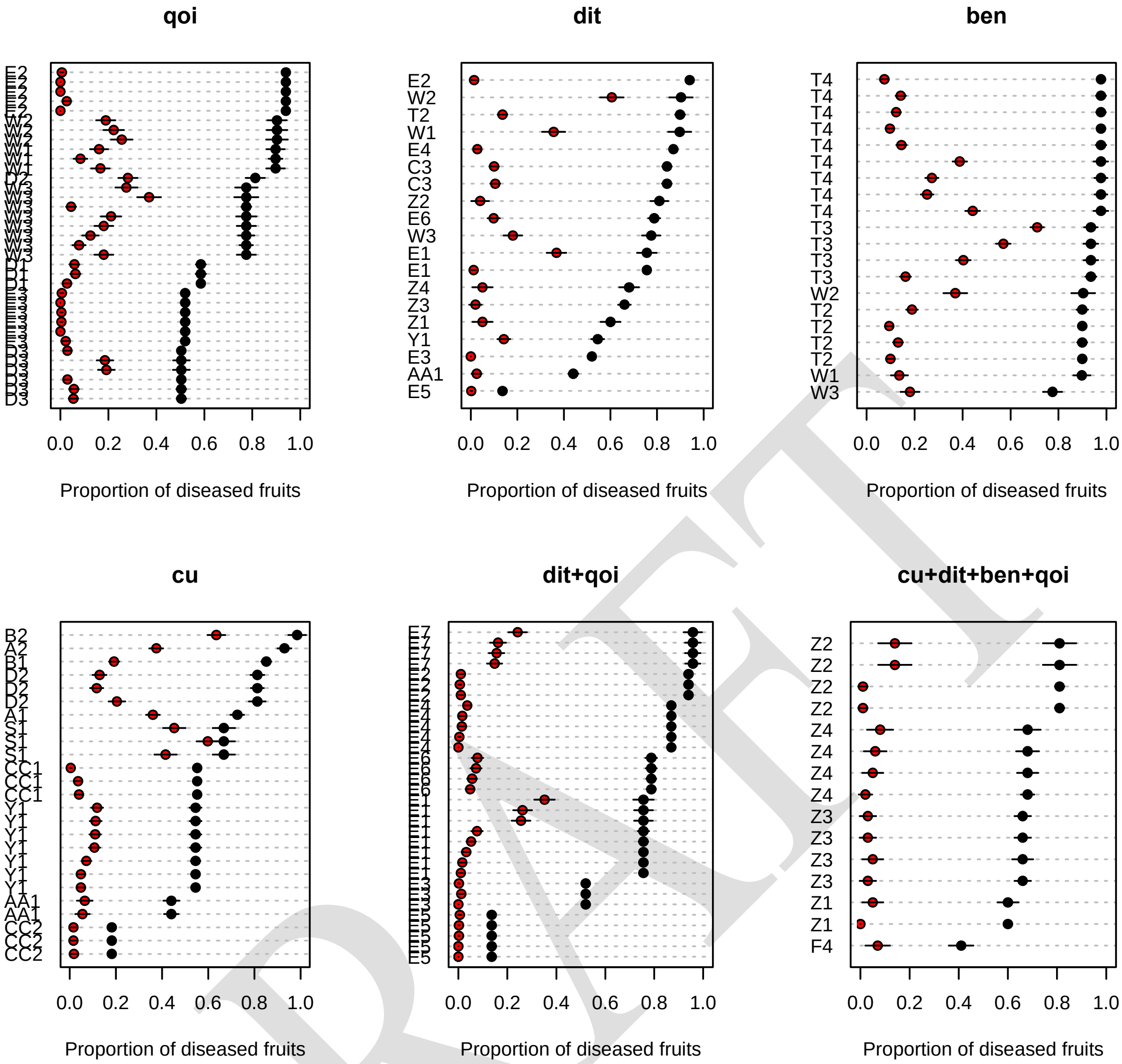

Figure 39: Observed proportions of CBS-affected fruits in untreated (black) and treated (red) trees for different types of fungicide (qoi, dit, cu, ben, dit+qoi, cu+dit+ben+qoi), and different plots. Bars indicate 95% confidence intervals. Plot names are given on the y-axis.

cu: copper
dit: dithiocarbamates
qoi: quinone outside inhibitors (strobilurines)
ben: benzimidazoles

Table 11: Estimated proportions of CBS-affected fruits in untreated plots and in plots treated with fungicides. Proportion of diseased fruits, odd (ratio of the proportion of diseased fruits to the proportion of healthy fruits), and odds ratio (ratio of the odds in treated plots to the odds in untreated plots) were estimated for six types of fungicide using a generalized mixed effect model including a fungicide effect but no covariable related to the number of sprays. 95 % confidence intervals are given between brackets. Effects of all types of fungicide were statistically significant (p<0.001).

Fungicide type	Number of plots	Untreated		Treated		Odds ratio
		Estimated proportion	Odds	Estimated proportion	Odds	
qoi	8	0.78 (0.64, 0.88)	3.6 (1.81, 7.16)	0.068 (0.03, 0.17)	0.07 (0.03, 0.21)	0.02 (0.01, 0.04)
dit	17	0.74 (0.63, 0.83)	2.89 (1.71, 4.9)	0.055 (0.02, 0.12)	0.058 (0.03, 0.14)	0.02 (0.016,0.03)
dit+qoi	7	0.77 (0.51,0.92)	3.34 (1.04,10.7)	0.022 (0.006,0.07)	0.022 (0.006,0.08)	0.0066 (0.004,0.011)
ben	6	0.91 (0.86,0.95)	10.7 (5.9, 19.4)	0.23 (0.15, 0.33)	0.3 (0.18, 0.51)	0.028 (0.02, 0.036)
cu	10	0.74 (0.52, 0.87)	2.77 (1.1, 6.97)	0.16 (0.07, 0.32)	0.19 (0.08, 0.48)	0.069 (0.059,0.08)
cu+dit+ben +qoi	5	0.64 (0.52,0.75)	1.79 (1.06,3.0)	0.047 (0.035,0.063)	0.049 (0.036,0.067)	0.027 (0.025, 0.03)

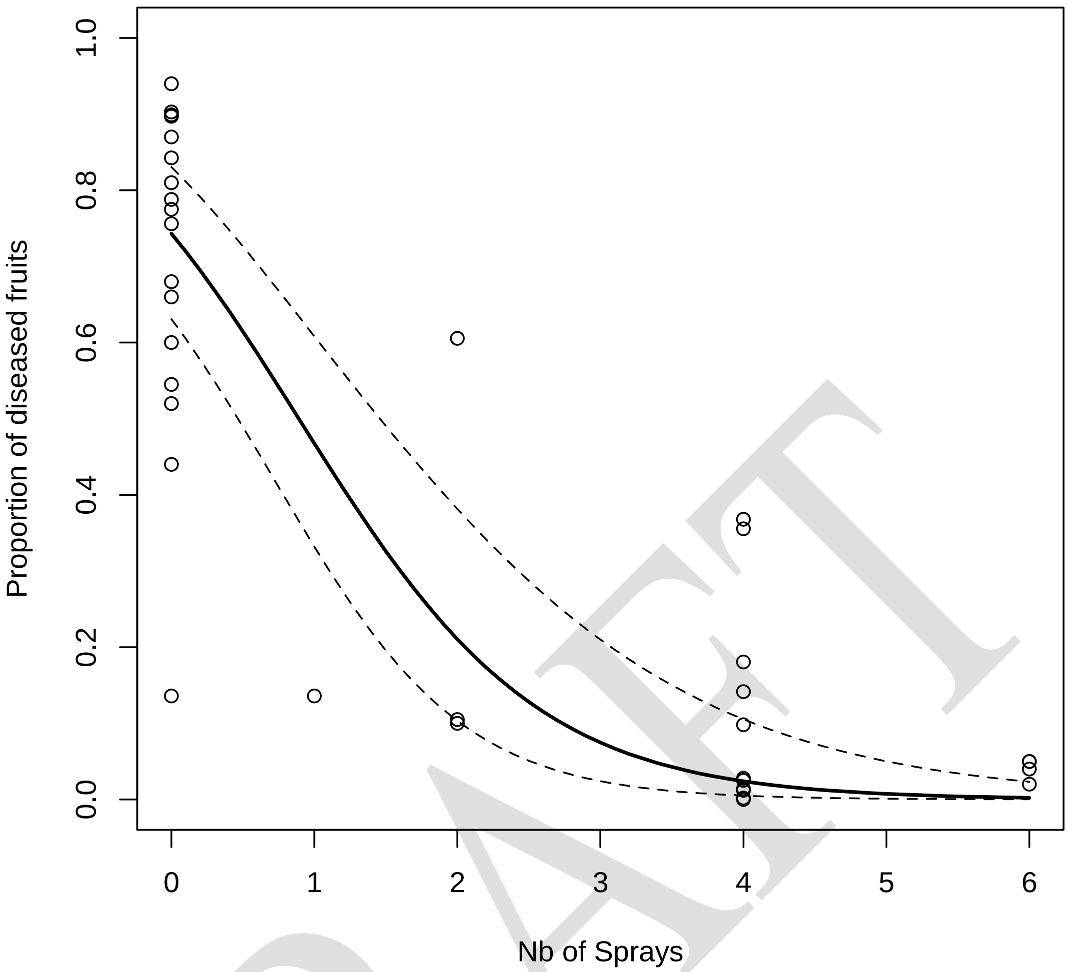

Figure 40: Proportion of CBS-affected fruits as a function of the number of sprays with dithiocarbamate fungicides. Points are data, continuous line shows the fitted curve, and the dashed lines indicate the 95% confidence intervals. The fitted model did not perform better in terms of AIC and BIC than the model including a fungicide effect but no covariable related to the number of sprays.

3.6.1.2. Pest effects on citrus crops in the risk assessment area

In a previous continent-wide simulation study, environmental conditions in EU citrus-growing areas were considered to be generally favourable for ascospore infection during the periods of April-May and September-October, when both susceptible leaves and fruit are widely available in the PRA area (Section 3.3.1.1). However, broader potential infection periods were obtained when data from on-site microclimatic weather stations located in citrus orchards were used (EFSA, 2008). Similar results were obtained for pycnidiospore infection. However, based on current knowledge the Panel considers infections from pycnidiospores mainly as relevant for establishing the first infection point following an entry of CBS-affected fruit into the risk assessment area (Section 3.3.2.6). According to the integrated results from the simulation of inoculum production and ascospore infection based on historical weather data, the Panel found that epidemics will develop only sporadically in time and space during spring in EU citrus-production areas. During the summer months, production of inoculum continues and tends to increase towards the end of the summer, while the weather is generally not conducive to infection. In autumn however, disease epidemics are predicted to happen again, and more regularly than in spring because the main proportion of the ascospores are released during the months of September and October which is also the time period of the year having the greatest percentage of hours with weather conducive to infection.

As described previously, *P. citricarpa* is known to have a relatively long incubation time, which means that it will take several weeks from when the infection takes place, until symptoms become visible on the fruit. After infection, the duration of the incubation period until symptom appearance is about 2 to 12 months, depending on environmental factors, tree age and condition (Brodrick and Rabie, 1970; Kotzé, 1981; Kotzé, 1963; Ninin et al., 2012). Fruit age at the time of infection has a considerable influence on the duration of the incubation period. Sweet orange fruits infected by *P. citricarpa* when they are 3 cm in diameter, needed about eight months to develop symptoms. However, fruits infected at 7 cm only needed two months to express symptoms (Aguiar et al., 2012). Therefore, the timing of epidemics compared to the harvesting calendar of the different citrus fruit varieties is a key factor for the assessment of direct pest effects.

Figure 41 provides the harvesting calendar for citrus fruit in the EU citrus-production areas. It is apparent that fruit infected in September-October would eventually develop symptoms in the field only if they hang on the tree for some months after infection. Early-maturing mandarins like Satsumas, that are usually harvested in September and October, may not have enough time for symptom development in the field, and will already be harvested and even also be consumed before symptoms become visible. Nevertheless, latent infections might develop after harvest during transport and storage. Mid season and late maturing mandarins and sweet oranges would stay on the tree for several months after infection in September-October, especially the sweet orange cultivars of the 'Valencia' group which are harvested as late as May or June. Cultivar field trials conducted in Brazil as well as studies comparing the rate of disease progress indicated that cultivar reaction to the disease is more linked to the interaction of environmental factors with the dynamics of fruit maturation (Sousa and de Goes, 2010; Sposito et al., 2004), than cultivar resistance. Lemons have several flowerings per season, so fruits with different growth stages would be present at the time of potential infection by *P. citricarpa*.

These conclusions are in line with the available literature, which indicates that lemons and late maturing sweet orange cultivars are most affected by CBS (Kotzé, 1981; Timmer, 1999; Spósito et al., 2004)

For the rarer events of disease development starting in spring, e.g. during the month of May, these epidemics will be most damaging, because there will be sufficient time for disease symptoms to develop on the fruits on both the early, and the late maturing citrus-varieties and lemons. Due to this differentiation, the Panel has decided to also differentiate the risk rating accordingly, with minor consequences for early maturing citrus-varieties, while the risk of direct pest effects is characterised as moderate for the late maturing citrus.

The EU citrus industry is strongly oriented towards the production of fruit for the fresh market (Agustí, 2000; Cutuli et al., 1985). According to international quality standards (Section 3.1.1.5), the presence of more than one spot per fruit is considered detrimental to the quality, and fruits with more than six spots are considered to be out of grade (OECD, 2010). Thus, even relatively low disease severities would cause significant negative impacts in the fresh fruit industry. The presence of fruit spots is not a major factor for the citrus processing industry, so CBS is not expected to cause major impacts in this sector. Under high disease pressure conditions in Brazil, CBS also induces premature fruit drop (Reis et al., 2006), which impacts both the fresh fruit and processing industries. However, the information available on CBS does not indicate that the disease will reach such high levels of disease intensity in the EU endangered areas.

In EU citrus-growing areas, early and late maturing cultivars are generally grown together in the same regions, so the areas indicated in Figures 25 and 26 can be considered to show the potential geographical limits to impacts. In Spain, lemons are mainly grown in the south of Alicante and Murcia provinces (Agustí, 2000). In Italy, lemon production is mainly concentrated in the island of Sicily (Cutuli et al., 1985). As this citrus species is considered to be the most susceptible to CBS, these regions might experience higher impacts than other citrus-growing areas in the EU. Nevertheless, new cultivars are being introduced by the European citrus industry to extend the harvesting period, so the diversity of cultivated genotypes in the EU is changing and new varietal scenarios may be expected in the future (Aleza et al., 2012).

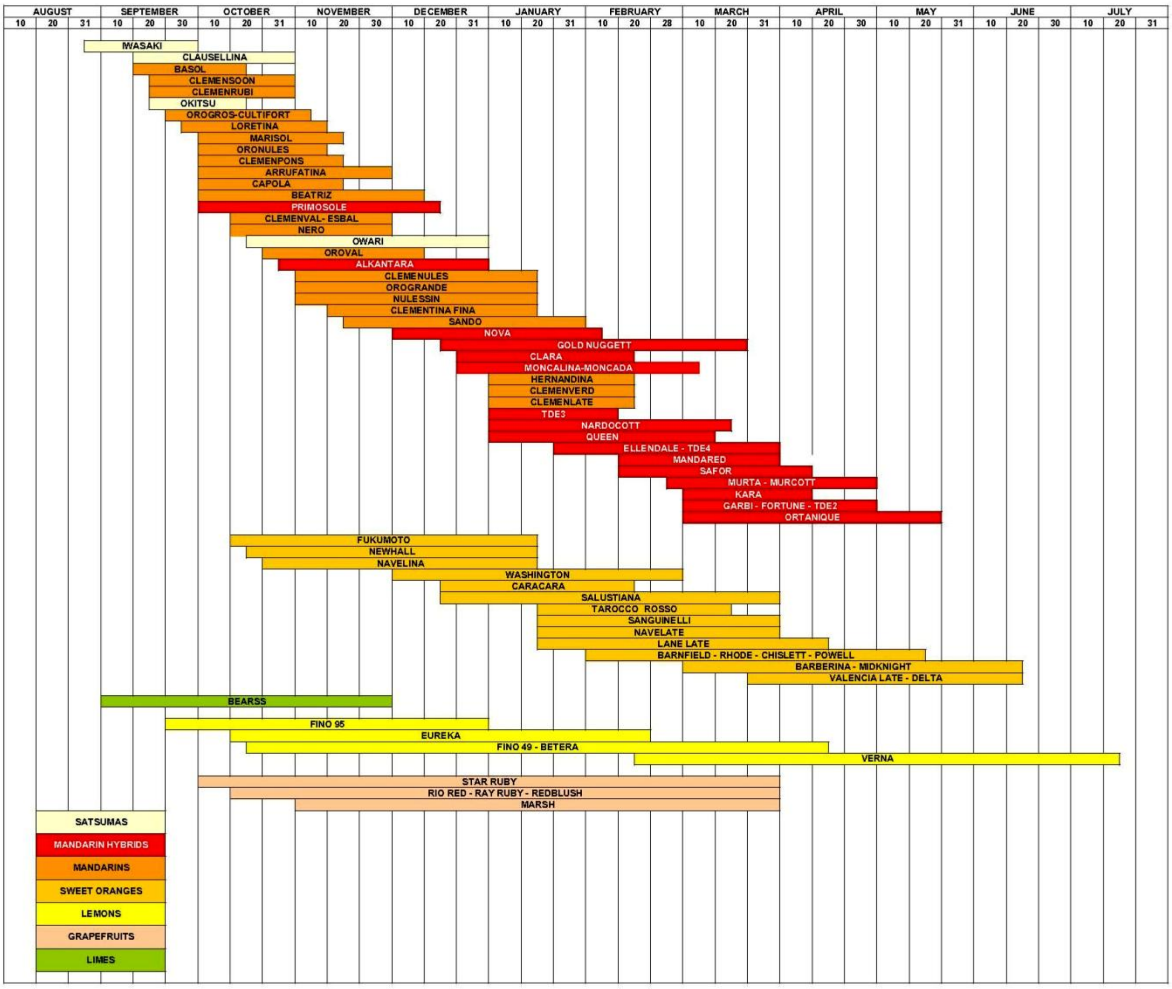

Figure 41: Harvesting calendar of citrus fruit in the EU citrus-production areas (Pardo et al., 2013)

3.6.2. Control

Agronomic practices, such as leaf litter and soil cover management, irrigation and early fruit harvesting, are used to some extent for CBS control in the areas where the disease is present (Kotzé, 1981; Timmer, 1999; Miranda-Bellote et al., 2013). However, chemical control involving the use of protective and curative fungicides is generally necessary for economic disease management (Kotzé, 1981; Sposito et al., 2011). In order to obtain a summary of the fungicide schedules used for CBS control elsewhere and to quantify their efficacy, data were obtained from fungicide control trials conducted in areas of Australia, Argentina, Brazil, South Africa and Taiwan where CBS is present (Section 3.6.1.1). Results from the meta-analysis showed that copper-based compounds were the least efficient fungicides for the control of CBS. The highest disease control levels were obtained with fungicide schedules including dithiocarbamates, benzimidazoles and strobilurines (QoI).

Copper compounds and mancozeb (dithiocarbamate) are the only fungicides currently registered for citrus in the EU (Directive 91/414/CEE) than may have some effect against *P. citricarpa*. Strobilurin fungicides (QoI) and benzimidazoles, which are highly effective for CBS control, are not currently labeled for citrus in the EU, and their future use will depend on private or public funding resources to cover the registration costs. Assuming a potential infection period of about two months in September and October, between one and two fungicide sprays would be necessary to protect the fruit, depending on rainfall and other meteorological factors. For some cultivars, compliance with maximum residue limits (MRL) will be challenging considering the time lapse between the timing of fungicide application and harvest. In years having an infection period also in spring, between one and two additional fungicide sprays would be needed during this season for effective disease control.

3.6.3. Environmental consequences

In addition to the economic costs of the fungicides and their application in the orchards, environmental side-effects should be also considered. Environmental consequences are envisaged due to the additional fungicide treatments required for the control of *P. citricarpa* once the pathogen is established (Cunha et al., 2013). Copper compounds and mancozeb have been associated with environmental concerns (Alva et al., 1993; Houeto et al., 1995) and in fact, the use of copper in organic production in the EU is strictly limited (Regulation EC/473/2002) to reduce environmental pollution of soil and changes in microbial communities (Zhou et al., 2011). Moreover, the effective life of fungicide is shortened if they are used more frequently, reducing their effectiveness for management of other diseases and jeopardizing the effectiveness and sustainability of IPM approaches (van den Bosch and Gilligan, 2008).

3.6.4. Indirect pest effects

In case of *P. citricarpa* establishment in the EU, indirect pest effects could be linked to the need of implementing eradication and/or containment measures to prevent establishment and spread to other EU citrus growing areas (section 3.6.4.1, below), as well as additional fungicide treatments and/or quality controls in packing houses for citrus fruit exported to non-EU citrus growing areas.

3.6.4.1. Indirect pest effects: eradication and/or containment

Although eradication has never been proved successful for this disease, containment measures may be needed to prevent or limit the spread of *P. citricarpa* (Section 3.4.4) to other citrus growing areas, if the disease became established in an EU location. These measures are described in details in Section 3.4.4 and further evaluated in Section 4 of this opinion.

3.6.4.2. Indirect pest effects: additional fungicides treatements and quality controls for export of citrus fruit

During the last 6 years (2007-2012), citrus fruit was exported from the EU (EU 27) to 38 countries where CBS is not present and where citrus is also cultivated in commercial orchards (Figure 38). Among these 38 countries, in five of them (Bosnia and Herzegovina, United Arab Emirates, Montenegro, Russia and Algeria), citrus export were larger than 3,500 tons/year.

Table 12: List of countries where EU citrus fruit is exported. Only those countries where citrus cultivation exists and Citrus Black Spot is not present are listed. Data are yearly average values for the period 2007 to 2012.

Country	Average yearly export of citrus fruit from EU 27 (tonnes/year)
Russia	129,789
Bosnia and Herzegovina	25,951
Algeria	10,749
United Arab Emirates	5,322
Montenegro	3,563
Angola	793
Turkey	653
Philippines	588
Malaysia	538
Korea	492
Costa Rica	470

Kuwait	301
Japan	276
Mexico	236
Colombia	199
Liberia	198
Azerbaijan	196
Georgia	172
Libya	156
Cote D'ivoire	140
Panama	108
Jordania	103
Afghanistan	101
Mali	98
Honduras	75
Senegal	62
El Salvador	55
Egypt	45
Mauritius	30
Congo	27
Dominican Republic	27
Israel	23
Morocco	21
Iraq	20
Madagascar	16
Lebanon	12
Eritrea	3
Chile	1

Among these 38 countries, Turkey, Jordan, Israel and Chile are currently regulating *P. citricarpa* as a quarantine organism in citrus fruit commodities (see Section 3.1.4). In addition, *P. citricarpa* is in the A1 List of the Caribbean Plant Protection Commission (CPPC) and in the A2 Lists of Asia and Pacific Plant Protection Commission (APPC), Comitè de Sanidad Vegetal del Cono Sur (COSAVE), Interafrican Phytosanitary Council (IAPSC) and Pacific Plant Protection Organisation (PPPO). Thus if *P. citricarpa* became established in the EU, to export citrus fruit, additional fungicide treatments in the orchards, official inspections, quality controls in packinghouses and/or establishment of pest free areas might be needed to meet the phytosanitary requirements of these countries.

3.6.5. Conclusion of the assessment of consequences

Rating	**Justification**
Moderate for fresh fruit of late maturing citrus varieties and lemons	Due to the required incubation time with a minimum of 2 months and to results from the simulations showing more frequent autumn infection, late maturing citrus varieties and lemons are likely to express more symptoms in the field. The main impact will be on quality for the fresh market (fruit with more than 1 lesion is reduced in quality and with more than 6 lesions is not suitable for the fresh market). There would a potential for reduction in disease incidence by chemical treatment, but this would cause environmental impacts owing to the fact that in most EU citrus growing areas fungicides are not widely applied and that the most effective fungicide products are not currently registered for use in citrus by the EU MSs. In addition, to export citrus fruit to areas where CBS is regulated, additional fungicide treatments in the orchards, official inspections, quality controls in packinghouses and/or establishment of pest free areas might be needed to meet the phytosanitary requirements of

	these countries.
Minor for fresh fruit of early maturing citrus varieties	The impact on early maturing varieties would be sporadic in time and space, limited to years with rainy springs and/or to specific locations. However the impact could be higher in areas where spring infection, based on simulation results, is expected to be more frequent, such as some locations in Southern Spain, Cyprus, Malta and Greek islands.
Minimal for citrus for processing	External lesions or spots on citrus fruit are not a quality issue for citrus for processing.

3.6.6. Uncertainties on the assessment of consequences

Rating	**Justification**
High	High uncertainties about the time from infection to symptom expression (incubation period) High uncertainties due to: the lack of information on key parameters in the epidemiological models, the lack of knowledge about the rate of disease dynamics and inoculum build-up for this pathogen, especially in marginal areas within the current area of distribution, e.g. Eastern Cape Province of South Africa, where environmental conditions is more similar to those in the risk assessment area; the limited information available about the impact and the fungicides treatments in these marginal areas.

3.7. Conclusion and uncertainties of the pest risk assessment

Under the scenario of absence of regulations, the conclusions of the pest risk assessment are as follows:

<u>Entry</u>

The probability of entry is rated as:

- moderately likely for the citrus fruit trade pathway (medium uncertainty),
- very unlikely for the Tahiti lime (*Citrus latifolia*) fruit trade pathway (high uncertainty),
- unlikely for citrus fruit import by passengers traffic pathway (medium uncertainty),
- likely for the citrus fruit with leaves trade pathway (medium uncertainty),
- likely for the citrus plants for planting trade pathway (low uncertainty),
- likely for the Tahiti lime (*Citrus latifolia*) plants for planting trade pathway (high uncertainty),
- likely for the citrus plants for planting import by passengers traffic (medium uncertainty),
- unlikely for the citrus leaves (medium uncertainty)

<u>Establishment</u>

The probability of establishment is rated as moderately likely because of:

- the widely availabity of suitable hosts (no uncertainty),
- the climate suitability for ascospores maturation, dispersal and infection of many EU citrus growing areas in September and October and for specific location also in May (high uncertainty),
- cultural practices (fungicides) not preventing establishment (low uncertainty),
- sprinkle and micro-sprinkle irrigation (still used in part of the EU citrus growing areas) favouring establishment (low uncertainty),
- the simultaneous occurrence of host susceptibility and of weather conditions suitable for ascospore production and release and weather conditions for ascospore germination and infection (high uncertainty).

Overall, uncertainty on the probability of establishment is rated as high, mainly due to lack of knowledge on how *P. citricarpa* will respond under the EU climatic conditions. Although it is known for the organism which environmental factors are important in the various stages of the life cycle, there is lacking scientific evidence to precisely determine the exact threshold values the organism require, e.g. for the temperature and wetness levels and durations. Further validation of the models applied by the Panel, especially for marginal areas within the current distribution of the citrus black spot disease would be needed to reduce the uncertainty on the probability of establishment of *P. citricarpa* in the EU.

Spread

Natural spread of *P. citricarpa* is known to mainly happen by airborne ascospores. The distances it can spread by natural means are poorly known, The pathogen is very likely to spread with human assistance along the commercial fruit and plants for planting pathways. However, because spread is defined as the expansion of the geographical distribution of a pest within an area, the rate of spread depends not only on the rapidity of movement and the number of spread pathways but also the likelihood of finding a suitable environment for establishment. When the proportion of the citrus growing areas identified as potentially suitable for *P. citricarpa* is taken into account, the Panel considered that a rating of moderately likely is most appropriate for spread

There is uncertainty about the potential natural spread of ascospores carried by wind over long distances, but this uncertainty does not concern the two main pathways of spread (intra-European trade of commercial fruit and plants for planting). There is uncertainty about the structure of the EU intra-trade network for the citrus plants for planting owing to lack of data, however this does not influence the conclusions above.

Endangered area

The risk assessment has identified parts of the EU where host plants are present and where, based on simulation results, the climatic conditions are suitable for ascospore maturation and release followed by infection.

Conclusions from simulations of the release of ascospores based on gridded interpolated climate data of the EU citrus producing areas show that, in almost all years (for the 95 percentile), ascospore release in areas of Cyprus, Crete, Southern Greece, Italy, Spain and Portugal will start early enough to coincide with climatic conditions that are conducive to infection in September and October as simulated by EFSA (2008). However the same simulations indicate that the onset of ascospore release in most areas will start too late to coincide with the climatic conditions conducive to infection in April-May. Therefore, early maturing orange varieties might generally be infected in autumn only, which is when the availability of inoculum coincides with suitable conditions for infection. Due to the long incubation time, fruits from these early varieties will be harvested before symptoms appear. The late maturing oranges varieties and lemons are expected instead under such scenario to show CBS symptoms.

There are some areas however, such as locations in Cyprus, Greek islands, Malta and Southern Spain where onset of ascospores is expected also in May in half of the years simulated. In those years, it is expected that symptoms can develop on the fruit before harvest, and therefore have an impact on the fruit quality.

The results from the simulations on interpolated (grid-based) weather data are consistent with the simulations run on weather data measured by agrometereological stations. The uncertainty is high as indicated in the establishment section.

<u>Consequences</u>

The results from the simulation of ascospore maturation, release and infection show that citrus black spot will develop and express symptoms mainly in late maturing sweet orange varieties and lemons grown within the endangered area. The expected consequences will be moderate for fresh fruit of late maturing citrus varieties and lemons. There would a potential for reduction of disease incidence by chemical treatments, but this would cause environmental impact owing to the fact that in most EU citrus growing areas fungicides are not widely applied and that the most effective fungicide products are not currently registered for use in citrus in the EU MSs. In addition, to export citrus fruit to areas where CBS is regulated, additional fungicide treatments in the orchards, official inspections, quality controls in packinghouses and/or establishment of pest free areas might be needed to meet the phytosanitary requirements of these countries.

The consequences for fresh fruit of early maturing citrus varieties are assessed as minor. The impact on early maturing varieties would be sporadic in time and space, limited to years with rainy springs and/or to specific locations. However the impact could be higher in areas where spring infection, based on simulation results, is expected to be more frequent, such as some locations in Southern Spain, Cyprus, Malta and Greek islands.

The consequences would be minimal for citrus for processing, as external lesions or spots on citrus fruit are not a quality issue for the citrus for processing.

As for establishment, the uncertainties about consequences are high due to: the lack of information on key parameters in the epidemiological models and on te incubation period, the lack of knowledge about the rate of disease build-up for this pathogen; the limited information available about the impact and the fungicides treatments in marginal areas within the current CBS area of distribution, eg Eastern Cape, where environmental conditions are more similar to those in the pest risk assessment area.

4. Identification of risk reduction options and evaluation of their effect on the level of risk and of their technical feasibility

Section 4 assesses the effectiveness of potential options for reducing the risk of entry, establishment and spread of *P. citricarpa* following the ‘Guidance on methodology for evaluation of the effectiveness of options to reduce the risk of introduction and spread of organisms harmful to plant health in the EU territory’ (EFSA Panel on Plant Health (PLH), 2012). Section 4.1 first presents a systematic evaluation of options for reducing the probability of entry. This considers all the entry pathways analysed in sections 3.2.2 – 3.2.9. Section 4.2 evaluates the options for reducing the probability of establishment while section 4.3 evaluates options for reducing the probability of spread. Section 4.4 discusses the effectiveness of combining risk reduction options. The effectiveness of current EU phytosanitary measures is evaluated in section 4.5.

4.1. Systematic identification and evaluation of options to reduce the probability of entry

In this section, options to reduce the probability of entry of *P. citricarpa* are systematically identified and evaluated. Each of the eight introduction pathways described in the entry part of this opinion (sections 3.2.2 – 3.2.8) is considered and the citrus fruit commercial trade (section 4.2.1) and citrus plants for planting (section 4.2.5) pathways are analysed in detail. For these pathways, 14 potential risk reduction options identified by the EFSA Panel on Plant Health (PLH) (2012) have been evaluated as a stand-alone measure, assuming that other risk reduction options are not in force for that pathway or for the other pathways. The risk reduction options considered are listed in Table 13. This checklist has been followed to ensure that no options are overlooked and consistency and objectivity is maximized between opinions. For each risk reduction option[13] (RRO), the Panel assessed its *effectiveness* and *technical feasibility* together with the *uncertainty* in the ratings given. The effectiveness of a systems approach, integrating two or more independent RROs, is discussed in section 4.4.

Table 13: Potential risk reduction options, listed by the EFSA PLH Panel (2012) and used for this opinion

Options for consignments
1. Prohibition
2. Prohibition of parts of the host or of specific genotypes of the host
3. Pest freedom of consignments: inspection or testing
4. Pre- or post-entry quarantine system
5. Preparation of the consignment
6. Specified treatment of the consignment/reducing pest prevalence in the consignment
7. Restriction on end use, distribution and periods of entry
Options preventing or reducing infestation in the crop at the place of origin
8. Treatment of the crop, field or place of production in order to reduce pest prevalence
9. Resistant or less susceptible varieties
10. Growing plants under exclusion conditions (glasshouse, screen, isolation)
11. Harvesting of plants at a certain stage of maturity or during a specified time of year
12. Certification schemes
Options ensuring that the area, place or site of production at the place of origin remains free from the pest
13. Limiting import of host plant material to material originating in pest-free areas (PFAs)
14. Limiting import of host plant material to material originating in pest-free production places or pest-free production sites

[13] Hereinafter referred to as RRO

Prohibiting the import of consignments in theory closes a pathway, making all other RROs for that pathway redundant. The *effectiveness* of this RRO is **very high** for all pathways. The *technical feasibility* is **high** for all pathways because it already is, or can be, implemented in customs operations and phytosanitary import procedures. The level of *uncertainty* is **low**, for all pathways.

The effectiveness of individual RROs in one pathway on the overall probability of entry (via all pathways) is not discussed, nor is the effectiveness of an individual RRO in one pathway compared with RRO(s) in one or more other pathways. To undertake such a complex evaluation, ideally a fully quantitative probabilistic pathway model would be required. For example, the effectiveness of the treatment of consignments of citrus fruit in commercial trade in reducing the overall probability of *P. citricarpa* entry has not been compared to the effectiveness of post-entry quarantine for citrus plants for planting. However, it should be kept in mind that the overall reduction of probability of entry of *P. citricarpa* is determined by the combined set of RROs for all pathways.

4.1.1. RROs to reduce entry along the citrus fruit commercial trade (pathways I, II and IV)

This section deals with the identification and evaluation of RROs to reduce the probability of entry of *P. citricarpa* along the three pathways of citrus fruit commercial trade described in the entry section: the pathway (I) of commercial trade of citrus fruit (excluding Tahiti lime and citrus fruit with leaves), the pathway (II) of commercial trade of Tahiti lime fruit and the pathway (IV) of commercial trade of citrus fruit with leaves and peduncles (for a detailed description and analysis of these pathways please see sections 3.2.2, 3.2.3 and 3.2.5, respectively).

The results of this evaluation are summarised in Table 14.

A. Options for consignments

4.1.1.1. Prohibition

The prohibition of the import of all citrus fruit along the three pathways of commercial trade would prevent the entry of *P. citricarpa* into the EU along these pathways. The *effectiveness* is assessed as **very high**. The *technical feasibility* is **very high**, because it can be implemented in customs operations and phytosanitary procedures. The *uncertainty* on these ratings is assessed as **low**.

4.1.1.2. Prohibition of parts of the host or of specific genotypes of the host

a) Prohibition of parts of the host

This option would prohibit the import of citrus fruit with leaves and peduncles (pathway IV), therefore leading to an effective block on this pathway. The *effectiveness* is assessed as **high**. The *technical feasibility* is very **high**, because it is already implemented in customs operations and phytosanitary procedures. The *uncertainty* on these ratings is assessed as **low**.

b) Prohibition of specific genotypes of the host

All commercial citrus species and cultivars are considered to be susceptible to CBS, except for sour orange (*C. aurantium*) and Tahiti lime (*C. latifolia*) (see section 3.1.1.4). The host status of pomelo (*C. maxima*) is still uncertain (see section 3.1.1.4). The prohibition of import of fruit of susceptible citrus varieties would therefore default to a general prohibition of all citrus fruit except for Tahiti lime and sour orange fruit.

The *effectiveness* in reducing the probability of entry of *P. citricarpa* via this pathway is **very high**, as, considering the current trade flows, it is almost equivalent to the general prohibition of import of fruit from all citrus species. The *technical feasibility* is **moderate**, because fruits of sour orange and Tahiti limes cannot be clearly identified at import inspection unless inspectors are well trained or equipped with tools for fruit analysis. The *uncertainty* for these ratings is **medium**, owing to some publications indicating that sour orange and Tahiti lime can be colonised by *P. citricarpa* (see section 3.1.14).

4.1.1.3. Pest freedom of consignments: inspection or testing

The detection of *P. citricarpa* in consignments is based on visual inspection, sampling and laboratory testing. Inspection and sampling of the consignment should be performed according to guidelines in the IPPC Standards ISPM No 23 Guidelines for inspection (FAO, 2005) and ISPM No 31 Methodologies for sampling of consignments (FAO, 2009). For laboratory testing, *P. citricarpa* -specific detection methods have been developed (see section 3.1.1.3). Inspection or testing of consignments may be applied at the time of export and/or at the time of import. At export, inspection or testing may serve as a stand-alone measure without other official measures for production, harvest and packaging or as a measure to verify that other measures have been effective. At import, inspection generally serves to verify phytosanitary measures taken by the exporting country.

The CBS disease is characterized by a long incubation period (50 – 200 days; see 3.1.1.2). Fruit symptoms become visible only several months after infection and may not yet have appeared at the time of inspection (at export or at import). Infested lots may pass these inspections unnoticed, limiting the effectiveness of visual inspection. Laboratory testing using molecular procedures is generally required for the detection of latent infections and for accurate identification of the pathogen. Following the recent discovery of new *Phyllosticta* species on citrus, new molecular methods need to be validated and implemented routinely (see 3.1.1.3). The effectiveness of both visual inspection and laboratory testing for detection of *P. citricarpa* in consignments of citrus fruit depends on the sampling method and the sample size. No method will provide 100% effectiveness of detection. The effectiveness of visual inspection alone is further limited by the possible presence of latent or mildly infected fruits escaping detection in the sample.

The visual inspection of consignments combined with laboratory testing is effective in reducing the probability of entry of *P. citricarpa* along the citrus fruit commercial trade pathways, particularly when up to date molecular methods are used and a sampling procedure that gives high confidence in detecting low disease incidence is employed The *effectiveness* of visual inspection combined with laboratory testing is assessed as **moderate,** because of the occurrence of latent infections that will be overlooked by visual inspection.

The *technical feasibility* of visual inspection is **moderate,** owing to the huge volumes of imported citrus fruit that would have to be inspected to give a high confidence. Also some EU MSs may be following a reduced check regime on citrus under 1756/2004/EC (this is a voluntary system that can be applied by MSs if interceptions have only been found at a very low level in a large number of consignments inspected over the previous three years). The technical feasibility of routine laboratory testing for exported and imported consignments is **moderate** due to the relatively long duration of the laboratory procedures, although the development of new methods may reduce the time required (Tomlinson et al., 2013).

The *uncertainty* for these ratings is **medium** because of: the lack of knowledge on the proportion of CBS latent infection in citrus fruit consignments (see section 4.1.1.1 below), the lack of an estimate of the incidence of CBS in imported consignments and of a detailed analysis of the practical implementation of the inspections at EU ports together with the recent description of new *Phyllosticta* species associated with citrus for which validated molecular detection methods are not always available.

4.1.1.4. Pre- or post-entry quarantine system

Pre- or post-entry quarantine systems are not applicable to citrus fruit commercial trade at the ports of the exporting or importing country, due to the size of the consignments and to the difficulty of storing citrus fruit for long period to make the expression of symptoms possible.

Regarding pre-harvest inspections, Baldassari et al. (2007) have shown that treating asymptomatic fruit of orange 'Pêra-Rio', aged between 20 and 28 weeks after flowering by immersion in a solution of ethephon (2.10 g/l, 1 min) induced precocious symptom expression (assessed 28-35 days after treatment) of *P. citricarpa* in proportions equivalent to those observed in fruit matured on trees. This system applied to field samples of asymptomatic citrus fruit allows the detection of latent infections of *P. citricarpa* in advance in the country of origin before harvest and export. This technique could be applied in the country of origin before harvest with **high** *effectiveness* and **high** *technical feasibility*, with **medium** *uncertainty* due to the lack of information on the field sampling protocol applied.

4.1.1.5. Preparation of the consignment

The preparation of the consignment includes several stages, beginning with the handling of harvested fruit and transport to the packing station, to the closing of boxes or other packaging material prior to export. Specific conditions and procedures, particularly culling, may be implemented during this process to reduce the presence of *P. citricarpa* infected units in the consignment. Management procedures at citrus fruit packing stations can play an important role in reducing the incidence of CBS infected fruit in consignments. Packing stations should be registered and employ a system of record keeping, enabling quality control of packing house operations and the tracking and tracing of consignments from the production site and the recording of information on the disease management program. Fruit originating from official pest free areas and official pest free places of production should be packed at dedicated packing stations where handling of fruit from other places of production is not allowed.

The culling and cleaning of fruit may allow the removal of leaves, peduncles, other debris and many (but not all) symptomatic fruits. However,the *effectiveness* of this option when applied alone is assessed as **low,** because of the existence of latent infections and the similarity of unspecific CBS symptoms with those caused by other citrus pathogens as well as by mechanical or insect damage (see section 3.2.2.1). The *technical feasibility* is **very high** since such measures are currently implemented in citrus producing countries. The *uncertainty* of these ratings is **medium** due to the limited knowledge on the proportion of CBS latent infection in citrus fruit consignments.

4.1.1.6. Specified treatment of the consignment/reducing pest prevalence in the consignment.

During the preparation of consignments of citrus fruit, several treatments, such as waxing or hot water treatments, may be applied that can reduce or delay the post-harvest development of CBS symptoms, but they are unlikely to eliminate the pathogen (see section 3.2.2.1). Methods that eliminate *P. citricarpa* from infected fruit are not available.

It is recommended that registered packing houses have an approved system in place to limit the build-up in the treatment tank of extraneous organic matter, including leaves, twigs, grass, weed, soil, slime or any other material that would interfere with the treatment.

The *effectiveness* of post-harvest chemical treatments alone is **low**, the *technical feasibility* is **very high**, since such treatments are currently implemented in fruit packing houses, and the *uncertainty* is **low**.

4.1.1.7. Restriction on end use, distribution and periods of entry

It is not possible to identify periods of the year when the harvested citrus fruit in the country of origin is uninfected. Therefore the *effectiveness* of a restriction in the period of import of citrus fruit, to reduce the probability of entry of the pathogen, is assessed as **negligible**, although the *feasibility* is **very high**, with **low** *uncertainty*.

Another option would be restricting the end use of citrus fruit imported in the EU to fruit processing facilities. However, in the citrus producing EU MSs, implementing this option would not lead to a reduction of risk but potentially to an increase of risk, because large amounts of citrus cull fruit and waste, from areas where CBS occurs, would be concentrated around the citrus processing plants located in citrus growing areas (see Section 3.2.2.4). The *effectiveness* would therefore be **negligible**. *Feasibility* would be **negligible** due to the complex trade network of citrus fruit within the EU and the need to develop a traceability system to secure the intended end use (i.e. keep distinct the citrus for processing from the citrus for fresh consumption). *Uncertainty* is **medium** due to lack of information on the potential re-distribution of citrus fruit for processing as fresh fruit for consumption.

A restriction on the distribution of citrus fruit imported into the EU from areas where CBS occurs to areas in the EU without citrus production or where climate conditions are unsuitable for the development of CBS, might reduce the probability of transfer to a suitable host. However, the complex network of the intra-EU trade in fresh citrus fruit shows large volumes of citrus fruit traded among EU MSs (Section 3.4.2). Fruit imported from Third Countries into EU MSs without citrus production and subjected to import inspection may subsequently be traded to citrus producing areas of the EU without further inspections. For example, in 2009 the Netherlands imported around 450,000 tons of sweet orange and 170,000 tons of grapefruit from various countries (including Florida, Argentina, Brazil and Uruguay) and re-distributed almost 200,000 tons of sweet orange and 115,000 tons of grapefruit to other EU MSs, including citrus producing countries (Eurostat). Because of the complex trade network and high trade volumes of citrus fruit within the EU, the implementation of differentiated import requirements for EU MSs without citrus production compared to citrus producing EU MSs has a **high** *effectiveness* and **negligible** *technical feasibility*. The *uncertainty* of these ratings is **low.**

B. Options preventing or reducing infestation in the crop at the place of origin

4.1.1.8. Treatment of the crop, field or place of production in order to reduce pest prevalence.

Fungicide treatments against the infestation by *P. citricarpa* in orchards at the place of origin can reduce the incidence of the pathogen, but will not eliminate it (see the meta-analysis of CBS control trials in section 3.6.1.1). Culling and cleaning of fruit removes leaves, peduncles and many (but not all) symptomatic fruits but is not effective in reducing the presence of asymptomatic latent infections. The *effectiveness* of treatments of the crop and orchards at the origin is thus **moderate**. The *technical feasibility* is **very high**, given that these treatments are already applied. The *uncertainty* for these ratings is **low**.

4.1.1.9. Resistant or less susceptible varieties.

All citrus species and cultivars grown for fresh fruit production are susceptible to CBS caused by *P. citricarpa,* except for sour orange (*C. aurantium*) and Tahiti lime (*C. latifolia*). The host status of pomelo (*C. maxima*) is still uncertain (see section 3.1.1.4). The *effectiveness* of the use of cultivars that are resistant or less susceptible to *P. citricarpa* would be **high** for pathway II (Tahiti lime fruit commercial trade without leaves and peduncles), but would be **low** overall due the fact that currently Tahiti lime and sour orange constitute only a small fraction of the total import of citrus fruit**.** This rating could be increased in the future if citrus varieties genetically modified for CBS resistance traits (Kava-Cordeiro et al., in press) became available. The *technical feasibility* is very high for Tahiti lime and sour orange but is **low** overall given the current lack of resistant or less susceptible varieties of sweet oranges, mandarins or lemons. The *uncertainty* for these ratings is **high**, owing to some publications indicating that sour orange and Tahiti lime can be colonised by *P. citricarpa* and that ascospores are produced in the leaf litter of Tahiti lime.

Figure 42: Citrus orchards for commercial fruit production under nets for protection from hailstorms.

4.1.1.10. Growing plants under exclusion conditions (glasshouse, screen, isolation).

Growing commercial citrus orchards for fruit production under exclusion could theoretically limit infection by reducing the introduction of external inoculum but may require screening with very fine mesh nets and controlled ventilation. Very early varieties are sometimes grown under nets to protect from hailstorms (Fig. 42). However, such conditions are applicable to plant propagation material (Fig. 43), but not to commercial citrus orchards on a large scale.

The *effectiveness* is likely to be **low**, since, due to the microscopic dimensions of *P. citricarpa* spores and because of the difficulty of securely excluding the pathogen in close proximity to outdoor grown citrus that may be infected, infection cannot be completely excluded. The *technical feasibility* is **negligible**, because of the difficulty of implementation in citrus orchards for fruit production over large areas. The *uncertainty* of these ratings is **medium,** due to the lack of data on the effectiveness of exclusion and the dispersal potential of the pathogen.

4.1.1.11. Harvesting of plants at a certain stage of maturity or during a specified time of year

Citrus fruit is susceptible to infection by *P. citricarpa* for several months after petal fall (Reis et al., 2003; Brentu et al., 2012). Following fruit infection, the latent period can last between 2 and 12 months (McOnie, 1967; Kellerman and Kotzé, 1977; Kotzé, 1981; Aguiar et al., 2012), depending on the citrus variety and growing conditions.

The *effectiveness* of harvesting citrus fruit during a specified time of the year is **negligible**, due to the long latent period between fruit infection and symptoms development.

The *technical feasibility* is **low**, because of the need to harvest citrus fruit at commercial maturity.

The *uncertainty* for these ratings is **low**.

4.1.1.12. Certification scheme

Citrus plants for planting, produced under a certification scheme, will initially be free from *P. citricarpa*. However, these plants can become infected when planted in a citrus orchard where *P. citricarpa* occurs. The prevalence of *P. citricarpa* in the orchard will then become dependent on the measures discussed in section 4.1.1.8. In areas where *P. citricarpa* occurs, the *effectiveness* of this option in reducing the probability of entry via the fruit pathway is likely to be **low**, the *technical feasibility* is **low**, and the *uncertainty* is **medium**, due to the lack of data on the local incidence of the pathogen in CBS-infested countries.

C. Options ensuring that the area, place or site of production at the place of origin remains free from the pest

4.1.1.13. Limiting the import of host plant material to material originating in pest-free areas

A pest-free area is defined as 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 (ISPM No.4; FAO, 1995). A pest-free area may be an entire country, a non-infested part of a country in which a limited infested area is present, or a non-infested part of a country situated within a generally infested area. Pest freedom of the area must be supported by general surveillance, delimiting surveys to demarcate the area and detection surveys to demonstrate the absence in the area and its buffer zone (for guidance on surveys and surveillance, see EFSA Panel on Plant Health (PLH), 2012). Phytosanitary measures must be in place to prevent the movement of potentially infested material into the area and to prevent natural spread of the pest into the area.

Surveys for CBS to demonstrate the pest-free status of a region within a CBS-infested country are not without their limitations, because of the likelihood of latently infected plants or *P. citricarpa* populations at low incidence being undetected in surveys. In the areas where the pathogen is currently distributed, CBS is usually first detected on lemons. Therefore, lemon trees should be the first to be inspected in an area for the detection of the pathogen. Based on the slow rate of spread and the frequent occurrence of latent infection, effective buffer zones are difficult to implement.

When the import of citrus fruit is restricted to material originating in pest free areas, the probability of introduction of *P. citricarpa* into the risk assessment area is reduced. The *effectiveness* is assessed as **high**, but this depends on the frequency and the confidence level of detection surveys to confirm the absence of *P. citricarpa* in the pest free area and on the intensity of phytosanitary measures to prevent entry of plant material (including fruit) infected by *P. citricarpa* into the pest free area. The design and frequency of surveys to confirm absence of *P. citricarpa* in the area should take into account the scattered presence of unmanaged citrus plants in private gardens, public areas or in uncultivated areas and the possible presence of latently infected plants in order to reach the required confidence level of the surveys.

The *technical feasibility* of the establishment and maintenance of a pest free area for *P. citricarpa* is **high** in countries where *P. citricarpa* is absent. The *feasibility* of establishment and maintenance of pest free areas in proximity to CBS infested areas is assessed as **moderate**, owing to the difficulties of detecting latent infection and low disease incidence in combination with the long lag phase observed between the first establishment and the development of CBS epidemics (see sections 3.1.1.2, 3.3.4 and 3.4.4). The *uncertainty* for these ratings is **low**.

4.1.1.14. Limiting import of host plant material to material originating in pest-free production places or sites of production

The designation and maintenance of pest free places or sites of production with respect to CBS is limited because of the presence of latent mycelium on infected citrus fruits and the difficulties in distinguishing CBS symptoms from those caused by other citrus pests. Also, as stated above (section 4.1.1.3), growing citrus orchards under exclusion conditions has **low** *feasibility*.

The *effectiveness* of this option is **low**. The *technical feasibility* is **low**, given the difficulties in mantaining the pest-free status of place and sites of production within CBS-infested countries due to latent fruit infections, the rarity of foliar symptoms and the postulated long lag phase between the first establishment and the development of the epidemics (see sections 3.1.1.2, 3.3.4. and 3.4.4).

The *uncertainty* is **medium**, due to the lack of detailed information on the incidence or absence of the pathogen at local level, as well as the lack of knowledge on the development of CBS epidemics at its inception in new sites.

4.1.1.15. Systems approaches integrating individual RROs

Systems approaches combining individual RROs may further reduce the probability of entry of *P. citricarpa* along this pathway. The following combinations are proposed:

For fruit originating from pest free areas or pest free production places, harvest and transport to packing stations should be done using clean boxes free of plant material. Packing should be in designated packing houses registered for packing of fruit from CBS-free areas and production places only. The *effectiveness* and *feasibility* is **high** with **low** *uncertainty*.

For fruit originating from CBS-affected areas, cultural measures and fungicide treatments to prevent *P. citricarpa* infections in the orchards should be combined with handling procedures and post-harvest treatments for fruit during packing to suppress the pathogen during handling and packing. Packing houses should keep a register of all processed fruit lots to allow tracking and tracing of infestations. Detection of latent infections in fruit prior to harvest by using ethephon dips and incubation will reduce the possibility of further symptom development during transport and storage. The *effectiveness* of each of these measures individually is assessed as **low**, except for the treatments in the orchard which has **moderate** *effectiveness* and etephon detection which has **high** *effectiveness*. The *effectiveness* of the integrated approach combining these measures together with appropriate official inspections is assessed as **high**. The *technical feasibility* is **high**, and the *uncertainty* is assessed as **medium**.

For citrus fruit imported in the EU from areas where *P. citricarpa* occurs, the end use could be restricted in combination with a restriction of its distribution within the EU. For example, citrus fruit might be imported into MSs without citrus production, only if this fruit is immediately processed in that MSs and waste disposal is conducted under a strict protocol to prevent the spread of *P. citricarpa.* The *effectiveness* is assessed as **high**. However, the *technical feasibility* is assessed as **low** due to the complex trade network of citrus fruit within the EU, the need to develop a traceability system to secure the intended end use (i.e. to keep distinct the citrus for processing from the citrus for fresh consumption) and for maintaining separate control systems for different citrus fruit pathways. *Uncertainty* is **medium** due to lack of information on the potential re-distribution of citrus fruit for processing as fresh fruit for consumption.

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Table 14: Summary of the risk reduction options identified and evaluated to reduce the entry along the citrus fruit commercial trade (pathways I, II and IV)

Category of options	Type of measure (for details, see EFSA Panel on Plant Health (PLH), 2012)	Position in the pathway	Existing measure	Effectiveness	Technical feasibility	Uncertainty
Options for consignments	Prohibition	Before shipment	No	Very high	Very high	Low
	Prohibition of parts of the host	Before shipment	Yes	High	Very high	Low
	Prohibition of specific genotypes of the host	Before shipment	No	Very high	Moderate	Medium
	Pest freedom of consignments; inspection or testing	Before shipment and/or at import	Yes	Moderate	Moderate	Medium
	Pre- or post-entry quarantine system: at harbours of exporting or importing country	Before shipment and/or at import	No		Not applicable	
	Pre- or post-entry quarantine system; in the country of origin at the orchard before harvest (induction of precocious symptoms expression in citrus fruit samples)	Before shipment and/or at import	No	High	High	Medium
	Preparation of consignment	Before shipment	No	Low	Very high	Medium
	Specified treatment of consignment	Before shipment	Yes	Low	Very high	Low
	Restriction on end use, distribution and periods of entry: period of entry	Before shipment and/or at import	No	Negligible	Very high	Low
	Restriction on end use, distribution and periods of entry: end use	After import	No	Negligible	Negligible	Medium
	Restriction on end use, distribution and periods of entry: distribution	After import	No	High	Negligible	Low
Options for the crop at the place of origin	Treatment of the crop, field or place of production	Before shipment	No	Moderate	Very high	Low
	Resistant or less susceptible varieties	Before shipment	No	Low	Low	High
	Growing plants under exclusion conditions	Before shipment	No	Low	Low	Medium

	Harvesting of plants during a certain period	Before shipment	No	Negligible	Low	Low
	Certification scheme	Before shipment	Yes	Low	Low	Medium
Options ensuring that the area, place or site of production at the place of origin, remains free from the pest	Limiting import of host plant material to material originating in pest-free areas	Before shipment	Yes	High	Moderate to high	Low
	Limiting import of host plant material to material originating in pest-free production places or pest-free production sites	Before shipment	Yes	Low	Low	Medium
Systems approaches	Pest free areas and production places combined with dedicated packing stations	Before shipment	No	Very high	High	Low
	Infested production places: measures in orchards combined with: handling procedures and treatments during packing; detection of latent infections in fruit prior to harvest by using ethephon dips and incubation; visual inspection and testing.	Before shipment	No	High	High	Medium
	Combined restriction on end use and distribution of imported citrus fruit	After import	No	High	Low	High

4.1.2. RROs to reduce entry along the citrus fruit import by passenger traffic (pathway III)

The RROs for this pathway are similar to those of the previous section. The results are summarised in Table 15.

A. Options for consignments

4.1.2.1. Prohibition

The prohibition of import of citrus fruit by passengers would prevent the entry of *P. citricarpa* into the EU along this pathway. Such a prohibition requires compliance by passengers, which can be influenced by the intensity and clarity of communication and by the frequency of passenger checks. The *effectiveness* is **high**, although it would depend on the level of compliance by passengers. Results from audits performed in Australia, where such a prohibition is implemented, show that fruit interceptions on passengers are made regularly, despite the communication campaigns. Moreover, there is a need for a high frequency of the passenger checks. The *technical feasibility* is therefore **low.** The *uncertainty* is **medium** due to the lack of EU data on the frequency of citrus fruit transport by passengers.

4.1.2.2. Prohibition of parts of the host or of specific genotypes of the host

Not applicable.

4.1.2.3. Phytosanitary certificates and other compliance measures

Not applicable.

4.1.2.4. Pest freedom of consignments: inspection or testing

The *effectiveness* of visual inspection of citrus fruit, carried by passengers, for symptoms of *P. citricarpa* is **moderate**, due to possible latent infections and confusion with symptoms by other injuries and pests. Testing is not applicable, since passengers would not be expected to await the results of the test. The *technical feasibility* of inspection of citrus fruit carried by passengers as an option to reduce the risk of entry of *P. citricarpa* is **low**. With an estimated 0.1% of passengers carrying on average one citrus fruit and thousands of passengers arriving daily in the EU, the frequency of passenger checks would have to be high. The *uncertainty* on these ratings is **low**.

4.1.2.5. Pre- or post-entry quarantine system.

Not applicable.

4.1.2.6. Preparation of the consignment

Not applicable.

4.1.2.7. Specified treatment of the consignment/reducing pest prevalence in the consignment.

Not applicable.

4.1.2.8. Restriction on end use, distribution and periods of entry

Not applicable.

B. Options preventing or reducing infestation in the crop at the place of origin

Such options are not applicable to citrus fruit carried by passengers.

C. Options ensuring that the area, place or site of production at the place of origin, remains free from the pest

Such options are not applicable to citrus fruit carried by passengers.

Table 15: Summary of applicable risk reduction options identified and evaluated for the pathway III: citrus fruit import by passenger traffic

Category of options	Type of measure (for details, see EFSA Panel on Plant Health (PLH), 2012)	Position in the pathway	Existing measure	Effectiveness	Technical feasibility	Uncertainty
Options for consignments	Prohibition	During customs checks	No	High	Low	Medium
	Visual inspection for pest freedom	During customs checks	No	Moderate	Low	Low

4.1.3. RROs to reduce entry along the commercial trade of citrus plants for planting, excluding seeds (pathway V and VI)

This section deals with the identification and evaluation of RROs to reduce the probability of entry of *P. citricarpa* along the two pathways of commercial trade of citrus plants for planting, excluding seeds, described in the entry section: the pathway (V) of commercial trade of citrus plants for planting and the pathway (VI) of commercial trade of Tahiti lime plants for planting (for a detailed description and analysis of these pathways please see sections 3.2.6 and 3.2.7, respectively). The plants for planting of Tahiti lime are dealt here together with the general pathway of citrus plants for planting, owing to the fact that Baldassarri et al. (2008) demonstrated that ascospores of *P. citricarpa* could also be produced on leaves of Tahiti lime (see section 3.1.1.4). There is therefore no difference in RROs along pathways V and VI.

Seeds are not included as they are not considered to be a potential entry pathway for *P. citricarpa* (see section 3.2.1).

The results of the evaluation are summarised in Table 16.

<u>A Options for consignments</u>

4.1.3.1. Prohibition

The *effectiveness* of prohibition is **very high**. The prohibition of imports of citrus plants for planting would be likely to prevent the introduction of the organism into the EU territory on citrus plant material for propagation purposes as well as on ornamental citrus plants for planting, particularly when these are latently infected.

The *technical feasibility* is **very high**, because it can be implemented in phytosanitary import procedures and customs operations and is already implemented in the EU (Council Directive 2000/29/EC, Annex III, point 16).

The *uncertainty* is assessed as **low**.

4.1.3.2. Prohibition of parts of the host or of specific genotypes of the host

- Prohibition of specific genotypes

As far as citrus species grown for propagating purposes (e.g. rootstocks) are concerned, sour orange (*C. aurantium*) was traditionally considered resistant to be CBS, but experimental studies would be needed to demonstrate whether it could still carry the pathogen if imported. The *effectiveness* of prohibiting plants for planting of all citrus species apart from sour orange is **high**, also considering that sour orange rootstocks are mostly propagated by seed (sour oranges for ornamental purpose can also be vegetatively propagated). The *technical feasibility* of limiting the prohibition of citrus propagating material imports to specific genotypes is however **low**, given the expertise required to distinguish between plants for planting of different citrus genotypes. The *uncertainty* is **high**, due to the fact that *P. citricarpa* has been isolated from asymptomatic leaves of sour orange in Brazil (Wickert et al., 2009), although *P. citricarpa* has not been observed to reproduce on this host.

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- Prohibition of parts of the host

Citrus vegetative plant propagation material always include leaves or buds which are likely to transport the pathogen if infected, so this option is not applicable.

4.1.3.3. Pest freedom of consignments: inspection or testing

Due to the potential presence of latent mycelium, the *effectiveness* is **low**. The *technical feasibility* is **low**. *P. citricarpa*-infected host plant material for propagating purposes can be asymptomatic and, thus, not detectable by visual inspection. The *uncertainty* is **low**, as there is a consensus in the scientific literature that plants for planting are the main pathway for long-distance dispersal of the pathogen.

4.1.3.4. Pre- or post-entry quarantine system.

P. citricarpa-infected citrus seedlings, scions and budwood are likely to remain asymptomatic and there is no validated method reported in the literature for accelerating CBS symptom development on living plants. Since latent infection of plants for planting may occur, post entry quarantine measures may be applied. Post-entry quarantine is applied for import of citrus nursery stock in EU MSs (Council Directive 2008/61/EC) and in other citrus producing countries (e.g. Biosecurity New Zealand, 2010; Vidalakis et al, 2010). For example, in New Zealand, the imported propagation material must be grown for a minimum period of 6 to 16 months in a post-entry quarantine facility where it will be inspected, treated and/or tested for regulated pests (Biosecurity New Zealand, 2010).

The effectiveness of pre- and post-entry quarantine systems depends on the level of containment established by the quarantine facilities, the quarantine period, and the methods and intensity of inspection and testing during the quarantine period. The *effectiveness* is **high** The *technical feasibility* is **high**, as this option is already implemented, but for limited import frequency of small consignments only. The *uncertainty* is **medium**, due to the lack of data on the specific effectiveness of such a scheme for *P. citricarpa*.

4.1.3.5. Preparation of the consignment

Culling of citrus planting material in the nursery is unlikely to detect CBS-infected plants, as young citrus seedlings/rootstocks/scions remain asymptomatic (see section 3.6.1.1).

The *effectiveness* is thus **negligible**, although the *technical feasibility* is **high**. The *uncertainty* is **low**.

4.1.3.6. Specified treatment of the consignment/reducing pest prevalence in the consignment.

Fungicide sprays applied to consignments of citrus planting material following their harvest, may reduce CBS incidence and severity but they cannot eliminate the pathogen.

The *effectiveness* is thus **low**, although the *technical feasibility* is **high**.

The *uncertainty* is **medium**, because there is no information on the use of fungicide sprays on citrus plant propagating material following harvest and before dispatch from the nursery .

4.1.3.7. Restriction on end use, distribution and periods of entry

Such restrictions are not applicable to citrus plants for planting: host plants of *P. citricarpa* may carry the pathogen year round, the end use is planting by definition, and the distribution is by definition to areas with host plants.

B. Options preventing or reducing infestation in the crop at the place of origin

4.1.3.8. Treatment of the crop, field or place of production in order to reduce pest prevalence.

Fungicide sprays in orchards may reduce CBS incidence and severity, but the pathogen is unlikely to be completely eliminated (see section 3.6.1.1). The *effectiveness* in reducing the probability of entry with plants for planting is thus **low**, although the *technical feasibility* is **high,** with **low** *uncertainty*.

4.1.3.9. Resistant or less susceptible varieties.

Given the lack of resistant cultivars, this option is not yet applicable.

4.1.3.10. Growing plants under exclusion conditions (glasshouse, screen, isolation).

To limit the introduction of inoculum, growing citrus plants for planting under exclusion conditions may require screening with very fine mesh nets and controlled ventilation to block airborne spores.

The *effectiveness* is likely to be **low**, as, due to the microscopic dimensions of the spores of *P. citricarpa* and because of the difficulty of securely excluding the pathogen in close proximity to outdoor grown citrus that may be infected, infection cannot be completely excluded.

The *technical feasibility* is **high,** because it is regularly applied in nurseries against vectors of plant diseases (see Fig. 43). The *uncertainty* is **high,** due to the lack of data on the effectiveness of exclusion and the dispersal potential of the pathogen.

Figure 43: Citrus plant propagation material grown under nets

4.1.3.11. Harvesting of plants at a certain stage of maturity or during a specified time of year.

Given the year-round infectiousness and susceptibility of host plants, this option is not applicable.

4.1.3.12. Certification scheme of plant propagation material.

For plants for planting of citrus, certification schemes have been developed worldwide (e.g, see: Von Broembsen and Lee, 1988; Passos et al., 2010; Vidalakis et al., 2010: Australian Citrus Propagation Association Inc., undated). Citrus plants for planting produced according to such a scheme are however unlikely to be completely free from *P. citricarpa*, unless they are produced in a pest free area. The *effectiveness* of this risk reduction option is **low,** unless this option is combined with pest free area (then the *effectiveness* is **high**) .The *technical feasibility* is **very high**.The *uncertainty* is **low**.

C. Options ensuring that the area, place or site of production, remains free from the pest

4.1.3.13. Limiting import of host plant material to material originating in pest-free areas

This is a viable risk reduction option, but the long period of latent infection can reduce the *feasibilty* of this option when pest free areas are in proximity to CBS infested areas. The *effectiveness* is **high**. Due to the difficulties of detecting latent infection and low CBS incidence, the *technical feasibility* for maintenance of pest free areas in proximity to CBS infested areas is **moderate**, whereas it is **high** in CBS-free countries,

The *uncertainty* is **medium,** due to the difficulties in detecting latent infections and to lack of studies on the maximum distance of ascospore dispersal.

4.1.3.14. Limiting import of host plant material to material originating in pest-free production places or pest-free production sites

The *effectiveness* of establishing pest-free production places/ sites for plants for planting is **low**, due to the spread potential of the disease (see section 4.1.1.7). The *technical feasibility* is **moderate**. The *uncertainty* is **medium**, due to the lack of knowledge on long-distance dispersal of *P. citricarpa* spores.

4.1.3.15. Systems approaches integrating individual RROs.

A possible systems approach for the production of plants for planting is the application of a certification scheme in citrus nurseries in pest free areas, including regular testing for *P, citricarpa* at different production stages, and the preparation and sealing of consignments at the nursery. The *effectiveness* of this approach is assessed as **high**, with **very high** *technical feasibility* and **low** *uncertainty*.

Table 16: Summary of the risk reduction options identified and evaluated for the commercial trade of citrus plants for planting (pathways V and VI)

Category of options	Type of measure	Position in the pathway	Existing measure	Effectiveness	Technical feasibility	Uncertainty
Options for consignments	Prohibition	Before shipment	Yes	Very high	Very high	Low
	Prohibition of specific genotypes	Before shipment	No	High	Low	High
	Prohibition of parts of the host	Before shipment	No	Not applicable		
	Visual inspection / testing for pest freedom	Before shipment and/or at import	No	Low	Low	Low
	Pre- or post-entry quarantine systems	Before / After shipment	No	High	High	Medium
	Preparation of consignment	Before shipment	No	Negligible	High	Low
	Specified treatment of consignment	Before shipment	No	Low	High	Medium
	Restriction on end use. distribution and period of entry	After shipment	No	Not applicable		
Options for the crop at the place of origin	Treatment of the crop, field or place of production	Before shipment	Yes	Low	High	Low

	Growing plants under exclusion conditions (glasshouse, screen, isolation)	Before shipment	No	Low	High	High
	Harvesting of plants during a specific time of the year	Before shipment	No	Not applicable		
	Certification scheme	Before shipment	No	Low	Very high	Low
Options ensuring that the area, place or site of production at the place of origin, remains free from the pest	Limiting import of host plant material to material originating in pest-free areas	Before shipment	No	High	Moderate to high	Medium
	Limiting import of host plant material to material originating in pest-free production places or pest-free production sites	Before shipment	No	Low	Moderate	Medium

Summary of applicable risk reduction options identified and evaluated for the pathway of citrus plants for planting by passenger traffic (pathway VII)

Category of options	Type of measure (for details, see EFSA Panel on Plant Health (PLH), 2012a)	Position in the pathway	Existing measure	Effectiveness	Technical feasibility	Uncertainty
Options for consignments	Prohibition	During customs checks	No	Moderate	Low	High
	Visual inspection for pest freedom	During customs checks	No	Low	Negligible	Medium

Summary of the risk reduction options identified and evaluated for the commercial trade of citrus leaves (pathways VIII)

Category of options	Type of measure	Position in the pathway	Existing measure	Effectiveness	Technical feasibility	Uncertainty
Options for consignments	Prohibition	Before shipment	Yes	Very high	Low	Low
	Prohibition of parts of the host	Before shipment	No	Not applicable		
	Prohibition of specific genotypes	Before shipment	No	Not applicable		
	Visual inspection / testing for pest freedom	Before shipment and/or at import	No	Low	Negligible	Medium
	Pre- or post-entry quarantine systems	Before / After shipment	No	Not applicable		
	Preparation of consignment	Before shipment	No	Low	High	Low
	Specified treatment of consignment	Before shipment	No	No info available, not evaluated		
	Restriction on end use. distribution and period of entry	After shipment	No	Restriction on period not applicable. Restriction in distribution to EU MSs where citrus is not grown: effective but feasibility negligible. Uncertainty high.		
Options for the crop at the place of origin	Treatment of the crop, field or place of production	Before shipment	Yes	Moderate	High	Medium

	Resistant or less susceptible varities	Before shipment	No	Not applicable		
	Growing plants under exclusion conditions (glasshouse, screen, isolation)	Before shipment	No	Low	High	High
	Harvesting of plants during a specific time of the year	Before shipment	No	Not applicable		
	Certification scheme	Before shipment	No	Low	High	Low
Options ensuring that the area, place or site of production at the place of origin, remains free from the pest	Limiting import of host plant material to material originating in pest-free areas	Before shipment	No	High	High	Medium
	Limiting import of host plant material to material originating in pest-free production places or pest-free production sites	Before shipment	No	Low	Moderate to high	High
Systems approaches	Certification scheme + Pest Free Area + preparation and sealing of consignment on nursery	Before shipment	No	High	Very high	Low

4.1.4. RROs to reduce entry along the pathway of import of citrus plants for planting by passenger traffic (pathway VII)

The risk reduction options for this pathway are similar to those of the general citrus plants for planting pathway, although there is very little information on the frequency of transport of citrus plants for planting along this pathway. The results of the evaluation are summarised in Table 17.

A. Options for consignments

4.1.4.1. Prohibition

The prohibition of import of citrus plants for planting for citrus fruit production by passenger traffic would prevent the entry of *P. citricarpa* into the EU along this pathway. Such a prohibition requires compliance by passengers which can be influenced by the intensity and clarity of communication of this measure to passengers and the intensity of passenger checks. Results of audits performed in Australia for citrus fruit show that interceptions on passengers are made regularly, despite communication and inspection. There are no specific data on interception of citrus plants for planting for citrus fruit production carried by passengers, but the frequency of passengers carrying such material is assumed to be lower than the frequency of passengers with fruit for consumption. The *effectiveneness* is assessed as **moderate**, although it would depend on the level of compliance by passengers. The *technical feasibility* is **low**, because of the need to implement it on a very large volume of passenger luggage at all entry points over the whole year and because it would require the technical ability to identify citrus plants for planting at the border (e.g. citrus plants, rootstocks or buds). The *uncertainty* of these ratings is **high**, due to the lack of data on the frequency of transport of citrus plants for planting along this pathway and on the compliance by passengers.

4.1.4.2. Prohibition of parts of the host or of specific genotypes of the host

Not applicable.

4.1.4.3. Pest freedom of consignments: inspection or testing

The *effectiveness* of visual inspection of citrus plants for planting carried by passengers for symptoms of *P. citricarpa* is **low**, mainly due to the possible presence of latent infections. The leaves of young citrus plants infected by *P. citricarpa* are usually asymptomatic and, thus, not detectable by visual inspection. Sample testing could be applicable, however the plants for planting should be stored until the results of the test are available before further customs procedures. Therefore, the *technical feasibility* of inspection of citrus plants carried by passengers as an option to reduce the risk of entry of *P. citricarpa* is **negligible**. The fraction of passengers carrying such planting material is likely to be much lower than the estimated 0.1% of passengers carrying on average one citrus fruit (see section 4.1.2.4), and therefore a very large number of passenger luggages would need to be inspected to detect citrus plants for planting. The *uncertainty* on these ratings is **medium**, due to the lack of data on the frequency and origin of transport of citrus plants for planting along this pathway.

4.1.4.4. Pre- or post-entry quarantine system.

Not applicable.

4.1.4.5. Preparation of the consignment

Not applicable.

4.1.4.6. Specified treatment of the consignment/reducing pest prevalence in the consignment.

Not applicable.

4.1.4.7. Restriction on end use, distribution and periods of entry

Not applicable.

B. Options preventing or reducing infestation in the crop at the place of origin

Such options are not applicable to plants for planting carried by passengers.

C. Options ensuring that the area, place or site of production at the place of origin, remains free from the pest

Such options are not applicable to plants for planting carried by passengers.

4.1.5. RROs to reduce entry along the pathway of import of citrus leaves (pathway VIII)

The results of the evaluation of RROs for this pathway are summarised in Table 18.

A. Options for consignments

4.1.5.1. Prohibition

The prohibition of import of citrus leaves commercial trade would prevent the entry of *P. citricarpa* into the EU along this pathway. The *effectiveness* is assessed as **very high**. The *technical feasibility* is **low,** because citrus leaves could be sent in non-declared packages escaping customs operations and phytosanitary procedures. The *uncertainty* on these ratings is assessed as **low**.

4.1.5.2. Prohibition of parts of the host

Not applicable to citrus leaves commercial trade.

4.1.5.3. Prohibition of specific genotypes

Not applicable to citrus leaves, commercial trade. The host status of *C. hystrix* and other exotic citrus species for *P. citricarpa* is highly uncertain and there is no information on the use for cooking purposes of other citrus species apparently not affected by CBS, such as sour orange or Tahiti lime. In addition, it was reported that *P. citricarpa* can colonize leaves of both citrus species and, in the case of Tahiti lime, even reproduce on them (see section 3.1.1.4).

4.1.5.4. Pest freedom of consignments: inspection or testing

The detection of *P. citricarpa* in consignments is based on visual inspection, sampling and laboratory testing. Inspection or testing of consignments may be applied at the time of export and/or at the time of import. At export, inspection or testing may serve as a stand-alone measure, without other official measures for production, harvest and packaging, or as a measure to verify that other measures have been effective. At import, inspection generally serves to verify phytosanitary measures by the exporting country.

The *effectiveness* of visual inspection of citrus leaves for symptoms of *P. citricarpa* is **low**, mainly due to the possible presence of latent infections. The leaves of citrus infected by *P. citricarpa* are usually asymptomatic and, thus, not detectable by visual inspection. Sample testing could be applicable, but without a reliable detection of symptoms on leaves, very large sample sizes would be required. Therefore, the *technical feasibility* of inspection of citrus leaves as an option to reduce the risk of entry of *P. citricarpa* is **negligible**. The *uncertainty* on these ratings is **medium**, due to lack of data on the amounts, origin and end use of citrus leaves along this pathway, as well as due to the unknown host status of *C. hystrix* and other exotic citrus species for *P. citricarpa*.

4.1.5.5. Pre- or post-entry quarantine system.

Not applicable to citrus leaves.

4.1.5.6. Preparation of the consignment

The preparation of the consignment includes several stages, including handling and transport of harvested leaves and packing prior to export. Culling and cleaning of leaves may theoretically allow the removal of leaves infected by *P. citricarpa*, but leaves with latent infections or with small lesions will not be detected and eliminated by these procedures. The *effectiveness* is assessed as **low**. The *technical feasibility* is assessed as **high**. The *uncertainty* on these ratings is **low**.

4.1.5.7. Specified treatment of the consignment/reducing pest prevalence in the consignment.

No information is available on treatments against *P. citricarpa* on citrus leaves for flavouring or cooking. This RRO has therefore not been evaluated.

4.1.5.8. Restriction on end use, distribution and periods of entry

It is not possible to identify periods of the year when citrus leaves are not infected, nor periods of the year when host plants are not susceptible to infection. Therefore a restriction on the period of entry of citrus leaves is not applicable.

A restriction on the distribution of the imported citrus leaves for flavouring to the EU MSs where citrus is not grown could be potentially effective, however the entry through this pathway is considered unlikely. The *feasibility* is **negligible** because of the free internal market of the EU. *Uncertainty* is **high** due the lack of information and data on the trade of citrus leaves for flavouring,

B. Options preventing or reducing infestation in the crop at the place of origin

4.1.5.9. Treatment of the crop, field or place of production in order to reduce pest prevalence

Treatments of citrus plants against *P. citricarpa* to reduce the incidence of CBS may be routinely applied by citrus producers in the absence of official phytosanitary requirements, although the combination of fungicide treatments, cultural and other methods may vary among producers. However, these measures will not eliminate *P. citricarpa* in production places and the harvest of infected leaves cannot be prevented due to the scarcity of CBS leaf symptoms. The incidence of *P. citricarpa* in harvested leaves remains variable, depending on the intensity of the control programs and the weather conditions during the growing season.The *effectiveness* of control program is assessed as **moderate**. The *technical feasibility* is assessed as **high**. The *uncertainty* on these ratings is **medium**.

4.1.5.10. Resistant or less susceptible varieties

The host status of *C. hystrix* and other exotic citrus species for *P. citricarpa* is highly uncertain and there is no information on the use for cooking purposes of other citrus species apparently not affected by CBS, such as sour orange or Tahiti lime. In addition, it was reported that *P. citricarpa* can colonize leaves of both citrus species and, in the case of Tahiti lime, even reproduce on them (see section 3.1.1.4). This RRO is not applicable to citrus leaves.

4.1.5.11. Growing plants under exclusion conditions (glasshouse, screen, isolation)

This RRO may be applicable to production places producing citrus leaves, if the plants are kept sufficiently small so that they can grow in areas screened with very fine mesh nets and controlled ventilation to block airborne fungal spores.. The *effectiveness* is likely to be **low** due to the microscopic dimensions of the spores of *P. citricarpa* and because of the difficulty of securely excluding the pathogen in close proximity to outdoor grown citrus that may be infected, infection cannot be completely excluded. The *technical feasibility* is **moderate,** as it is regularly applied in nurseries against vectors of plant diseases and could be used also for small citrus trees (Fig. 41). The *uncertainty* is **high,** due to the lack of data on the effectiveness of exclusion and the dispersal potential of the pathogen.

4.1.5.12. Harvesting of plants at a certain stage of maturity or during a specified time of year.

Not applicable since citrus leaves are susceptible to *P. citricarpa* for 8-10 months and new leaves are produced year-round (see section 3.3.1.1).

4.1.5.13. Certification scheme

Plants for production of citrus leaves, produced under a certification scheme, will initially be free from *P. citricarpa*. However, these plants can become infected when planted in areas where *P. citricarpa* occurs. The *effectiveness* of a certification scheme is **low.** The technical feasibility is assessed as **high**. The *uncertainty* on these ratings is assessed as **low**.

C. Options ensuring that the area, place or site of production at the place of origin, remains free from the pest

4.1.5.14. Limiting import of host plant material to material originating in pest-free areas

The different aspects of this RRO are discussed in section 4.1.1.14.

The *effectiveness* of pest-free areas is assessed as **high**, on the condition that procedures for maintaining the pest free area and its buffer zone are documented, regularly officially evaluated, and the results reported.

The establishment and maintenance of a pest free area for *P. citricarpa* is technically feasible, but surveys with adequate attention to the distribution of managed and unmanaged host plants in the pest free area should be performed when designating the pest free area and its buffer zone. The *technical feasibility* is assessed as **high**. The *uncertainty* is **medium**, due to the difficulties in detecting latent infections and to the lack of studies on the maximum distance of airborne ascospore dispersal.

4.1.5.15. Limiting import of host plant material to material originating in pest-free production places or pest-free production sites

The *effectiveness* of this measure to establish CBS-free production sites for production of citrus leaves is assessed as **low**, but depends on the intensity of monitoring. Due to latent infections, the *technical feasibility* is **moderate to high.** The *uncertainty* is **high**.

4.2. Systematic identification and evaluation of options to reduce the probability of establishment and spread

This section analyses the RROs that can be applied in the EU to prevent the establishment and spread of *P. citricarpa*. However some of the RROs to reduce the probability of transfer to a suitable host in the entry pathways are the same as those that can reduce spread and are therefore also included in this section. The results are summarised in table 19.

4.2.1. Pruning

The trade in citrus fruit has been considered as a pathway for both the entry and spread of *P. citricarpa*. In both steps the transfer of *P. citricarpa* to a citrus plant depends on the splash dispersal of pycnidiospores from culled fruit, waste or peel. The transfer may be favoured by low-hanging citrus branches in orchards, private gardens, roadsides and parks; therefore pruning the lower branches of citrus trees could theoretically reduce the probability of transfer. However, a requirement for pruning the low branches of citrus trees in parks, roadsides and private gardens is difficult to implement. This measure is not feasible in commercial orchards, because low hanging branches are the most productive and they are more easily harvested, so citrus trees in the EU are trained and pruned to maximize this part of the canopy (see section 3.3.3.3). This option is considered as having a **low** *effectiveness* and a **negligible** *technical feasibility*, with **low** *uncertainty*.

4.2.2. Irrigation and other cropping practices

There is a trend to move away from the irrigation systems that use large amounts of water (see sections 3.3.3.1 and 3.3.3.2). A wider use of drip irrigation can reduce the risk of establishment as this method does not wet citrus leaf surfaces. Instead, micro-sprinkler irrigation uses spray jets under the tree canopy that not only wet the soil but also the leaves in the lower canopy of the tree, thereby significantly increasing leaf wetness and aiding *P. citricarpa* establishment. In addition, *P. citricarpa* pseudotecia production in leaf litter is favored by alternating leaf wetting and leaf dryness. When microsprinkler irrigation is applied, apparently the number of dead leaves with pseudothecia in the orchard floor is much higher (10 x) than with drip or flood irrigation (Alcoba et al., 1999, cited in Feichtenberger E, Citrus black spot and its management in Brazil, ppt at Packinghouse Day & The Indian River Postharvest Workshop). However, these data should be interpreted toghether with leaf litter decomposition rates, which may differ depending on the irrigation system used. Cover crops and mulching of the orchard floor with grass cuttings after the leaf drop can accelerate the decomposition of the citrus leaves bearing the perithecia, limiting ascospore dispersal and thus reducing the inoculum. In addition, since citrus trees in poor conditions are more susceptible to CBS, it is important to maintain tree vigour (Schutte, 2009). Therefore, the *effectiveness* of the application of drip and flood irrigation to reduce the probability of CBS establishment is assessed as **moderate**, with **high** *technical feasibility* and **low** *uncertainty*.

4.2.3. Hygiene measures: waste management

However, adopting hygiene practices specific to CBS in citrus waste management in citrus packing houses and citrus processing plants is likely to be more effective and easier to implement than cultural practices in orchards, gardens, roadsides and parks. The implementation of strict containment and waste processing measures (according to the guidelines for handling of such biowaste in EPPO Standard PM 3/66(2)) at citrus packing houses or processing industries handling citrus fruit imported from areas where CBS occurs, would reduce the probability of transfer to a suitable host, and thus establishment and spread. However, large amounts of culled fruit and waste are produced by citrus packing houses and processing plants located in citrus producing areas of the EU (Section 3.2.2.4) and high safety standards would have to be set for these facilities. Moreover, a considerable proportion of citrus fruit imported in the EU is destined for direct consumption via various markets ranging from supermarkets to small outdoor markets, where standards for waste management cannot be controlled other than by making consumers aware of the phytosanitary risk. Therefore, the effectiveness and feasibility are limited by the scattered distribution of the numerous points of potential transfer in the citrus growing EU MSs. The *effectiveness* of such measure is assessed as **high**, however with **low** *technical feasibility* due to the need for the application of specific measures for strict citrus waste management in all the citrus growing EU MSs. *Uncertainty* is **high**, particularly on the feasibility of the practical implementation and on the lack of studies on survival of *P. citricarpa* in citrus waste and in the compost derived from citrus waste.

4.2.4. Eradication

Following the discovery of an outbreak of *P. citricarpa*, eradication measures should be implemented immediately. An eradication programme includes surveys to determine the limits of the outbreak, eradication actions to eliminate a pest from an area, containment action to prevent pest spread and surveys to verify absence of that pest (ISPM 9 by FAO, 1998), and the eradication measures themselves. CBS has never disappeared or declined after the epidemic stage has been reached, and successful disease eradication has never been achieved. Field surveillance for CBS eradication is challenging, considering that *P. citricarpa* may be present as latent mycelia in asymptomatic citrus fruit and leaves and there is a long lag phase between the first establishment and subsequent epidemic development (section 3.4.4).

The *effectiveness* of CBS eradication is **low**, because there are no reports of successful eradication of CBS: once established, the disease is reported to expand slowly but relentlessly. The *technical feasibility* is **low**, and the *uncertainty* on these ratings is **low**.

4.2.5. Containment

Once the disease has established, RROs to reduce the spread of *P. citricarpa*, include: containment measures in infested areas (e.g. cultural/fungicide control measures in orchards); preventive measures in areas suitable for new infection foci (e.g. the adoption of drip instead of sprinkler irrigation and the avoidance of citrus mono-cultures; Bellotte et al., 2013); targeted surveys at high-risk nodes in the trade network of fruit and plants for planting; information campaigns for the local growers, the stakeholders and the public so as to raise awareness of the disease and increase the likelihood of implementation of containment and preventive measures.

The *effectiveness* of containment is assessed to be **low**, because there is little evidence from other regions that CBS can be successfully contained. Once established, the disease is reported to expand slowly but relentlessly. The *technical feasibility* is **moderate**, and the *uncertainty* on these ratings is **low**.

4.2.6. Surveillance

A surveillance program including regular detection surveys in commercial citrus orchards, abandoned citrus orchards and public areas, in areas with production of citrus fruit and/or plants for planting would contribute to eradication and containment. The *effectiveness* is determined by the intensity of the surveys including sampling, visual inspection and laboratory testing, however it is assessed as **moderate** due to latent asymptomatic infections and to the reported long lag phase between first introduction and development of the epidemic. The *technical feasibility* is **moderate**, due to the difficulty to organize surveys in public areas, and the *uncertainty* is **medium**.

Table 17: Summary of risk reduction options identified and evaluated to reduce the probability of establishment and spread

Type of measure (for details, see EFSA Panel on Plant Health (PLH), 2012a)	Existing measure	Effectiveness	Technical feasibility	Uncertainty
Pruning	No	Low	Negligible	Low
Irrigation and other cropping practices	No	Moderate	High	Low
Hygiene measures: waste management	No	High	Low	High
Eradication	No	Low	Low	Low
Containment	No	Low	Moderate	Low
Surveillance	No	Moderate	Moderate	Medium

4.3. Systems approach of Risk Reduction Options

With the exception of prohibition and of limiting the import from pest-free areas, the effectiveness of the risk reduction options evaluated is generally low (see summary Tables of RROs 13-19). Combining ineffective risk reduction options may slightly increase their overall effectiveness, but is unlikely to result in a significant risk reduction in the case of *P. citricarpa.*

The only risk reduction options with high effectiveness were found to be:

- Prohibition; prohibition of parts of the host; prohibition of specific genotypes of the host
- citrus fruit consignment testing using the method to induce precocious symptom expression (see section 4.1.1.4) together with validated molecular methods, which should thus be further developed and adopted in conjunction with the other options,

- and pest free areas

The *effectiveness* of a systems approach to RROs is assessed as **moderate**, with a **moderate** *technical feasibility* and **high** *uncertainty*.

4.4. Evaluation of the current phytosanitary measures to prevent the introduction and spread of *P. citricarpa*

The current phytosanitary measures in place in the EU are designed to prevent the introduction into and spread within the EU. These phytosanitary measures are listed in section 3.1.3 and evaluated below.

"Guignardia citricarpa Kiely all strains pathogenic to Citrus" is listed in the Annex II Part A Section I and Annex IV Part A Section I (see section 3.1.3.3) of the EU Plant Health Directive (Dierctive 2000/29/EC). However, following recent taxonomic and nomenclatural changes, the correct name for the causal agent of CBS is now *Phyllostica citricarpa* (McAlpine) Aa and *Guignardia citricarpa* Kiely is considered to be a synonym. While new knowledge on the *Phyllosticta* species associated with citrus is continuously emerging, the current knowledge supports the conclusion that only *P. citricarpa* has proven to be pathogenic to citrus and a threat to citrus cultivation in regions that are suitable for this pathogen (see section 3.1.1.1), therefore the specification "all strains pathogenic to Citrus" is not needed when using the taxonomically updated name *Phyllosticta citricarpa* (McAlpine) Aa.

The combination of the requirements listed in Directive 2000/29/EC for all citrus pathways could be considered as being highly effective in preventing the introduction of *P. citricarpa* into the EU because there have been no outbreaks of CBS. However, it has also been argued that successful introductions have not taken place due to the unsuitable climate for *P. citricarpa* in the risk assessment area despite very large shipments of citrus into Europe over many decades from areas where the disease is present (Kotzé, 2000). In this respect, it it is important to note that, until 1993, when the EC phytosanitary directive established common quarantine requirements for import of citrus fruit to prevent the entry of *P. citricarpa* into the EU, most citrus growing EU countries had a very strict national quarantine for citrus, with a general prohibition of citrus fruit import (see section 3.1.3.1). For this reason the trade in citrus fruit from CBS areas in the world to EU citrus growing areas has been very limited, with the only exception of Italy where the import of grapefruit was allowed (although still in limited quantities) from countries where CBS is present (see section 3.1.3.2). Responding to the requirements of the EU directive, imports of citrus fruit were first allowed in 1993 and 1999 for Spain and Italy respectively.

The current phytosanitary measures are designed to prevent the entry of *P. citricarpa* into the EU. The effectiveness and technical feasibility of risk reduction options in preventing or reducing entry has already been evaluated in section 4.1. The Panel has therefore taken the conclusions from section 4.1 in determining the effectiveness of the EU's phytosanitary measures in preventing or reducing *P. citricarpa* entry along each of the pathways identified in section 3.2. Pathways that are prohibited under regulations in Annex III are evaluated together.

4.4.1. General remarks

"Guignardia citricarpa Kiely all strains pathogenic to Citrus" is listed in Annex II Part A Section I (c) of Directive 2000/29/EC as a harmful organism whose introduction into and spread within all EU MSs should be banned if present on plants of *Citrus* L., *Fortunella* Swingle, *Poncirus* Raf. and their hybrids, other than seeds.

As stated above, the correct name of the organism should be *Phyllosticta citricarpa* (McAlpine) Aa.

The subject of contamination is indicated as “plants,,,,, other than seeds”. The term “plants” as described in art, 2 1 (a) of Directive 2000/29/EC includes the following items of relevance for citriculture: living plants, fruit, cut flowers, branches with foliage, cut trees retaining foliage, plant tissue culture. Of these items, living plants, fruit and any plant part bearing leaves are entry pathways for *P. citricarpa*. Flowers do not harbour *P. citricarpa* unless attached to a branch with foliage. Seeds are correctly excluded as they are not considered an entry pathway for this pathogen.
With regard to the botanical genera, generally all *Citrus* species are susceptible to *P. citricarpa*. The only exceptions are:

- *Citrus latifolia* Tanaka (Tahiti lime) can be colonised by *P. citricarpa*. It does not produce symptoms/pycnidia on fruit but it produces ascospores and therefore entry is unlikely with fruit (with high uncertainty) but is more likely with plants and plant parts with leaves.
- *Citrus aurantrium* L. (sour orange) is considered resistant although sometimes *P. citricarpa* has been isolated from asymptomatic sour orange.

With regard to *Fortunella* Swingle (kumquat), this species is recorded by Kiely (1948) as moderately susceptible to CBS under conditions of natural infection.

No definitive information has been found on the susceptibility of *Poncirus* Raf. (trifoliate orange) to CBS.

4.4.2. Remarks concerning the pathway ‘citrus fruit by commercial trade’

The entry of fruit of *Citrus*, *Fortunella*, *Poncirus* and their hybrids with *P. citricarpa* is banned under Annex IIAI of EU Plant Health Directive (2000/29/EC). In order for Third Countries to export such fruit to the EU, in brief, Annex IVAI states that fruits must not only be free from peduncles and leaves and the packaging shall bear an appropriate origin mark but also that the fruit must be accompanied by an official statement to confirm that they originate from either:

a) a country that is free from the pest

b) a pest free area

c) a pest free place of production (no symptoms observed in the field of production and in its immediate vicinity since the beginning of the last cycle of vegetation, and none of the fruits harvested in the field of production has shown, in appropriate official examination, symptoms of this organism)

d) a field of production subjected to appropriate treatments against *Guignardia citricarpa* Kiely (all strains pathogenic to *Citrus*), and none of the fruits harvested in the field of production has shown in appropriate official examination, symptoms of this organism.

Options (a) and (b) have already been evaluated in section 4.1.1.13. This section concluded that the establishment and maintenance of pest free areas for *P. citricarpa* has high effectiveness and moderate to high technical feasibility with medium uncertainty but such pest free areas need to be based on surveys with adequate attention to the distribution of managed and unmanaged host plants in the pest free area. The uncertainty was rated is low. Examples of country freedom and pest free areas for *P. citricarpa* include New Zealand and the Hartswater and Warrenton magisterial districts of Northern Province, South Africa, respectively (Commission Decision 2006/473/EC (OJ L 187, 8.7.2006, p. 35). Cartsens et al (2012) have recently justified area freedom for Western Cape Province, South Africa.

Option (c) has already been partly evaluated in section 4.1.1.4 and assessed as having a low effectiveness, a low technical feasibility and medium uncertainty. Option (c) includes also an inspection that is separately evaluated in section 4.1.1.3. This inspection in Brazil can be connected with the ethephon pre-export test to induce precocious symptoms expression (see section 4.1.1.4).

For option (d), appropriate treatments (section 4.1.1.8) are assessed as having a moderate effectiveness but with a very high technical feasibility and low uncertainty. For the second part of option (d), section 4.1.1.4 covers the "pest freedom of consignments: inspection or testing": the visual inspection of consignments combined with laboratory testing is assessed as moderate with moderate technical feasibility and medium uncertainty.

Between 1999 and 2012, *P. citricarpa* was detected in 961 consignments by 11 EU MS indicating that exporting countries (see section 3.2.2 and figure 8) have difficulties in implementing the special requirements of the EU concerning *P. citricarpa*. For these 11 EU MS, there is a good correlation between the amount of citrus fruit imported and the number of *P. citricarpa* inspections. However, it is not possible from the data available in the Europhyt database to analyse which of the options listed in Annex IVAI has been applied to the consignments within which interceptions were made.

The analysis of the risk reduction options in the EC Plant Health Directive utilised to prevent the entry of *P. citricarpa* on fruit indicates that only country and area freedom are highly effective. However from section 4.1.1.15 a system approach is also shown to have high effectiveness.

4.4.3. Remarks concerning the pathway 'lime fruit (Citrus latifolia) by commercial trade'

Although no interceptions on Tahiti lime have been made in EU border inspections, *P. citricarpa* has been shown to colonize Tahiti lime fruit under natural conditions, however without expressing symptoms in fruit. This implies a high probability of association of the pathogen with the pathway of Tahiti lime fruit without leaves and peduncles at origin as latent mycelia in asymptomatic fruits but a very unlikely probability of transfer to suitable host. The overall probability of entry with the pathway of Tahiti lime fruit, without leaves and peduncles, is rated as very unlikely, with high uncertainty.

4.4.4. Remarks concerning the pathway 'citrus fruit with passenger traffic'

Currently, under EU legislation, measures to prevent the entry of *P. citricarpa* via citrus fruit may not be applied to citrus fruit carried by passengers since the special requirements for plants, plant products and other objects listed in Annex IV, Part A and in Annex V B need not apply for small quantities of plants, plant products, foodstuffs or animal feedingstuffs where they are intended for use by the owner or recipient for non-industrial and non-commercial purposes or for consumption during transport, provided that there is no risk of harmful organisms spreading (Council Directive 2000/29/EC, Art. 5 paragraph 4; Art 13b paragraph 3). According to the risk assessment (section 3.2.4.) the movement of *P. citricarpa* on fruit carried by passengers is very likely, but the transfer to a suitable host is unlikely, although with high uncertainty. The frequency of passengers carrying citrus fruit was estimated as 0.1 % (Section 4.1.2.4) and a large sample of passengers would need to be inspected to reduce the rate of entry of citrus fruit by passengers. A combination of improved communication measures to inform incoming passengers of their obligations with incidental targeted inspection of passengers might be more effective.

4.4.5. Remarks concerning pathway 'citrus fruit with leaves by commercial trade'

Although importation from Third Countries of citrus fruit with leaves is not permitted by EU legislation, a number of interceptions have been made by EU MS of consignments with citrus fruit with leaves originating from Third Countries during recent years (see section 3.2.5).

According to Annex IV part A Section I item 16.1, fruits of *Citrus* L., *Fortunella* Swingle, *Poncirus* Raf. and their hybrids, originating in Third Countries, shall be free from peduncles and leaves and the packaging shall bear an appropriate origin mark. This measure has been evaluated in section 4.1.1.2 as having high effectiveness and very high technical feasibility, with low uncertainty. It has to be noted that the pathway of citrus fruit with leaves and peduncles has a likely rating for entry due to the likely transfer to suitable host by ascospores produced on leaf litter. This also applies to Tahiti lime fruit with leaves and peduncles, since *P. citricarpa* ascospores have been reported from Tahiti lime leaf litter.

4.4.6. Remarks concerning pathways 'citrus plants for planting for citrus fruit production, commercial trade', 'lime (*Citrus latifolia*) plants for planting' and 'citrus leaves'

Since the import of citrus plants into the EU is prohibited, no trade data are available on the volume of citrus plant propagation material from countries of current distribution of *P. citricarpa* to the EU.

According to Annex III part A item 16 of Directive 2000/29/EC, the introduction into the EU of plants of *Citrus* L., *Fortunella* Swingle, *Poncirus* Raf. and their hybrids, other than fruit and seeds, originating in Third Countries, is prohibited. This measure has been evaluated in section 4.1.3.1 as having very high effectiveness and technical feasibility, with low uncertainty. It has to be noted that the pathway of citrus plants for planting, as well as that of Tahiti lime plants for planting, has a likely overall rating for entry and a very likely transfer to suitable host.

4.4.7. Remarks concerning pathway 'citrus plants for planting for citrus fruit production, passenger traffic'

Since citrus plants for planting are subject to prohibition of import according to Annex III of Council Directive 2000/29/EC instead of special requirements of Annex IV Part A, the exceptions of Article 5 point 4 of the Directive do not apply. The prohibition of import of plants for planting via passengers traffic is the only option for this pathway evaluated (see section 4.1.4.1) with at least moderate effectiveness, although with low feasibility.

4.5. Conclusions on the analysis of risk reduction options and on the current phytosanitary measures

For the reduction of the probability of entry of *P. citricarpa*, prohibition and import from pest-free areas have overall a high to very high effectiveness with moderate to high feasibility for all pathways. Prohibition of parts of the host also has high effectiveness and very high feasibility with regard to the prohibition of citrus fruit with leaves and peduncles. For the fruit pathway, systems approaches as well as the induction of precocious symtoms expression also have high effectiveness and feasibility. For plants for planting, certification and pre- and post-entry quarantine systems were also found to have high effectiveness and feasibility.

For reduction of the probability of establishment and spread, the application of strict waste processing measures would be highly effective in reducing the transfer of *P. citricarpa* from infected citrus fruit, both for entry and spread, although with low feasibility. The effectiveness of eradication, as well as of containment, is assessed as low. The application of drip irrigation practices will moderately reduce the probability of establishment.

Current EU phytosanitary measures appear to be focusing on the correct strategy to reduce the risk of *P. citricarpa* introduction. Once established, CBS is reported to be very difficult to eradicate or contain. The Panel thus concludes that every effort should be made to avoid the entry of the pathogen. Should the disease be reported from the risk assessment area, limited options are available to reduce the risk of establishment and spread. Current EU measures are overall judged to be effective in preventing the introduction of *P. citricarpa*.

CONCLUSIONS

The Panel on Plant Health (PLH) conducted the risk assessment following the guidance documents for producing standardized assessments of pest risk (EFSA Panel on Plant Health (PLH), 2010) and risk reduction options (EFSA Panel on Plant Health (PLH), 2012). The Panel conducted the risk assessment in the absence of current and potential new risk reduction measures in place. The risk assessment therefore evaluates the full risk posed by *P. citricarpa* to the EU territory corresponding to a situation where all current EU requirements listed in Council Directive 2000/29/EC and Commission Decisions 2004/416/EC and 2006/473/EC are lifted without being replaced by any other risk reduction measures.

The risk assessment covers *Guignardia citricarpa* Kiely, which has since been renamed *Phyllosticta citricarpa*. Other *Phyllosticta species* are not included.

After consideration of the evidence, the Panel reached the following conclusions:

With regard to the pest categorisation:

P. citricarpa is absent from the EU and has a potential for establishment and spread and for causing consequences in the risk assessment area

With regard to the assessment of the risk to plant health for the EU territory:

Under the worst case scenario in which all current EU requirements listed in Council Directive 2000/29/EC and Commission Decisions 2004/416/EC and 2006/473/EC would be lifted, the conclusions of the pest risk assessment are as follows:

<u>Entry</u>

The probability of entry is rated as:

- Moderately likely for the citrus fruit trade pathway (medium uncertainty),
- very unlikely for the Tahiti lime (*Citrus latifolia*) fruit trade pathway (high uncertainty),
- unlikely for citrus fruit import by passengers traffic pathway (medium uncertainty),
- likely for the citrus fruit with leaves trade pathway (medium uncertainty),
- likely for the citrus plants for planting trade pathway (low uncertainty),
- likely for the Tahiti lime (*Citrus latifolia*) plants for planting trade pathway (high uncertainty),
- likely for the citrus plants for planting import by passengers traffic (medium uncertainty),
- unlikely for the citrus leaves (medium uncertainty)

<u>Establishment</u>

The probability of establishment is rated as moderately likely because of:

- the widely availabity of suitable hosts (no uncertainty),

- the climate suitability for ascospores maturation, dispersal and infection of many EU citrus growing areas in September and October and for specific location also in May (high uncertainty),
- cultural practices (fungicides) not preventing establishment (low uncertainty),
- sprinkle and micro-sprinkle irrigation (still used in part of the EU citrus growing areas) favouring establishment (low uncertainty),
- the simultaneous occurrence of host susceptibility and of weather conditions suitable for ascospore production and release as well as for ascospore germination and infection (high uncertainty).

Overall, the uncertainty on the probability of establishment is rated as high, mainly due to lack of knowledge on how *P. citricarpa* will respond under the EU climatic conditions. Although it is known for the organism *which* environmental factors are important in the various stages of the life cycle, there is lacking scientific evidence to precisely determine the exact threshold for *what* values the organism require, e.g. for the temperature and wetness levels and durations. Further validation of the models applied by the Panel, especially for marginal areas within the current distribution of the citrus black spot disease would be needed to reduce the uncertainty on the probability of establishment of *P. citricarpa* in the EU.

<u>Spread</u>

Natural spread of *P. citricarpa* is known to mainly happen by dispersalo of airborne ascospores. There is little evidence about the dispersal distances of the pathogen by natural means, The pathogen is very likely to spread with human assistance along the commercial fruit and plants for planting pathways. However, because spread is defined as the expansion of the geographical distribution of a pest within an area, the rate of spread depends not only on the rapidity of movement and the number of spread pathways but also the likelihood of finding a suitable environment for establishment. When the proportion of the citrus growing areas identified as potentially suitable for *P. citricarpa* is taken into account, the Panel considered that a rating of moderately likely is most appropriate for spread.

Although there is uncertainty about the potential natural spread of ascospores carried by wind over long distances, this uncertainty does not concern the two main pathways of spread (intra-European trade of commercial fruit and plants for planting). There is uncertainty about the structure of the EU intra-trade network for the citrus plants for planting owing to lack of data, however this does not influence the conclusions above.

<u>Endangered area</u>

The risk assessment has identified parts of the EU where host plants are present and where, based on simulation results, the climatic conditions are suitable for ascospore maturation and release followed by infection.

Conclusions from simulations of the release of ascospores based on gridded interpolated climate data of the EU citrus producing areas show that, in almost all years (for the 95 percentile), ascospore release in areas of Cyprus, Crete, Southern Greece, Italy, Spain and Portugal will start early enough to coincide with climatic conditions that are conducive to infection in September and October as simulated by EFSA (2008). However the same simulations indicate that the onset of ascospore release in most areas will start too late to coincide with the climatic conditions conducive to infection in April-May. Therefore, early maturing orange varieties might generally be infected in autumn only, which is when the availability of inoculum coincides with suitable conditions for infection. Due to the long incubation time, fruits from these early varieties will be harvested before symptoms appear. The late maturing oranges varieties and lemons are expected instead under such scenario to show CBS symptoms.

There are some areas however, such as locations in Cyprus, Greek islands, Malta and Southern Spain where onset of ascospores is expected also in May in half of the years simulated. In those years, it is expected that symptoms can develop on the fruit before harvest, and therefore have an impact on the fruit quality.

The results from the simulations on interpolated (grid-based) weather data are consistent with the simulations run on weather data measured by agrometereological stations. The uncertainty is high as indicated in the establishment section.

Consequences

The results from the simulation of ascospore maturation, release and infection show that citrus black spot will develop and express symptoms mainly in late maturing sweet orange varieties and lemons grown within the endangered area. The expected consequences will be moderate for fresh fruit of late maturing citrus varieties and lemons. There would a potential for reduction of disease incidence by chemical treatments, but this would cause environmental impacts owing to the fact that in most EU citrus growing areas fungicides are not widely applied and that the most effective fungicide products are not currently registered for use in citrus in the EU MSs. In addition, to export citrus fruit to areas where CBS is regulated, additional fungicide treatments in the orchards, official inspections, quality controls in packinghouses and/or establishment of pest free areas might be needed to meet the phytosanitary requirements of these countries.

The consequences for fresh fruit of early maturing citrus varieties are assessed as minor. The impact on early maturing varieties would be sporadic in time and space, limited to years with rainy springs and/or to specific locations. However the impact could be higher in areas where spring infection, based on simulation results, is expected to be more frequent, such as some locations in Southern Spain, Cyprus, Malta and Greek islands.

The consequences would be minimal for citrus for processing, as external lesions or spots on citrus fruit are not a quality issue for the citrus for processing.

As for establishment, the uncertainties about consequences are high due to: the lack of information on key parameters in the epidemiological models and on te incubation period, the lack of knowledge about the rate of disease build-up for this pathogen; the limited information available about the impact and the fungicides treatments in marginal areas within the current CBS area of distribution, eg Eastern Cape, where environmental conditions are more similar to those in the pest risk assessment area.

With regard to risk reduction options, the Panel notes that, for reduction of the probability of entry of *P. citricarpa*, prohibition and import from pest-free areas have overall high to very high effectiveness with moderate to high feasibility for all pathways. Prohibition of parts of the host has also high effectiveness and very high feasibility with regard to the prohibition of citrus fruit with leaves and peduncles. For the fruit pathway, systems approaches as well as induction of precocious symtoms expression have also high effectiveness and feasibility. For plants for planting, certification and pre- and post-entry quarantine systems were also found having high effectiveness and feasibility.

For reduction of the probability of establishment and spread, the application of strict waste processing measures would be highly effective in reducing the transfer of *P. citricarpa* from infected citrus fruit, both for entry and spread, although with low feasibility. The effectiveness of eradication, as well as of containment, is assessed as low. The application of drip irrigation practices cover crops and mulching will moderately reduce the probability of establishment.

Current EU phytosanitary measures appear to be focusing on the correct strategy to reduce the risk of *P. citricarpa* introduction. Once established, CBS is reported to be very difficult to eradicate or contain. The Panel thus concludes that every effort should be made to avoid the entry of the pathogen. Should the disease be reported from the risk assessment area, limited options are available to reduce the risk of establishment and spread. Current EU measures are overall judged to be effective in preventing the introduction of *P. citricarpa*.

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APPENDICES

Appendix A. Rating descriptors

In order to follow the principle of transparency as described under Paragraph 3.1 of the Guidance document on the harmonised framework for risk assessment (EFSA, 2010)—"*…Transparency requires that the scoring system to be used is described in advance. This includes the number of ratings, the description of each rating … the Panel recognises the need for further development…*"—the Plant Health Panel has developed specifically for this opinion rating descriptors to provide clear justification when a rating is given.

Table 18: Ratings used in the conclusion of the pest risk assessment

In this opinion of EFSA's Plant health Panel on the risk assessment of *P. citricarpa* and the evaluation of the effectiveness of the risk reduction options, a rating system of five levels with their corresponding descriptors has been used to formulate separately the conclusions on entry, establishment, spread and impact as described in the following tables.

1.1. Rating of probability of entry

Rating for entry	**Descriptors for *P. citricarpa***
Very unlikely	The likelihood of entry would be very low because the pest: • is not, or is only very rarely, associated with the pathway at the origin; and/or • may not survive during transport or storage; and/or • cannot survive the current pest management procedures existing in the risk assessment area; and/or • may not transfer to a suitable host in the risk assessment area.
Unlikely	The likelihood of entry would be low because the pest: • is rarely associated with the pathway at the origin; and/or • survives at a very low rate during transport or storage; and/or • is strongly limited by the current pest management procedures existing in the risk assessment area; and/or • has considerable limitations for transfer to a suitable host in the risk assessment area.
Moderately likely	The likelihood of entry would be moderate because the pest: • is frequently associated with the pathway at the origin; and/or • survives at a low rate during transport or storage; and/or • is affected by the current pest management procedures existing in the risk assessment area;

	and/or • has some limitations for transfer to a suitable host in the risk assessment area.
Likely	The likelihood of entry would be high because the pest: • is regularly associated with the pathway at the origin; and/or • mostly survives during transport or storage; and/or • is partially affected by the current pest management procedures existing in the risk assessment area; and/or • has very few limitations for transfer to a suitable host in the risk assessment area.
Very likely	The likelihood of entry would be very high because the pest: • is usually associated with the pathway at the origin; and/or • survives during transport or storage; and/or • is not affected by the current pest management procedures existing in the risk assessment area; and/or • has no limitations for transfer to a suitable host in the risk assessment area.

1.2. Rating of probability of establishment

Rating for establishment	Descriptors for *P. citricarpa*
Very unlikely	The likelihood of establishment would be very low because, although the host plants are present in the risk assessment area, the environmental conditions are unsuitable and/or the host is susceptible for a very short time during the year; other considerable obstacles to establishment occur.
Unlikely	The likelihood of establishment would be low because, although the host plants are present in the risk assessment area, the environmental conditions are mostly unsuitable and/or the host is susceptible for a very short time during the year; other obstacles to establishment occur.
Moderately likely	The likelihood of establishment would be moderate because, although the host plants are present in the risk assessment area, the environmental conditions are frequently unsuitable and/or the host is susceptible for short time; other obstacles to establishment may occur.
Likely	The likelihood of establishment would be high because the host plants are present in the risk assessment area, they are susceptible for a long time during the year, and the environmental conditions are frequently suitable; no other obstacles to establishment occur.
Very likely	The likelihood of establishment would be very high because the host plants are present in the risk assessment area, they are susceptible for a long time during the year, and the environmental conditions are suitable for most of the host growing season; no other obstacles to establishment occur. Alternatively, the pest has

	already been established in the risk assessment area.

1.3. Rating of probability of spread

Rating for spread	**Descriptors for *P. citricarpa***
Very unlikely	The likelihood of spread would be very low because the pest: • has only one specific way to spread (e.g., a specific vector) which is not present in the risk assessment area; and/or • highly effective barriers to spread exist; and/or • the host is not or is only occasionally present in the area of possible spread; and/or • the environmental conditions for infestation are unsuitable in the area of possible spread.
Unlikely	The likelihood of spread would be low because the pest: • has one or only a few specific ways to spread (e.g., specific vectors) and its occurrence in the risk assessment area is occasional; and/or • effective barriers to spread exist; and/or • the host is not frequently present in the area of possible spread; and/or • the environmental conditions for infestation are mostly unsuitable in the area of possible spread.
Moderately likely	The likelihood of spread would be moderate because the pest: • has few specific ways to spread (e.g., specific vectors) and its occurrence in the risk assessment area is limited, and/or • effective barriers to spread exist; and/or • the host is moderately present in the area of possible spread; and/or • the environmental conditions for infestation are frequently unsuitable in the area of possible spread.
Likely	The likelihood of spread would be high because the pest: • has some unspecific ways to spread, which occur in the risk assessment area; and/or • no effective barriers to spread exist; and/or • the host is usually present in the area of possible spread; and/or • the environmental conditions for infestation are frequently suitable in the area of possible spread.

Very likely	The likelihood of spread would be very high because the pest: • has multiple unspecific ways to spread, all of which occur in the risk assessment area; and/or • no effective barriers to spread exist; and/or • the host is widely present in the area of possible spread; and/or • the environmental conditions for infestation are mostly suitable in the area of possible spread.

1.4. Rating of magnitude of the potential consequences

Rating of potential consequences	**Descriptors for *P. citricarpa***
Minimal	Differences in crop production are within normal day-to-day variation; no additional control measures are required.
Minor	Crop production is rarely reduced or at a limited level; additional control measures are rarely necessary.
Moderate	Crop production is occasionally reduced to a limited extent; additional control measures are occasionally necessary.
Major	Crop production is frequently reduced to a significant extent; additional control measures are frequently necessary.
Massive	Crop production is always or almost always reduced to a very significant extent (severe crop losses that compromise the harvest); additional control measures are always necessary.

1. Ratings used for the evaluation of the risk reduction options

The Panel developed the following ratings with their corresponding descriptors for evaluating the effectiveness of the risk reduction options to reduce the level of risk.

1.1. Rating of the effectiveness of risk reduction options

Rating	**Descriptors for *P. citricarpa***
Negligible	The risk reduction option has no practical effect in reducing the probability of entry or establishment or spread, or the potential consequences.
Low	The risk reduction option reduces, to a limited extent, the probability of entry or establishment or spread, or the potential consequences.
Moderate	The risk reduction option reduces, to a substantial extent, the probability of entry or establishment or spread, or the potential consequences.
High	The risk reduction option reduces the probability of entry or establishment or spread, or the potential consequences, by a major extent.

Very high	The risk reduction option essentially eliminates the probability of entry or establishment or spread, or any potential consequences.

1.2. Rating of the technical feasibility of risk reduction options

Rating	**Descriptors for *P. citricarpa***
Negligible	The risk reduction option is not in use in the risk assessment area, and the many technical difficulties involved (e.g., changing or abandoning the current practices, implement new practices and or measures) make their implementation in practice impossible.
Low	The risk reduction option is not in use in the risk assessment area, but the many technical difficulties involved (e.g., changing or abandoning the current practices, implementing new practices and/or measures) make its implementation in practice very difficult or nearly impossible.
Moderate	The risk reduction option is not in use in the risk assessment area, but it can be implemented (e.g., changing or abandoning the current practices, implementing new practices and/or measures) with some technical difficulties.
High	The risk reduction option is not in use in the risk assessment area, but it can be implemented in practice (e.g., changing or abandoning the current practices, implement new practices and or measures) with limited technical difficulties.
Very high	The risk reduction option is already in use in the risk assessment area or can be easily implemented with no technical difficulties.

2. Ratings used for describing the level of uncertainty

For the risk assessment chapter—entry, establishment, spread and impact—as well as for the evaluation of the effectiveness of the risk reduction options, the level of uncertainty has been rated separately in coherence with the descriptors that have been defined specifically by the Panel in this opinion.

Rating	**Descriptors for *P. citricarpa***
Low	No or little information or no or few data missing, incomplete, inconsistent or conflicting. No subjective judgement is introduced. No unpublished data are used.
Medium	Some information is missing or some data are missing, incomplete, inconsistent or conflicting. Subjective judgement is introduced with supporting evidence. Unpublished data are sometimes used.
High	Most information is missing or most data are missing, incomplete, inconsistent or conflicting. Subjective judgement may be introduced without supporting evidence. Unpublished data are frequently used.

Appendix B. Additional trade and interceptions figures

Table 19: Import of citrus fruit (tons) from Third Countries where P. citricarpa is reported into the EU. Product = 0805 citrus. Yearly data (from January to December) from 2002 to 2011. Extracted from Eurostat on 22/02/2013.

PERIOD/ PARTNER	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011
ARGENTINA	285842	358827	315900	385569	331398	391136	424803	307875	315528	280264
AUSTRALIA	1271	2743	1875	4808	1775	5950	2697	3983	1622	465
BRAZIL	47200	88630	86358	65004	96139	85222	78142	73094	89206	83795
BHUTAN										
CHINA	41	361	1524	6166	17110	43715	68235	75452	72478	47803
CUBA	22217	31598	18964	15989	10363	7281	2979	2197	1374	1375
GHANA	7	33			20	0.7	1232	2064	672	312
INDONESIA	18				0.1					
KENYA		9	0							0.1
MOZAMBIQUE		94	910	1082	121			285	989	1587
NEW ZEALAND	172	73		20	130	219	82	122	2	11
PHILIPPINES				0			3			0.3
TAIWAN	16	1.2			0.3	0.8		0.5	0.1	
UGANDA	22	13	0			0	0	0.3	0	0
UNITED STATES	122324	107838	100255	54926	55532	65665	89823	62964	58743	63673
URUGUAY	68167	95219	85302	122878	115349	116738	99045	103616	118951	90050
VANUATU										
SOUTH AFRICA	463832	473011	407357	539363	468412	641822	676519	527017	604160	535988
ZAMBIA	0	1.8		12						
ZIMBABWE	37626	37023	18882	36046	15666	29409	18104	15519	25816	13873

EU = EU27 (AT, BE, BG, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LT, LU, LV, MT, NL, PL, PT, RO, SE, SI, SK)

Interception of living stages of *P. citricarpa* on citrus fruit consignments imported into the EU-27 MSs.

According to Europhyt (2010), during the period 2005-2010, there have been 560 notifications from EU-MSs of interceptions of *P. citricarpa* on imported citrus fruit consignments originated in CBS-infested countries. In the Netherlands alone, 20 consignments of citrus fruit infected with *P. citricarpa* have been intercepted in 2010. The average size of the intercepted consignments was 20 tonnes (source: NPPO of the Netherlands, 2010).

Figure 44 shows the trend through time in the number of yearly interceptions (all EU countries, 1999-2012). There is an increase through time until the onset of the financial crisis (2008), after which the number of interceptions goes back to levels comparable to the beginning of the 2000s (with the exception of 2011).

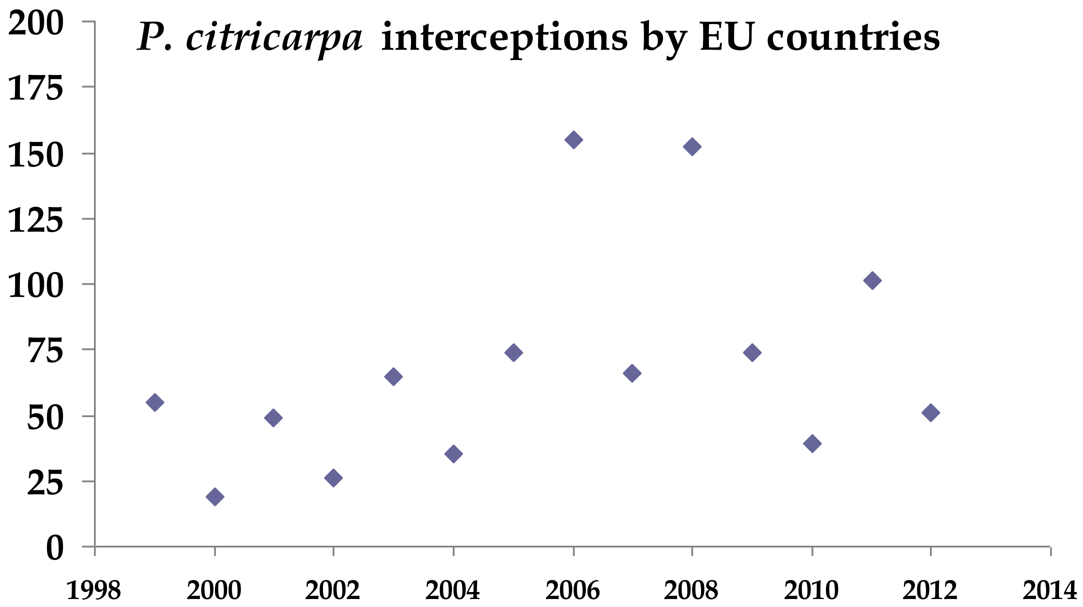

Figure 44: Temporal trend in the number of *P. citricarpa* EU interceptions on citrus fruit consignments from Third Countries (1999-2012).

Figure 45 shows the temporal trend in the number of *P. citricarpa* interceptions per unit of trade (100,000 tons of citrus fruit, to all EU countries, from all origins, but only between 2002 and 2011 for lack of trade data in 1999-2001 and 2012). The trend is towards a weak increase (R^2 = 0.09) in the number of interceptions per unit of trade, with two outliers in 2006 and 2010.

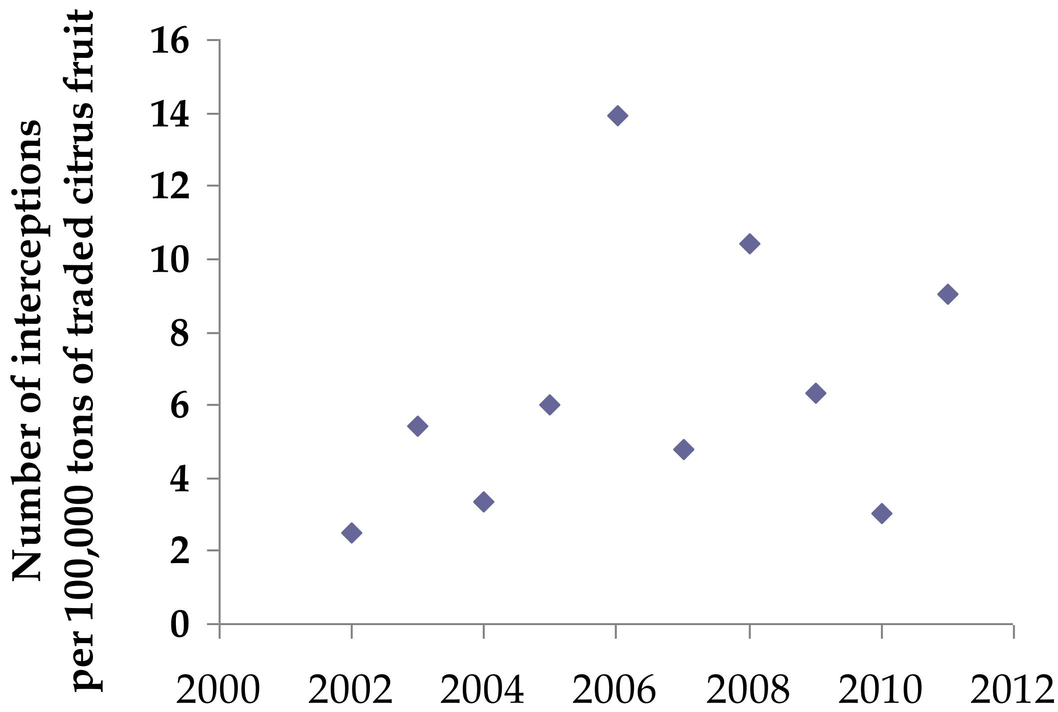

Figure 45: Temporal trend in the number of *P. citricarpa* EU interceptions *(per unit of trade)* on citrus fruit consignments from Third Countries (2002-2011).

The trend in citrus fruit imported by EU countries from Third Countries (2002-2011) may also suggest an influence of the economic crisis since 2008 (unless other factors explain the reduced trade volumes for 2009-2010-2011) (Fig. 46). Please note that this overall trend for all exporting countries may not be matched by the trends for single countries. For example, over the studied period (2002-2011), citrus fruit imported by the EU from Cuba and the USA went considerably down, whereas imports from China went from insignificant in 2002 to more than imports from the USA in 2010.

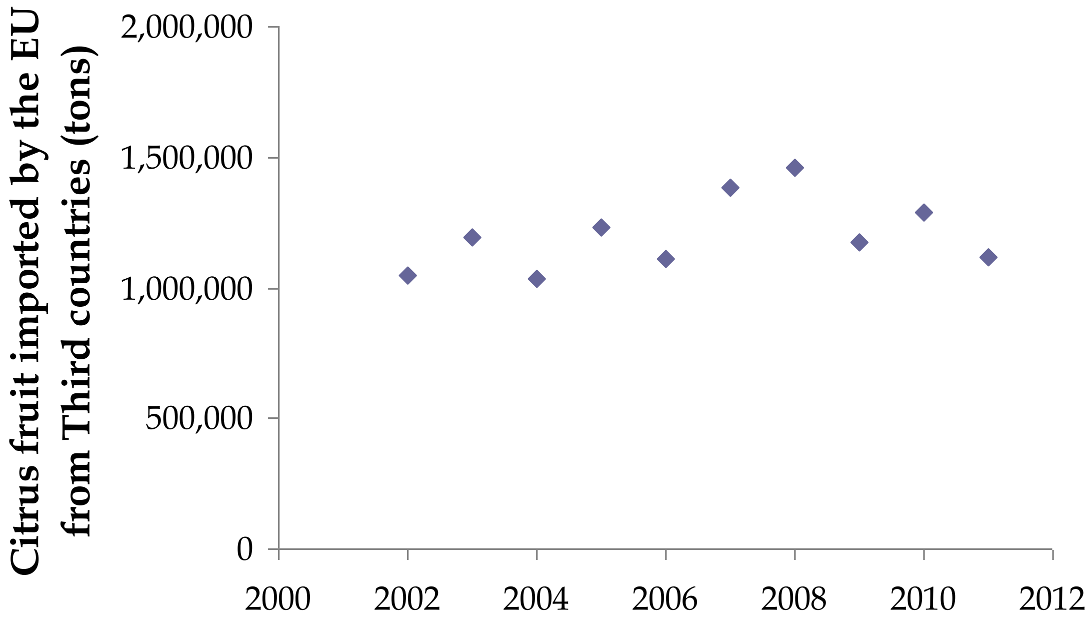

Figure 46: Temporal trend in the amount of citrus fruit imported by the EU from Third Countries (2002-2011).

There is a (weak; R^2 = 0.12) correlation between number of yearly *P. citricarpa* EU interceptions and amount of citrus fruit imported by the EU from Third Countries in the same year. The correlation is weak due to the high number of interceptions in 2006 (and, in part, in 2011) despite low trade volumes in the same year (Fig. 47). The weakness of the correlation does not imply that there is not an increased risk of entry of the pathogen with increasing amount of trade, as also other factors play a role in determining the number of interceptions over a whole year (e.g., climate, management practices and epidemic level during that particular year in the various countries of origin of consignments).

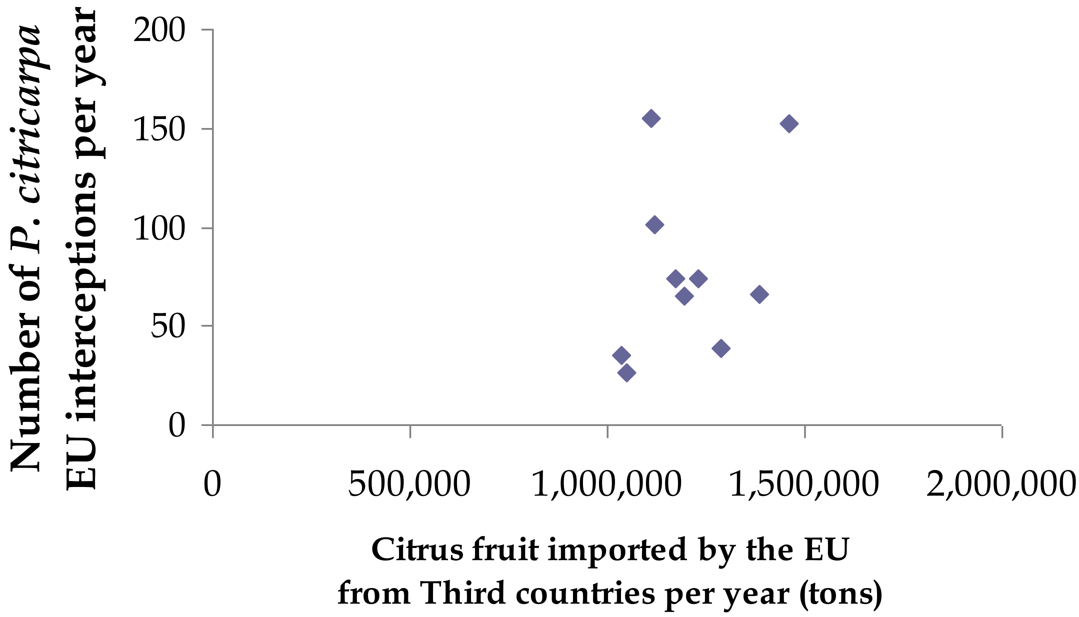

Figure 47: Correlation between number of *P. citricarpa* EU interceptions per year (all countries of origin, 2002-2011) and amount of citrus fruit imported by the EU from Third Countries (all countries of origin, 2002-2011).

Most of the oranges imported by the EU from South Africa and Argentina (two of the major citrus fruit exporters with reported presence of CBS) go to non citrus-producing countries, but the quantities of oranges imported by citrus-producing EU countries from these two countries are not negligibile (Fig. 48), and the pattern was reversed for Argentina in 2011. Mandarins from South Africa and Argentina go nearly exclusively to non citrus-producing EU countries. Also grapefruit from South Africa and Argentina is mostly imported by non-citrus producing EU countries. Most lemons exported by South Africa to the EU go to non citrus-producing countries, but this is not the case for Argentina, which mainly exports lemons to citrus-producing EU countries (Fig. 48).

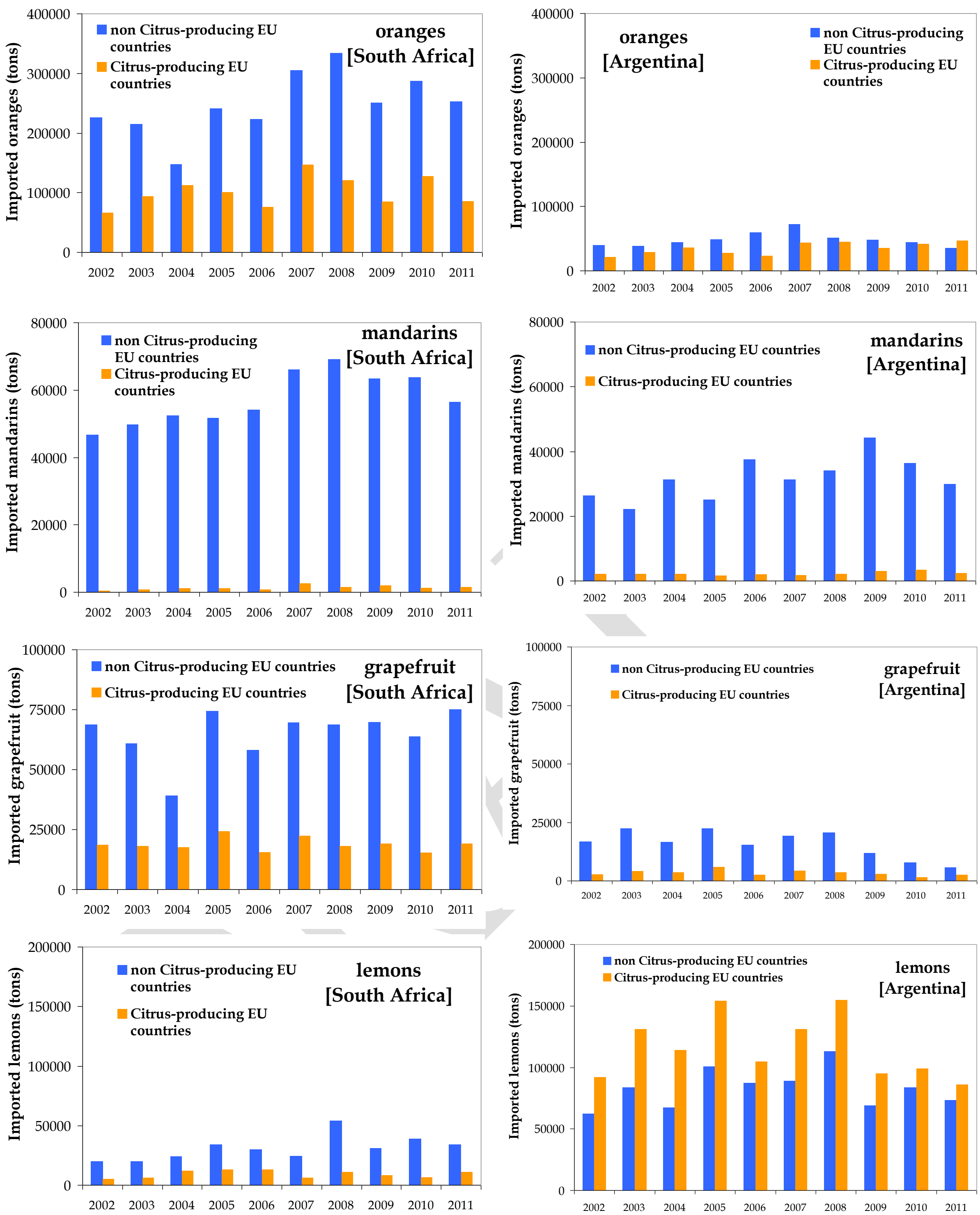

Figure 48: Imports of oranges, mandarins, grapefruit and lemons from (left-hand panel) South Africa and (right-hand panel) Argentina into citrus-producing and non-citrus producing EU countries (2002-2011). Note that the y-axis scales are consistent between South Africa and Argentina, but not among the different types of citrus fruit.

Appendix C. Comparison of ascospore release with time of possible start of infections for eight Italian stations in citrus growing areas

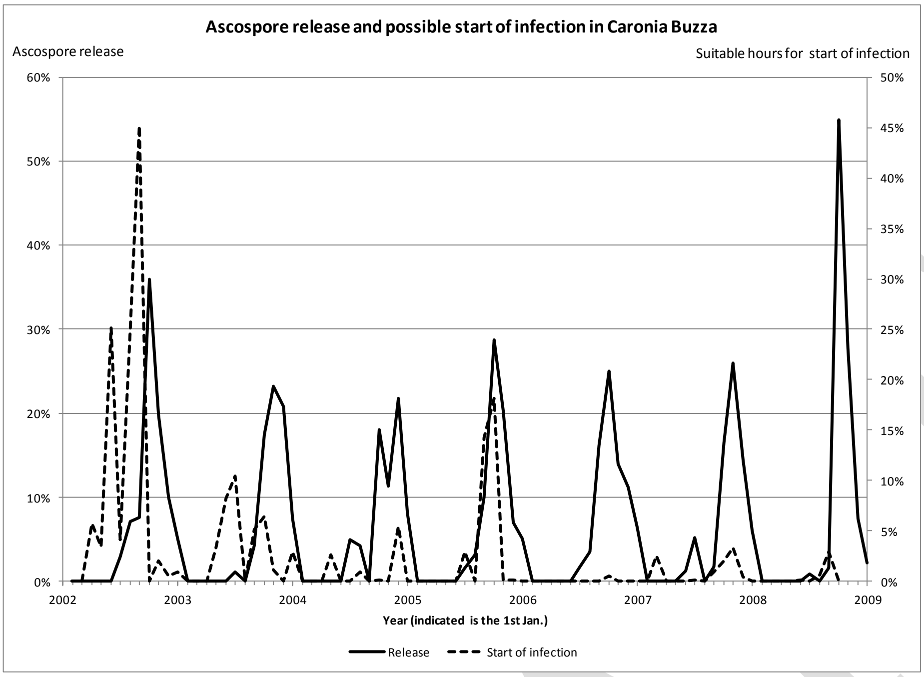

Figure 49: Comparison of ascospore release with time of possible start of infections

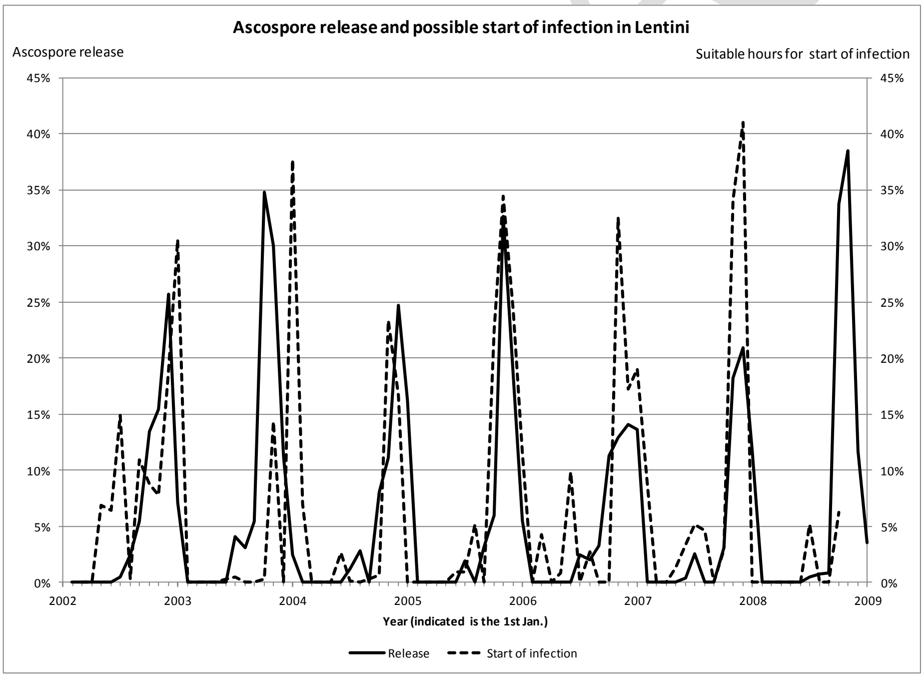

Figure 50: Comparison of ascospore release with time of possible start of infections.

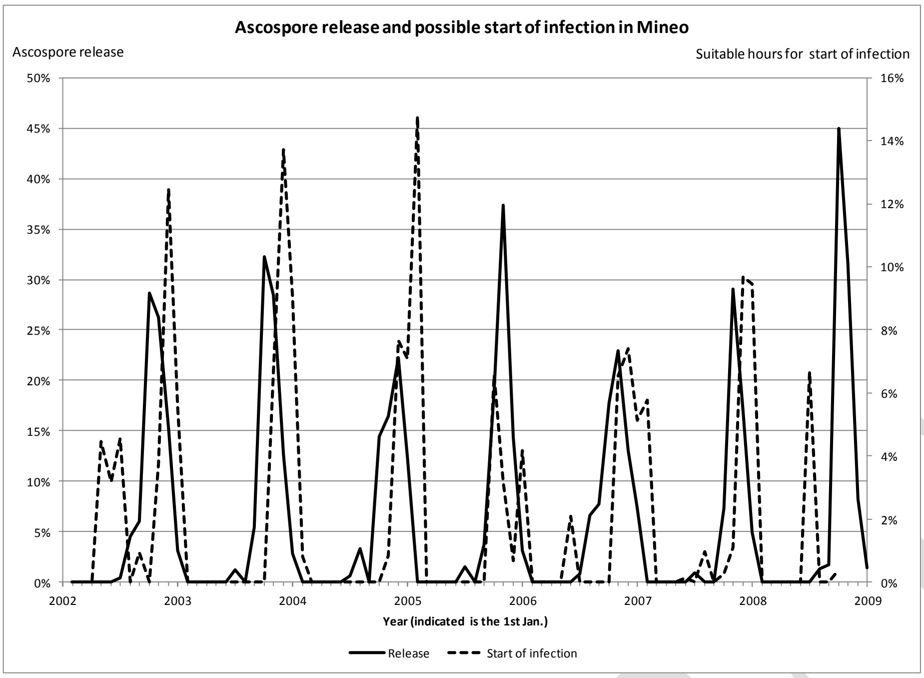

Figure 51: Comparison of ascospore release with time of possible start of infections

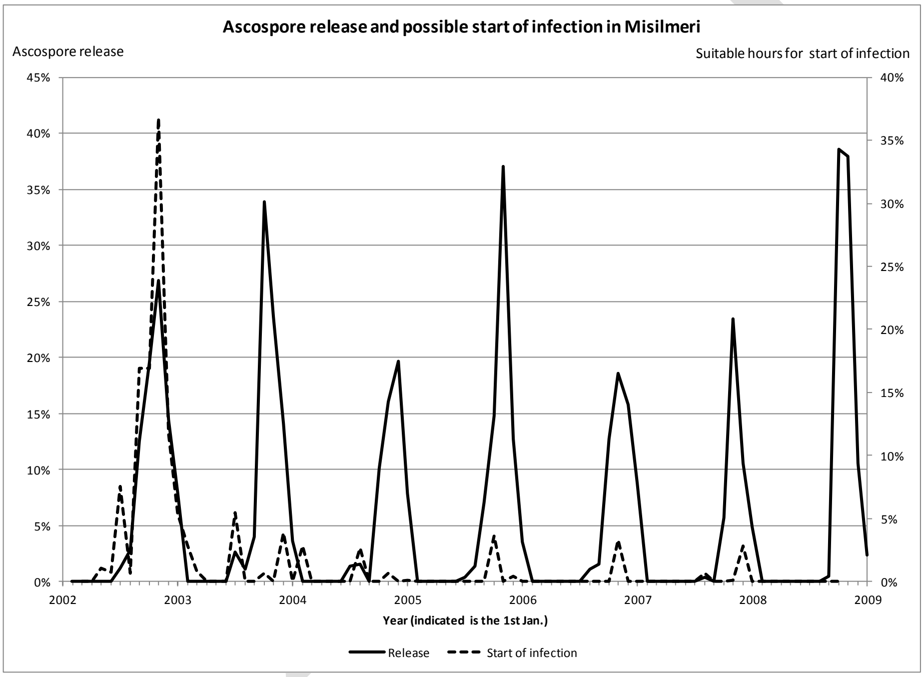

Figure 52: Comparison of ascospore release with time of possible start of infections.

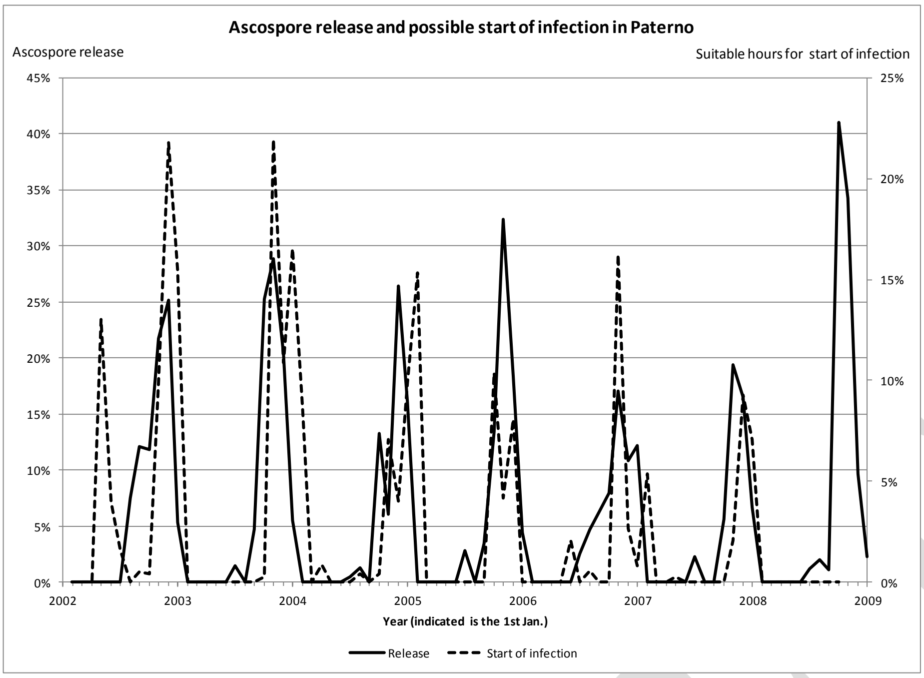

Figure 53: Comparison of ascospore release with time of possible start of infections

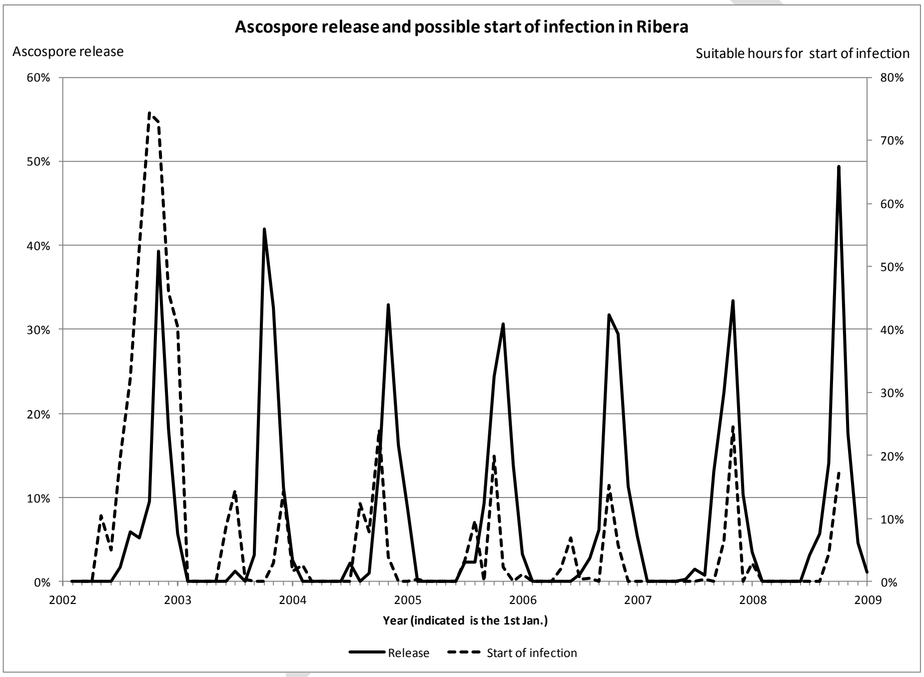

Figure 54: Comparison of ascospore release with time of possible start of infections.

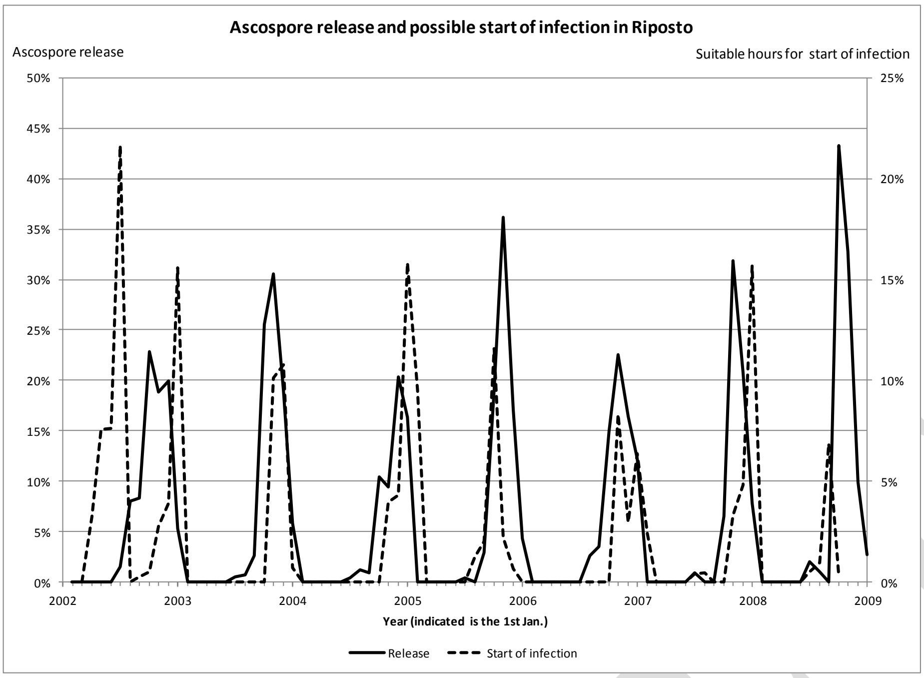

Figure 55: Comparison of ascospore release with time of possible start of infections.

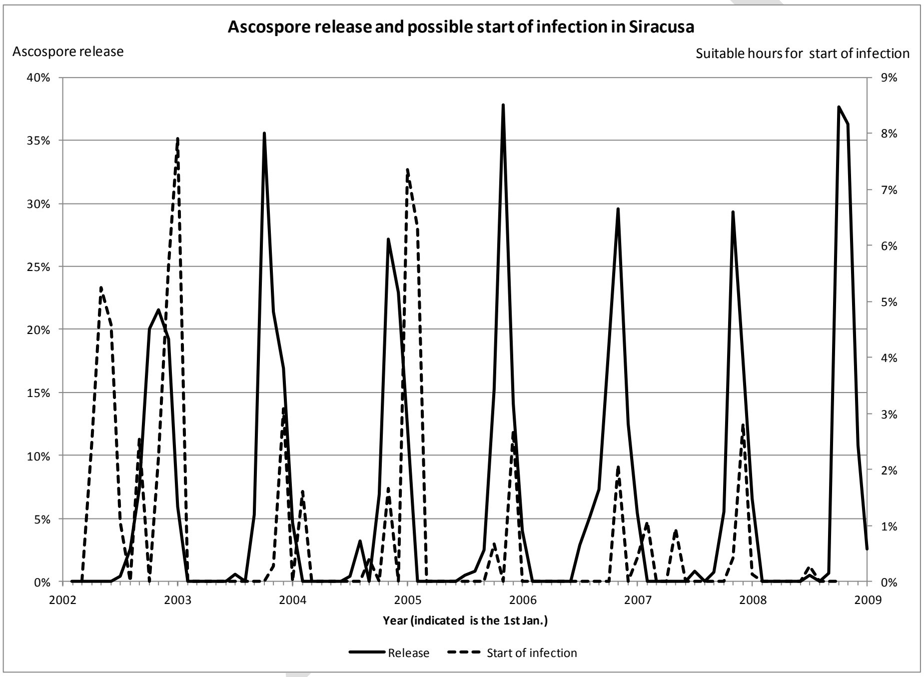

Figure 56: Comparison of ascospore release with time of possible start of infections.

Appendix D. Analysis of the sensitivity of the Yonow et al., 2013 Climex model results to European climate data with different spatial resolution and time periods coverage

Yonow et al (2013) recently parameterized a CLIMEX model for *P. citricarpa* to predict the potential global distribution of citrus black spot disease. Yonow et al (2013) paid particular attention to the risk of citrus black spot disease to Europe and found that "Within European citrus producing regions, suitable areas are highly constrained, never more than marginally suitable, and all have lower levels of suitability than any area in South Africa and Australia where *G. citricarpa* is known to occur." With regard to marginally suitable areas for the citrus black spot disease within its known current distribution range, Yonow et al (2013) mention Addo in South Africa, as an example of an area with marginal suitability for *P. citricarpa*. In Addo, *P. citricarpa* is present in the citrus growing areas and according to Yonow et al (2013) the pathogen persist there but "does not flourish" because of the marginal climatic suitability of the area for disease development.

The Panel notes that both of the above mentioned findings are of particular interest for the assessment of the risk posed by *P. citricarpa* to the EU territory. The Panel therefore explored the effect on the output of the Yonow et al (2013) model when (1) increasing the spatial resolution in the input climate data and (2) using more recent climate data (1961-90 vs. 1998-2007 EU climate averages).

Based on this analysis the Panel concludes that:

- The model results show sensitivity to variation in the spatial resolution and the time period of the input climate data

- For some of the EU citrus growing areas the climatic suitability classification varies from "marginally suitable", through "suitable" and even to "highly suitable" when changing either the spatial resolution or the temporal period covered by the input climate data (using the interpretation of the "Ecoclimatic index" used by Yonow et al 2013)

- The predicted potential for establishment in some EU citrus growing areas varies from EI =3 (1961-90 0.5° resolution), to EI=4 (1961-90 0.1° resolution) and EI = 11 (1998-2007 25 km resolution).

The Panel also undertook a limited investigation of the underlying CLIMEX calculations in order to reveal the climatic factors affecting the output. The CLIMEX model parameterised for *P. citricarpa* by Yonow et al. (2013) indicated that the northernmost citrus growing areas of Spain, located in the Ebro region, have a climate that is most suitable for citrus black spot. The grid cells with the highest "Ecoclimatic Index" as shown by Figure 57 indicate that there are two periods of the year, one in spring (week 15 – 22) and one in autumn (week 37 – 43) when temperature and moisture conditions occur at the same time to provide suitable climatic requirements according to the CLIMEX parameter set published by Yonow et al. (2013).

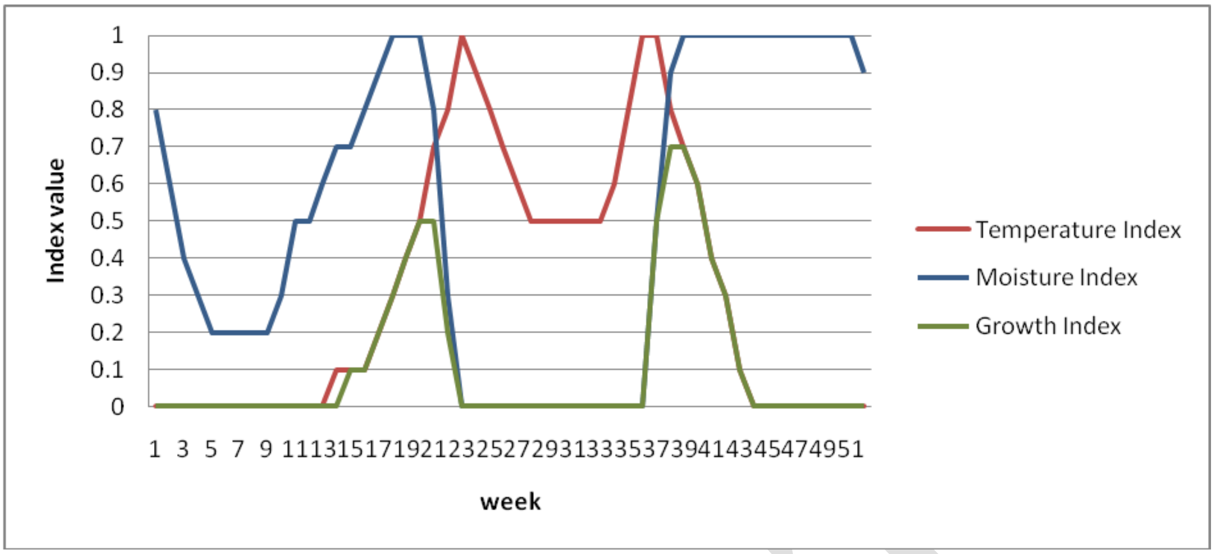

Figure 57: Weekly CLIMEX Temperature, Moisture and Growth Index values from the Ebro delta region in eastern Spain for 1961-90 at 0.1° latitude x longitude spatial resolution.

The Panel notes that understanding the climatic limitations of *P. citricarpa* is key to determining in which areas the organism can establish (i.e. persist) and it is important therefore to study the marginal areas to obtain these insights. Therefore, the Panel looked in detail at the underlying outputs from the model of Yonow et al (2013) for a limited number of locations of particular interest in order to understand how the model reacts to climate conditions indicated as marginal both within the EU citrus growing areas and the areas indicated as marginal within the known area of occurrence (e.g. Addo in Eastern Cape, SA) (Fig.58). The results show that the area around Addo is marginal for CBS as stated by Yonow et al (2013). However, when the spatial resolution is increased from 0.5° to 0.1° the Yonow et al (2013) model predicts the eastern part of Addo as unsuitable (Ecoclimatic index = 0).

As stated in section 3.3.2.2, CLIMEX cannot readily be used to analyse specific periods of the year when the host is at a susceptible stage and inoculum is potentially available. Moreover, it cannot directly take into account the effect of leaf wetness, a critical environmental variable for the successful infection of most fruit and foliage fungal pathogens including *P. citricarpa* (Kotzé, 1963 and 1981).

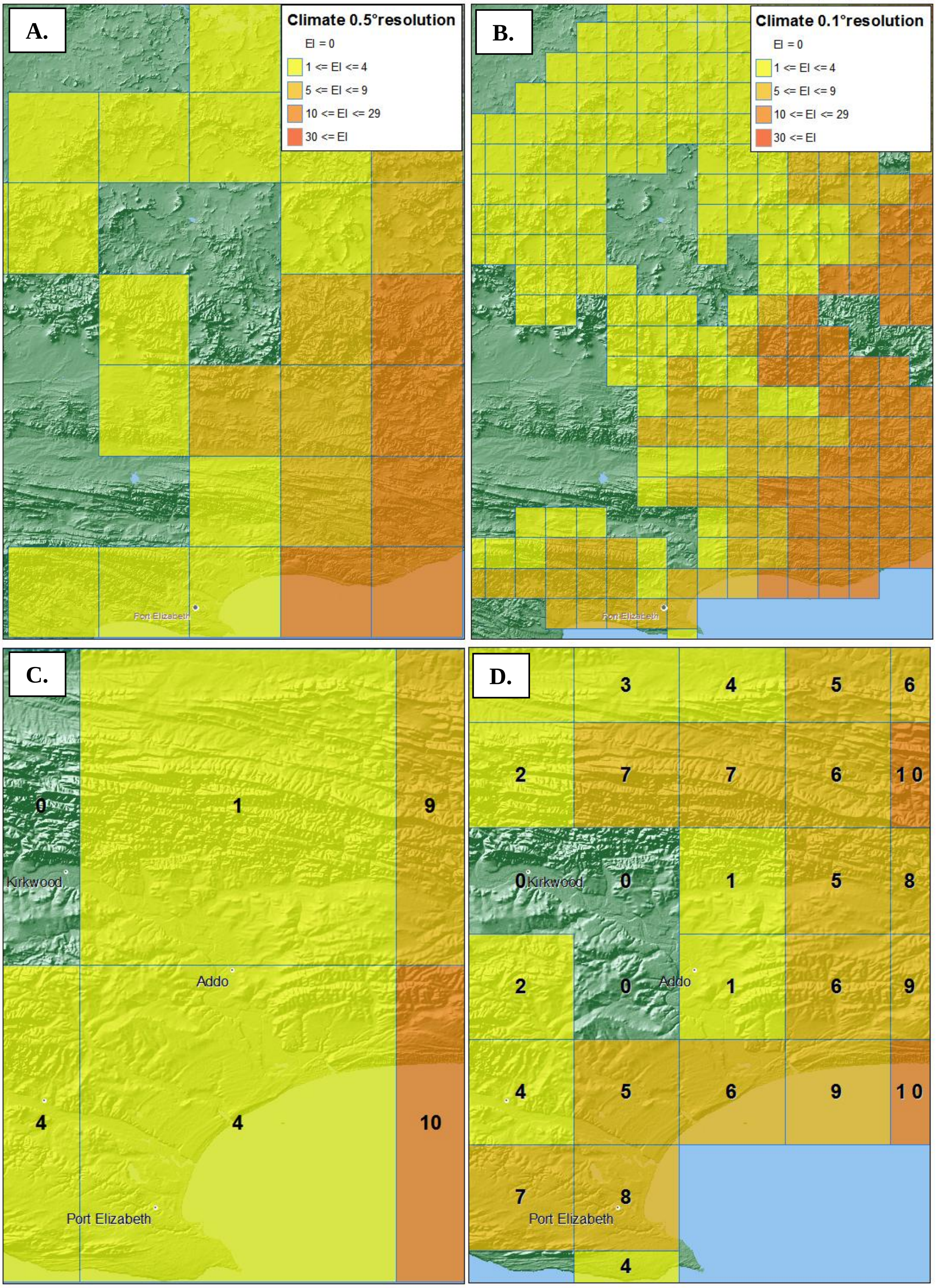

Figure 58: Detailed comparison of CLIMEX model output north of Port Elizabeth in South Africa, with two different spatial resolutions in the underlying interpolated climate data (sub-figures A. and C. 0.5°x0.5° latitude by longitude, sub-figures B. and D. 0.1°x0.1° latitude by longitude).