Fungi and Invertebrates associated with

Barberry (*Berberis* spp.) in New Zealand

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Contents

Summary

Project and Client

A survey of the invertebrate fauna and fungi associated with barberry (*Berberis glaucocarpa*), Darwin's barberry (*B. darwinii*), and European barberry (*B. vulgaris*) was carried out between October 2003 and March 2004 by Landcare Research for regional councils and the Department of Conservation. This was a recommendation of the feasibility study investigating the prospects for biological control of barberry (*Berberis* spp.) in New Zealand (McGregor 2002).

Objectives

- To survey fungi and invertebrates associated with barberry (*B. glaucocarpa*), Darwin's barberry (*B. darwinii*) and European barberry (*B. vulgaris*) in New Zealand and identify the herbivores (and their associated predators and parasitoids) and pathogens present.
- Based on these findings to recommend further action to be taken in the biological control of barberry programme.

Method

Barberry (*Berberis glaucocarpa*), Darwin's barberry (*B. darwinii*) and European barberry (*B. vulgaris*) sites were visited throughout New Zealand. Invertebrate fauna were sampled from barberry bushes, collected, preserved and identified. Fungi were also sampled at these sites, cultured and identified.

Results

- Fungi and invertebrates were recovered from 27 barberry (*Berberis glaucocarpa*), 13 Darwin's barberry (*B. darwinii*), and 1 European barberry (*B. vulgaris*) sites throughout New Zealand.
- 116 herbivorous invertebrate species were recorded from all barberry species – 108 species from *B. glaucocarpa*, 30 species from *B. darwinii*, and 2 species from *B. vulgaris.*
- The barberry aphid *Liosomaphis berberidis* (Kaltenbach) (Hemiptera: Aphididea) was the only herbivorous invertebrate species found to be specific to the barberry family Berberidaceae.
- The barberry aphid *L. berberidis* was the only herbivorous invertebrate to be classed as "abundant" (only on *B. glaucocarpa).*
- The overall damage that could be attributed to invertebrate herbivory was minimal.
- Generalist predators found on barberry included spiders, ladybirds, lacewings, earwigs, ants and a praying mantis.
- A total of 508 fungal isolates were obtained from a total 532 barberry tissues plated.
- A high frequency of fungal colonisation of barberry was observed with 95% of tissue fragments colonised.
- At least 24 fungal species were identified from the three barberry species.
- Three fungal species are potential primary pathogens of *B. glaucocarpa.*
- Four fungal species are potential primary pathogens of *B. darwinii.*
- No potential primary pathogens were isolated from *B. vulgaris.*
- A barberry rust, *Puccinia graminis*, was recorded from *B. glaucocarpa* at one site (Brancott Station), which is a new host record for New Zealand.

Conclusions

- A range of native and exotic invertebrate species in New Zealand attacks barberry but overall damage is minimal.
- Sap-feeders, e.g., the barberry aphid *Liosomaphis berberidis,* appear to be the most damaging invertebrates currently feeding on barberry in New Zealand

- None of the herbivore niches on barberry are well utilised in New Zealand, especially on *B. darwinii.*
- Specialised barberry feeding biological control agents are unlikely to meet with any significant competition from resident herbivores in New Zealand.
- There is scope for the introduction of host-specific biological control agents in New Zealand.
- Five primary plant pathogenic fungi were isolated from diseased barberry tissues, three of which have been the subject of various microbial weed biocontrol projects elsewhere. These fungi could potentially be investigated further for biocontrol potential against *Berberis* spp. in New Zealand.

Recommendations

In the light of our conclusions that only one host-specific invertebrate herbivore (*Liosomaphis berberidis)* contributes minimal damage to *Berberis* species in New Zealand, and with no specialised pathogenic fungi known to be present on these weeds in New Zealand, we recommend:

1. A classical biocontrol program for barberry should proceed along the following lines:

- Survey *B. darwinii* and other *Berberis* species in Chile and Argentina to identify prospective invertebrate and fungal biological control agents;
- Survey *B. glaucocarpa* and other *Berberis* species in the western Himalaya to identify prospective invertebrate and fungal biological control agents;
- Undertake host-range tests on plant species of importance to New Zealand for the most promising prospective invertebrates and fungal biological control agents;
- Introduce host-specific invertebrates and pathogenic fungi to New Zealand as classical biocontrol agents.

2. The inundative biocontrol potential of the *Colletotrichum gloeosporioides*, *Phomopsis* sp. and *Sclerotinia sclerotiorum* isolates associated with leaf, flower and fruit disease symptoms should be determined using inoculation trials to elucidate their taxonomy, epidemiology and pathogenicity.

3. The possible presence of bacterial or viral pathogens associated with leaf disease symptoms that were not associated with culturable fungal pathogens should be determined.

1. Introduction

Two of the five adventive species of barberry (*Berberis* spp.) in New Zealand are important environmental weeds, for which current control options are not adequate. As part of a biological control programme, sampling of invertebrate and fungal fauna associated with 3 barberry species in New Zealand – barberry (*Berberis glaucocarpa*), Darwin's barberry (*B. darwinii*), and European barberry (*B. vulgaris*) – was carried out between November 2003 and March 2004 by Landcare Research for regional councils and the Department of Conservation.

2. Background

Five species of Berberis have naturalised in New Zealand. *Berberis darwinii* (Darwin's barberry) and *B. glaucocarpa* (barberry) have been widely dispersed throughout New Zealand and have become serious environmental weeds. In this survey we have focused on these two species. A third barberry species, *B. vulgaris* (European barberry), although widespread in Canterbury and Otago (Webb et al. 1988) would not appear to be as problematic. The remaining two species (*B. soulieana* and *B. wilsoniae*) have very limited distributions (Webb et al. 1988).

Berberis darwinii is endemic to Chile and Patagonia (Webb et al. 1988). First introduced into New Zealand in 1946 this species is now widely distributed from the East Cape region southwards, with major stands occurring in Southland. An evergreen shrub growing to c. 4 m tall and found particularly in higher rainfall areas, this species is as happy growing under a native forest or plantation pine (*Pinus radiata*) canopy as it is infesting open hill slopes. Farmers (Martin Stott pers. comm.) and regional council staff (Randall Milne pers. comm.) view *B. darwinii* as an expanding threat to pastoral and conservation values, and as a recent immigrant it will likely continue to move into suitable habitat. Further large infestations are likely to be identified (McGregor 2002).

B. glaucocarpa, from the western Himalaya, is common throughout lowland areas of New Zealand (Webb et al. 1988). Extensively used as hedging, this species of barberry, like *B. darwinii*, is equally at home in a wide range of habitats with stands occurring in coastal reserves, grazed dairy pastures and drier sheep grazed slopes from Northland to Southland. First recorded in 1916, it arrived in New Zealand earlier than *B. darwinii*, and has had longer to invade and occupy its potential range. Large stands, as seen with gorse (*Ulex europaeus*) and broom (*Cytisus scoparius*), occur but are not yet common.

According to a feasibility study (McGregor 2002) both species are serious threats primarily to sparsely vegetated areas of bush and scrub. The invasiveness of these species arises mainly from their production of large quantities of fruits, which are eaten and subsequently dispersed by birds and possums (Allen & Wilson 1992; Williams & Karl 1996; Williams et al. 2000).

B. vulgaris has a more restricted distribution than either *B. darwinii* or *B. glaucocarpa*, being found predominantly in inland areas of Canterbury and Otago. Although first recorded in the wild in New Zealand some 40 years before any other species of barberry, it appears far less invasive than *B. darwinii* and *B. glaucocarpa*; its propensity to disperse seems to be more limited than that of those species (McGregor 2002).

3. Objectives

- Sample and identify the fungi, pathogens and invertebrates associated with barberry (*Berberis glaucocarpa*), Darwin's barberry (*B. darwinii*) and European barberry (*B. vulgaris*) from sites throughout New Zealand.
- Based on these findings, recommend further action to be taken in the biological control of barberry programme.

4. Method

In consultation with the Department of Conservation, regional councils and Landcare Research staff, barberry infestations throughout New Zealand were identified and a sampling programme was undertaken to determine if any damaging pathogens or invertebrates were present. Such a survey represents Step 4 in a classical biological control programme (Harley & Forno 1992).

Berberis glaucocarpa, B. darwinii and B. vulgaris were sampled from a total of 41 sites across both the North and South Islands between November 2003 and March 2004. Each of these sites was allocated a unique site specimen number (Appendix 1). The invertebrate fauna and pathogens of barberry (*B. glaucocarpa*), Darwin's barberry (*B. darwinii*) and European barberry (*B. vulgaris*) were sampled at 27, 13 and 1 sites, respectively (Figs. 1 and 2). Six *B. glaucocarpa* sites (Stonehurst Farm, Deadman's Island, Brancott Station, Kowhai River, Cheviot, and Kurow) were visited a second time during February or March 2004 to monitor any seasonal differences in the fungi and invertebrates present.

Plant specimens from 25 sites were collected and lodged with the Allen Herbarium (CHR) at Landcare Research, Lincoln, as voucher records of plant species from which both invertebrates and fungi were sampled.

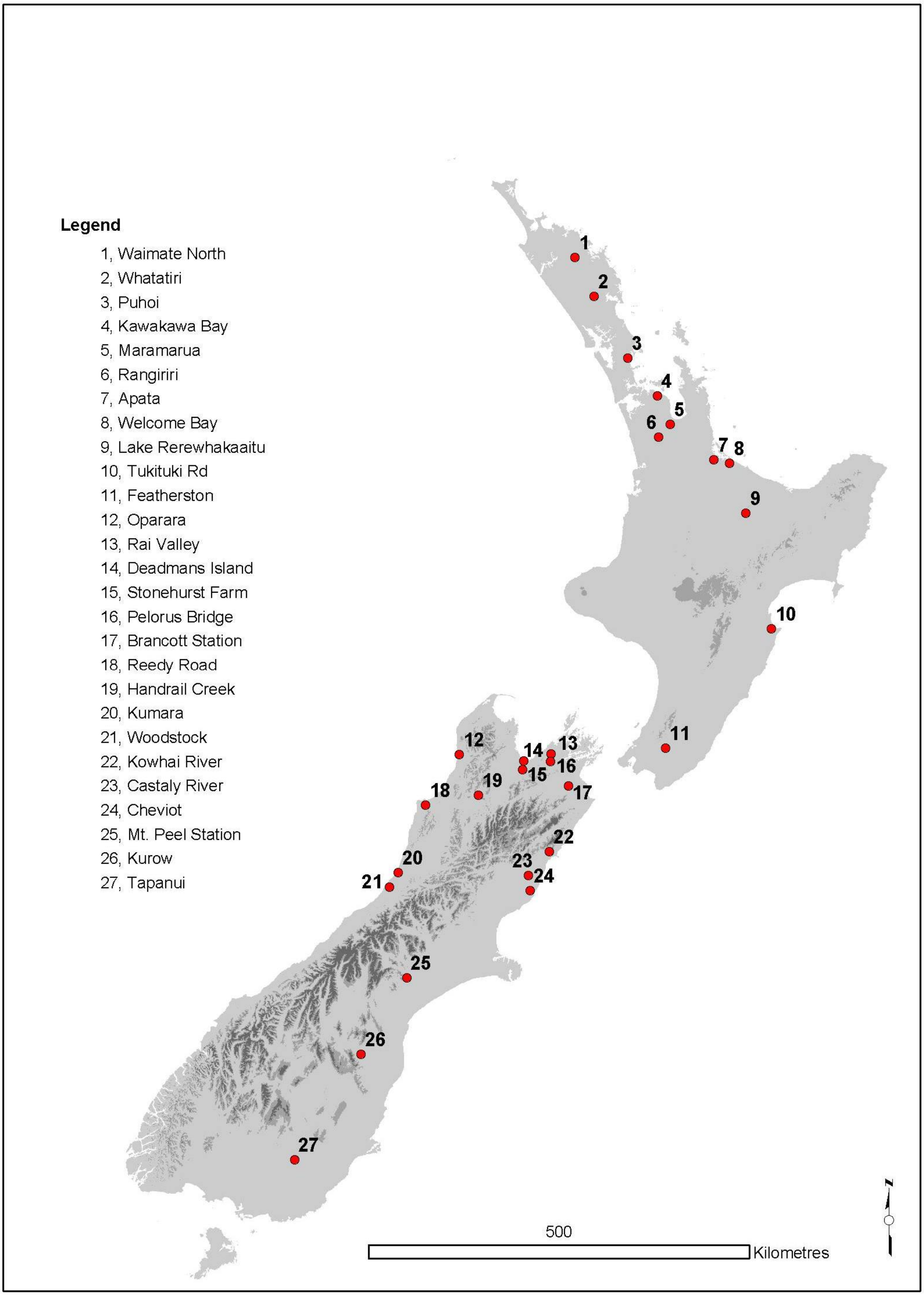

Fig. 1 Barberry (*Berberis glaucocarpa*) sites sampled for fungi and invertebrates 2003/04.

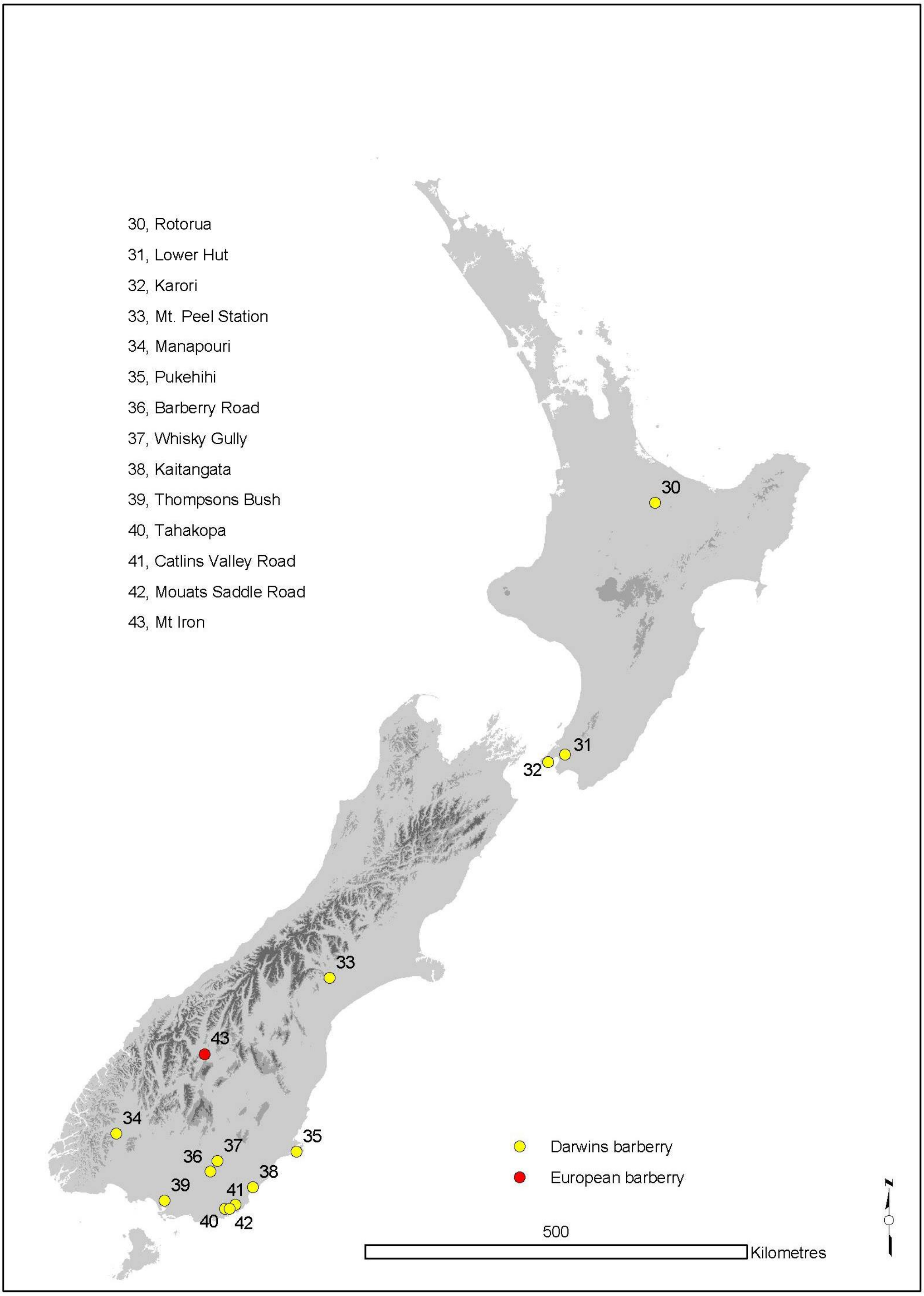

Fig. 2 Darwin's barberry (*Berberis darwinii*) and European barberry (*B. vulgaris*) sites sampled for fungi and invertebrates 2003/04.

4.1 Fungi

At each site, 10 plants selected for the invertebrate survey were inspected closely for signs of pathogen damage. Other barberry plants in the area were examined more superficially for obvious disease symptoms. Any diseased leaves, leaf petioles, stems, flower buds, flowers, flower petioles or fruit found were placed in paper bags, kept cool and sent to Landcare Research's laboratory in Tamaki for culturing. Collectors kept a special lookout for wilting plants, which might have been associated with a root disease previously recorded on barberry in New Zealand, and symptoms of rust infection as several rust species have been recorded on *Berberis* species overseas (McGregor 2002). Material was examined within 5 days of collection.

In the laboratory, disease symptoms were recorded and photographed. Necrotic areas were searched for fungal reproductive structures using a dissecting microscope. On microscopic examination, a number of samples exhibiting suspected fungal infection and disease symptoms were then selected for isolation onto microbiological media (Table 1). Small pieces of tissue (c. 3 mm^2) were cut from the edge of diseased areas and surface sterilised. Sterilisation was usually by immersion in 95% ethanol for 2–3 seconds, then in 2% Hypochlorite for 2 minutes, followed by rinsing twice in sterile water. The duration in hypochlorite was sometimes 1 or 3 minutes, depending on the toughness of the tissue, and immersion in ethanol was omitted on some occasions for tender young leaves or buds. The tissue fragments were blotted dry with sterile filter paper and placed on potato dextrose agar (Difco Labs, Detroit, MI, USA) with 0.02% streptomycin (Sigma, St Louis, MI, USA) contained in 9-cm petri dishes. Plates were incubated under near ultraviolet and white light (12 h photoperiod) at temperatures of 22 ± 2°C (day) and 18 ± 2°C (night).

Table 1 Number of fungi isolations from barberry samples onto media.

Host	Sample type			Total
	Leaf	Flower	Fruit	
B. glaucocarpa	32	5	11	48
B. darwinii	8	3	2	13
B. vulgaris	1	-	-	1
Total	41	8	13	52

Fungal colonies that grew out of the tissue fragments and produced spores were identified to the species level where possible. Taxonomic literature and fungal systematists were consulted to determine which of the identified fungi were likely to be causing the damage with which they were associated. Each identified isolate was given a unique number.

Representative portions of leaf, flower and fruit samples that exhibited leaf spot or brown lesions were also placed on moist blotting paper inside a sealable plastic tray (= a humidity chamber). Samples were incubated at room temperature and were misted with distilled water when required to maintain elevated humidity for 7–10 days or until any fungal sporulation developed from the diseased tissues. This method is often particularly useful to observe any latent *Botrytis cinerea* or *Colletotrichum* infection present on the samples. Fungal identification was undertaken by direct examination under the light microscope.

A rust fungus found at Brancott Station was initially given to Dr Eric McKenzie (Landcare Research) for identification using traditional mycological techniques. As these were not adequate to identify the rust, a specimen was sent to Dr Catherine Aime (USDA ARS Systematic Botany and Mycology Lab, Beltsville, MD, USA) for DNA sequencing. A dried specimen of the rust has been deposited in the New Zealand Fungal Herbarium (PDD), Tamaki, Auckland, as PDD 78317.

4.2 Invertebrates

At each site 10 barberry bushes were randomly selected from which insects were both beaten and hand collected. A collecting tray, 80 cm square, was placed under suitable parts of each selected plant, and the foliage above the tray was hit five times with a solid stick. Most invertebrates that fell onto the tray were collected with a pooter (aspirator) and preserved in 95% alcohol. Immature life stages, e.g., caterpillars (Lepidoptera) were collected live and placed, along with some foliage, in ventilated containers to rear through to adult stage in the laboratory for positive identification. Parasitoids emerging from these larvae or nymphs were also collected and identified. A rapid visual inspection, approximately 1 minute per plant, was made of foliage, growing points, and stems, for signs of invertebrates such as gall-formers, leaf miners, stem borers, and scale insects. Sections of the stems were cut to look for signs of invertebrates or their damage. Invertebrates found during the visual inspections were collected live, along with the plant material they were on, for identification. A visual estimate was made of the amount of herbivore damage, and the likely cause of the damage was noted (e.g., adult beetles, leaf-roller caterpillars).

Plants at three sites each for *B. glaucocarpa* (Kumara, Kowhai and Woodstock) and *B. darwinii* (Mouats Saddle, Catlins Valley Road and Pukehihi) were not beaten for invertebrate specimens as at the time of visiting rain precluded the use of an aspirator. The plants at these sites were still visually inspected and insects collected.

To identify seed-feeding insects, fruits and/or seeds were sampled by randomly collecting 10–20 stems bearing fruit or seed. These were returned to rearing facilities and held in ventilated containers for 8 weeks. Any insects emerging from these samples were collected and identified.

Most invertebrates collected were identified to species or genus level where feasible but some could only be allocated to a broader group (e.g., 'spiders'). All invertebrates were then ranked on a scale of abundance according to the total number of individuals collected, and the number of sites at which they were present. They were classed as rare, occasional, common or abundant according to the definitions below:

- **rare**: fewer than 5 individuals collected;
- **occasional**: 5–24 individuals collected,
  or >24 individuals but present at fewer than five sites;
- **common**: 25+ individuals collected **and/or** present at five or more sites;
- **abundant**: 100+ individuals collected **and** present at 10 or more sites.

Specimens of all invertebrates identified were lodged with the New Zealand Arthropod Collection (NZAC), Tamaki, Auckland.

5. Results and Discussion

5.1 Fungi

Many diseased plants were observed at sites where they were seemingly moisture stressed (e.g., wetter areas – Cheviot; drier areas – Brancott Station). *B. glaucocarpa* appeared to be more diseased than *B. darwinii*.

Fungal colonisation ([total number of fungal isolates divided by the number of tissue fragments] x 100) of *B. glaucocarpa, B. darwinii* and *B. vulgaris* averaged across all tissue types collected during our survey was found to be 95.7, 98.4 and 58.4 % respectively (Table 2). There was similarly high colonisation of fruit fragments on both *B. glaucocarpa* and *B. darwinii*, with several tissues, particularly the berry fruit, often having more than one fungus present. Fungal colonisation was also higher in the leaf and flower tissues of *B. darwinii* than in *B. glaucocarpa* (Table 2). Although a low colonisation was observed for *B. vulgaris* only one sample of leaves was plated, so further plating would be required to determine a more representative sample of the fungi present on this species. Across all barberry tissues plated, the mean fungal colonisation was 95.4%, which is relatively higher than that recorded from two previous weed surveys (moth plant, *Araujia sericifera,* 68%; tradescantia, *Tradescantia fluminensis,* 62%) but is similar to that observed for woolly nightshade, *Solanum mauritianum* (92%); nassella tussock, *Nassella trichotoma* (95%); and Chilean needle grass, *Nassella neesiana* (98%) (Winks et al. 2004).

Table 2 Mean fungal colonisation (%) of diseased barberry tissue fragments.

Host	Mean fungal colonisation			Total
	Leaf	Flower	Fruit	
B. glaucocarpa	88.9	91.4	115.6	95.7
B. darwinii	92.2	111.1	115.0	98.4
B. vulgaris	58.4	-	-	58.4
Total	88.7	98.1	115.5	95.4

A total of 508 fungal isolates were obtained from a total of 532 barberry tissues plated (Table 3). As most of the tissues plated were from leaf spots on *B. glaucocarpa* (261 pieces), the greatest numbers of fungi were correspondingly also from *B. glaucocarpa*. A total of 52 fungi were obtained from flower tissues and 134 fungi from barberry fruit (Table 3). Most fungi (375) were isolated from *B. glaucocarpa*, reflecting the higher number of samples collected for that host. *B. glaucocarpa* tissues also appeared to be more susceptible to infection by fungi than *B. darwinii*, a result that was supported by field observations.

Table 3 Mean fungal colonisation (%) of diseased barberry tissue fragments with the number of fragments plated in parentheses.

Host	Total fungi isolated			Total
	Leaf	Flower	Fruit	
B. glaucocarpa	232 (261)	32 (35)	111 (96)	375 (392)
B. darwinii	83 (90)	20 (18)	23 (20)	126 (128)
B. vulgaris	7 (12)	-	-	7 (12)
Total	322 (363)	52 (53)	134 (116)	508 (532)
Total mean % colonisation	88.7	98.1	115.5	95.5

A total of 21 fungal species were identified on isolation plates from the three barberry species: 19 from *B. glaucocarpa*, 14 from *B. darwinii*, and 2 from *B. vulgaris*. A further three species were identified separately from plant tissues: a barberry rust, *Puccinia graminis* was found on *B. glaucocarpa* flowers; and *Aspergillus niger* and a species of *Rhizopus* were directly observed sporulating on wizened *B. glaucocarpa* berries. Of the 24 species in total, only five species are likely to be potential pathogens of *Berberis* spp: *Colletotrichum acutatum*, *Colletotrichum gloeosporioides, Pestalotiopsis* sp., *Phomopsis* sp., and *Sclerotinia sclerotiorum* (Table 4). The remaining species are predominantly saprophytic or secondary/opportunistic pathogens, probably infecting the host after damage, insect feeding or infection by a primary pathogen, and are therefore unlikely to be sufficiently virulent and/or host specific to be useful as biocontrol agents (Table 5). Some of these species are also known to produce mycotoxins (Table 5), further reducing their potential as biocontrol agents.

Table 4 Relative abundance of pathogens collected from three *Berberis* species throughout New Zealand 2003–04

Species:	**Specimen no: ***	**Total no. sites (isolates)**	**Affected tissues, disease symptoms & previous host records**
Colletotrichum gloeosporioides	94, 95, 96, 98, 103, 104, 110, 127, 128, 129, 130, 131, 134.	13 (128)	**Young Leaves:** Isolated from black lesions that were variable in shape and size. Together, many of these lesions often appeared as a black-red freckled mottle on the leaves. **Mature Leaves:** Isolated from many different irregular shaped blotches and lesions variable in size and colour. On *B. glaucocarpa* the lesions were bright red-wine red brown in colour. On *B. darwinii* the lesions could be red in colour with a yellowish margin. Occurred on leaves of both species at most of the collection sites **Flowers:** Isolated from black-brown dieback on petioles of the flower inflorescence. Also isolated from black petioles and buds of the flower inflorescence where orange spore masses were observed within some lesions. **Fruit:** Isolated from shriveled blackened berries. Also from discoloured grey lesions on the berry stalks.
Colletotrichum acutatum	99, 126, 128, 129, 135, 136, 137, 138, 139.	9 (57)	**Leaves:** Isolated from lesions that were variable in colour, shape and size. Black–dark brown– purple–red lesions of circular to irregular shape and size. Lesions were also visible on the underside of the leaf.

Species:	Specimen no: *	Total no. sites (isolates)	Affected tissues, disease symptoms & previous host records
			Fruit: Isolated from blackish tissues on ripe fruit Both these *Colletotrichum* species can cause serious 'anthracnose' diseases on numerous plant species in NZ (Young & Fletcher 1997; Pennycook 1989), but can also be saprophytic on the plant tissues of many hosts. Pathogenic *Colletotrichum* isolates have successfully been used as biocontrol agents against a variety of environmental and agricultural weedy targets.
Pestalotiopsis sp*. * isolated from *B. darwinii* only	97, 114, 118, 125, 133.	5 (14)	**Leaves:** Isolated from reddish-brown lesions on the leaf edge. The lesions were surrounded by a yellowish chlorotic tissue that spread from the leaf edge into the middle of the leaf. Different species in the genus *Pestalotiopsis* are significant and primary pathogens that cause a range of serious diseases of a number of crop, ornamental and tree species (Farr et al. 2004). In New Zealand, this genus has been isolated from many different introduced and native plant species (Pennycook 1989).
*Sclerotinia sclerotiorum** *isolated from *B. darwinii* only	122	1 (2)	Symptoms appeared as small circular black lesions (2–5 mm) on unripe and ripening berries. *Sclerotinia sclerotiorum*, also known as white soft rot or white mould, is a virulent primary pathogen of many horticultural crop species with a very wide host range in NZ (Pennycook 1989) and an even wider range worldwide (Farr et al. 2004). A major research initiative is currently underway to develop this pathogen as a mycoherbicide against a number of agricultural and environmental weeds in New Zealand.
Phomopsis sp. (1-2 species isolated)	96, 98, 103, 107, 127, 128, 129, 130, 131, 137, 138, 140.	12 (64)	**Leaves:** Isolated lesions were variable in colour, shape and size. Dark brown–wine red–purple–bright red lesions of circular to irregular shape and size (1–10 mm). Lesions were also sometimes visible on the underside of the leaf. **Flower:** Appeared as blackened tissue in flower buds and flower petioles. **Fruit:** Isolated from ripening and ripe fruit that had blackened and had a withered appearance. Also isolated from purplish lesions on the fruit of *B. darwinii*. *Phomopsis* species are widespread saprophytes on plant tissues but some species can also be widely distributed primary pathogens of some plant species (Young & Fletcher 1997; Pennycook 1989; Farr et al. 1989; Farr et al. 2004).

* Specimen numbers relate to sites listed in Appendix 1

Colletotrichum spp. were also the most commonly isolated fungi across all tissues, with *C. gloeosporioides* being the most frequently isolated species (128 isolates) (Table 5). *Phoma, Phomopsis* and other unidentified Coelomycete fungi were also frequently isolated in the survey (Table 5). *Alternaria alternata* was the commonest secondary pathogen isolated in the survey with 33 isolates being recovered, mainly from leaf tissues. The remaining species were rare or occasionally isolated, with low frequency.

Table 5 Number of fungal isolates from *Berberis* species collected at 40 New Zealand sites during 2003/2004.

	B. glaucocarpa				***B. darwinii***				***B. vulgaris***	
Fungi	**Leaf**	**Flower**	**Fruit**	Total	**Leaf**	**Flower**	**Fruit**	Total	**Leaf**	**Total**
Acremonium spp. (n* = 2)	2	2	3	7	0	0	0	0	1	**8**
Alternaria alternata	11	1	7	19	9	3	2	14	0	**33**
Aureobasidium pullulans	3	2	9	14	4	3	2	9	0	**23**
Botrytis cinerea	3	2	1	6	1	2	2	5	0	**11**
Cladosporium spp. (2)	4	0	2	6	3	0	2	5	0	**11**
Coelomycete spp. (>4)	26	2	2	30	1	0	0	1	0	**31**
Colletotrichum cf. *acutatum*	41	0	14	55	2	0	0	2	0	**57**
Colletotrichum gloeosporioides	67	12	41	120	1	5	2	8	0	**128**
Diaporthe sp.	0	0	0	0	0	0	0	0	4	**4**
Epicoccum purpurascens	2	0	1	3	0	0	0	0	0	**3**
Fusarium spp. (4)	8	1	1	10	8	0	2	10	0	**20**
Fusicoccum sp.	1	0	3	4	1	0	0	1	0	**5**
Penicillium sp.	0	1	0	1	0	0	0	0	0	**1**
Pestalotiopsis sp.	0	0	0	0	13	1	0	14	0	**14**
Phoma spp. (>3)	32	6	9	47	10	0	9	19	0	**66**
Phomopsis sp.	16	3	17	36	23	5	0	28	0	**64**
Sclerotinia sclerotiorum	0	0	0	0	0	0	2	2	0	**2**
Stemphylium sp.	1	0	0	1	4	0	0	4	0	**5**
Sterile Dark	7	0	1	8	0	0	0	0	0	**8**
Sterile Hyaline	2	0	0	2	2	0	0	2	0	**4**
Unidentified	5	0	0	5	1	0	0	1	2	**8**
Yeast spp. (2)	1	0	0	1	0	1	0	1	0	**2**
Total fungi	**232**	**32**	**111**	**375**	**83**	**20**	**23**	**126**	**7**	**508**

* (n) = number of different species isolated

Excised plant tissues incubated in humidity chambers demonstrated that *Botrytis* infection caused significant disease of barberry plant tissues (Table 6) and was able to cause complete bunch rot of fruit and flower inflorescences. Although *Botrytis* was observed to be uncommon using culturing methods (Table 5), results from the humidity chambers showed the fungus was present and able to cause latent infections of all barberry tissues (Table 6).

Table 6 Number of samples with *Botrytis* & *Colletotrichum* sporulation present from diseased Barberry tissues.

	Botrytis & *Colletotrichum* sporulation						**Total**	
	Leaf		**Flower**		**Fruit**			
Host:	Bot	Col	Bot	Col	Bot	Col	Bot	Col
B. glaucocarpa	8 [A2]	2	4	1	4 [A1]	2	16	5
B. darwinii	-	-	2	-	1	1	3	1
Total	8	2	6	1	5	2	19	6

Bot = *Botrytis cinerea* / C = *Colletotrichum*/ A = *Alternaria* sporulation also observed (n = number of samples with *Alternaria*)

Barberry Rust

Orange pustules of a rust fungus were found on a few flowers of *B. glaucocarpa* collected from Brancott Station in November 2003 (Fig. 3). The rust was identified as *Puccinia graminis* using DNA sequencing techniques.

Fig. 3 *Puccinia graminis* pustules on flower petioles of *B. glaucocarpa*

The record of *P. graminis* on the flowers of *B. glaucocarpa* in New Zealand is very interesting scientifically. It represents a new host/pathogen record for this country and may also be the first time that the rust has been found on the flowers (normally found on leaves) of any *Berberis* species (C. Aime pers. comm).

The host specificity of most rusts mean they are often regarded as good candidates for use in classical biocontrol projects. However, for a variety of reasons, it will not be sensible to investigate using this rust as a biocontrol agent for barberry in New Zealand. The rust has a complicated life cycle (referred to as heteroecious), which means different life stages of the rust occur on different (and unrelated) host plants. Two of the life stages (uredinial and telial) of *P. graminis* are common in New Zealand on a broad range of grasses and cereals of economic importance (Pennycook 1989). This is the first time the additional life stage (aecial) has been found in New Zealand. This is significant because the aecial stage is common overseas on *Berberis* species, and is an integral part of the lifecycle in North America and Europe (McGregor 2002). Thus, *Berberis* species are the alternate hosts for *P. graminis* and provide an over-wintering reservoir for infection of cereals and grasses in the ensuing spring - this may be another good reason for controlling *Berberis* species. In addition, aecia are not a repeating stage in the life cycle. This means that aeciospores from the pustules found on *B. glaucocarpa* could only infect a cereal or grass host and would not spread directly to other *B. glaucocarpa* plants. In New Zealand, the rust also appears to be rare on *Berberis* spp. and causes limited disease symptoms on this host. In our nationwide survey this was the only specimen collected, and it was not recovered from the same site when it was revisited 4 months later.

Epiphytic colonisation on Darwin's barberry

Orange/red circular growths were observed on leaves of 8 samples of Darwin's barberry (Figure 4), in two locations (Barberry Rd and Mouats Saddle, Catlins). All leaves collected were covered in these growths, most probably caused by a very common alga, *Cephaleuros*, growing on the leaf surface (Shaun Pennycook, pers comm.). A leaf specimen was not available at the time of identification, so this conclusion is based on photographic evidence.

On further examination after scraping the orange/red spots from the leaf surface, it was found the leaves were still intact and the epidermis appeared unaffected. Further research would be required to ascertain if these have any impact on the host through possible reduction of direct photosynthesis due to these growths obscuring the leaf surfaces, but this was beyond the scope of the present study. An additional and different yellow species of lichen or alga was also present on the leaves of barberry plants sampled from Manapouri. This could not be identified. Again there was no apparent damage to the host leaf tissue.

Fig. 4 Colonisation of *B. darwinii* leaves probably caused by a common alga (*Cephaleuros*).

5.2 Invertebrates

A full list of phytophagous invertebrates found to be associated with the three species of barberry sampled is presented in Table 7; all other invertebrates collected (predators, parasitoids, saprophytes, etc.) are listed in Appendix 1.

Herbivores

Across the three *Berberis* species 101 herbivorous invertebrate species were identified, with another 12 taxonomically related groups of herbivorous species recorded for which identification to genus or species level was not feasible, e.g., unidentified snail. 108 species or groups were recorded from *B. glaucocarpa*, 25 species from *B. darwinii*, and 2 species from *B. vulgaris* (Table 7). This represents 40 herbivorous families collected from *B. glaucocarpa*, 16 families from *B. darwinii*, and 2 from *B. vulgaris*.

Table 7 Relative frequencies of phytophagous invertebrates associated with *Berberis* species through out New Zealand during 2003 and 2004.

Insect species	Common name	Feeding site	Origin	Site	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Phylum NEMATODA	**nematodes**						
unid. Nematoda		herbivorous		3	R		
Phylum MOLLUSCA	**molluscs**						
Class GASTROPODA	**slugs and snails**						
Helix aspersa Müller	brown garden snail	herbivorous	introduced	3,11	R		
unid. Snails		herbivorous		1,3,4,32,36,37,39	C	O	
unid. Slugs		herbivorous		10	R		
Phylum ARTHROPODA							
Class HEXAPODA							
INSECTA	**insects**						
COLEOPTERA	**beetles**						
Anobiidae	**borer beetles**						
Sphinditeles sp.		herbivorous	native	26	R		
Brentidae							
Exapion ulicis (Forster)	gorse seed weevil	herbivorous	introduced BCA	4,5,14,31	R	R	
Cerambycidae	**longhorn beetles**						
Coptomma sulcatum (F.)		herbivorous	native	27	R		
Hybolasius sp.		herbivorous	native	19,27	R		
Oemona hirta (F.)	lemon tree borer	herbivorous	native	12	R		
Psilocnaeia sp.		herbivorous	native	1,5,14,15,17,22,24,26,27,31,39	C	R	
Xyloteles sp.		herbivorous	native	4,10	O		
Xylotoloides huttoni (Sharpe)		herbivorous	native	17	R		
Zorion sp.		herbivorous	native	14,15,16,18,19,22,37	O	R	

Insect species	Common name	Feeding site	Origin	Site	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Chrysomelidae	**leaf beetles**						
Adoxia sp. 1		herbivorous	native	25	R		
Adoxia sp. 1		herbivorous	native	13	R		
Bruchidius villosus (F.)		herbivorous	introduced BCA	26	O		
Eucolaspis sp.		herbivorous	native	14,17,25	R		
Trachytetra rugulosa (Broun)		herbivorous	native	13	R		
Curculionidae	**weevils**						
Andracalles sp.		herbivorous	native	13	R		
Asynonychus cervinus (Boheman)	Fuller's rose weevil	herbivorous	introduced	1,3,4,5,6,10,11,15	C		
Catoptes sp. 1		herbivorous	native	39		R	
Catoptes sp. 2		herbivorous	native	18	R		
Didymus sp.		herbivorous	native	14,15,17	O		
Gymnetron pascuorum (Gyllenhal)		herbivorous	introduced	4	R		
Irenimus sp.		herbivorous	native	39		R	
Microcryptorhynchus sp.		herbivorous	native	1,5,12,14,17	O		
Pactolotypus sp.		herbivorous	native	39		R	
Peristoreus sp. 1		herbivorous	native	26	R		
Peristoreus sp. 2		larvae and adults in dead wood	native	14	R		
Phloeophagosoma sp.		herbivorous	native	8	R		
Phlyctinus callosus Boheman	garden weevil	herbivorous	introduced	1,9	R		
Psepholax sp.		larvae / adults in dead wood?		39		R	
Praolepra sp.		herbivorous	native	16	R		
Rhopalomerus sp.		pollen feeder	native	12,18,19,27,33,37	O	R	
Sericotrogus subaenescens Wollaston		larvae and adults in dead wood	native	2,14	R		

Insect species	Common name	Feeding site	Origin	Site	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Sitona lepidus Gyllenhal		herbivorous	introduced	9	R		
Synacalles sp.		herbivorous	native	15	R		
Dermestidae							
Trogoderma signatum Sharpe					R		
Elatridae	**click beetles**						
Conoderus exsul (Sharp)	pasture wireworm	herbivorous	introduced?	3,5,6	O		
Ctnicera sp.			native	15	R		
Scarabaeidae							
Costelytra zealandica (White)	grass grub	herbivorous	native	7,9	R		
Pyronota sp. 1		herbivorous	native	19	R		
Pyronota sp. 2		herbivorous	native	23	R		
Scraptiidae							
Nothotelus sp.		herbivorous	native	17,22,23,25,27,34	C	R	
Tenebrionidae	**darkling beetles**						
Artystona rugiceps Bates		herbivorous	native	14,24	O		
Zopheridae	**false darkling beetles**						
Bitoma rugosa Sharp		herbivorous	native	9,14,17	C		
HEMIPTERA	**bugs**						
Acanthosomatidae							
Oncacontias vittatus (Fabricius)		sap feeder	native	7,12,18,20,27,32,37,39	O	O	
Achilidae							
Agandecea annectens White		sap feeder	native	23	R		
Anthocoridae							
unid. Anthocoridae		sap feeder	native	1,9,15,20,30,31,39	O	O	
Aphididae	**aphids**						
Acyrthosiphon kondoi Shinji	blue-green lucerne aphid	sap feeder	introduced	2	R		

Insect species	Common name	Feeding site	Origin	Site	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Acyrthosiphon pisum (Harris)	pea aphid	sap feeder	introduced	17,23,24	O		
Aulacorthum solani (Kaltenbach)	foxglove aphid	sap feeder	introduced	1,12,15,16,22	O		
Brevicoryne brassicae (L.)	cabbage aphid	sap feeder	introduced	17,24	O		
Capitophorus elaeagni (Del Guer.)	thistle aphid	sap feeder	introduced	23,24	O		
Cavariella aegopodii (Scop.)	carrot aphid	sap feeder	introduced	23,24,43	R		R
Drepanosiphum platanoidis (Schrank)	sycamore aphid	sap feeder	introduced	25,27	O		
Liosomaphis berberidis (Kaltenbach)	barberry aphid	sap feeder	introduced	7,8,12,13,14,15,16,18,19,22,25,26,38,39	A	C	
Lipaphis pseudobrassicae (Davis)		sap feeder	introduced	15	R		
Macrosiphon euphorbiae (Thomas)	potato aphid	sap feeder	introduced	15,19,23,24	O		
Rhopalosiphum padi (L.)	cereal aphid	sap feeder	introduced	13,23,24	O		
Sitobion fragariae (Walker)	grain aphid	sap feeder	introduced	17,23,24	O		
Cercopidae	**spittle bugs**						
Carystoterpa fingens (Walker)		sap feeder	native	12	R		
Carystoterpa vagans Hamilton and Morales		sap feeder	native	18	R		
Philaenus spumarius (L.)	meadow spittle bug	sap feeder	introduced	9,12,14,15,18,20,22,31	C	R	
unid. Cercopidae				14,15,23	O		
Cicadellidae	**leafhoppers**						
Zelopsis nothofagi Evans		sap feeder	native	16	R		
Xestocephalus ovalis Evans		sap feeder	native	31		R	
Cicadidae	**cicadas**						
Kikihia sp.		sap feeder	native	9	R		
Cixiidae							
Oliarus oppositus (Walker)		sap feeder	native	9,11	O		
Koroana rufifrons (Walker)		sap feeder	native	3,7	R		
Koroana sp.		sap feeder	native	18,19	O		

Insect species	Common name	Feeding site	Origin	Site	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Coccidae	**soft scales**						
Saissetia coffeae (Walker)	hemispherical scale	sap feeder	introduced	8,15	R		
Saissetia oleae (Olivier)	olive scale	sap feeder	introduced	8,17	R		
Flatidae	**planthoppers**						
Anzora unicolor (Walker)	grey planthopper	sap feeder	introduced	5,14,15,17	C		
Siphanta acuta (Walker)	green planthopper	sap feeder	introduced	5,7,14,15,16,17,18	C		
Lygaeidae	**seed bugs**						
Rhypodes sp.		sap/seed feeder	native	7,17	R		
Margarodidae							
Coelostomidia sp.		sap feeder		14	R		
Icerya purchasi Maskell	cottony cushion scale	sap feeder	introduced	7,14,17	O		
Miridae	**mirid bugs**						
Chinamiris sp.		sap feeder	native	7,19,37	R	R	
Diomocoris ostiolum Eyles		sap feeder	native	7,12,25	O		
Romna sp.				7,8,10	O		
Xiphoides myersi (Woodward)			native	12,15,26	O		
unid. Miridae				15,16,22,24,27	O		
Pentatomidae	**shield bugs**						
Cuspicona simplex Walker	green potato bug	sap feeder	introduced	2,7,18,31	R	R	
Nezara viridula (L.)	green vegetable bug	sap feeder	introduced	2,7,31	R	R	
unid. pentatomid egg- batches				38		R	
Pseudococcidae	**mealybugs**						
unid. Pseudococcidae		sap feeder		14	O		
Ricaniidae	**planthoppers**						
Scolypopa australis (Walker)	passionvine hopper	sap feeder	introduced	3,4,5,6,8,10,14,15	C		

Insect species	Common name	Feeding site	Origin	Site	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
LEPIDOPTERA	**moths and butterflies**						
(collected as larvae and reared to adult for identification)							
Geometridae	**looper moths**						
Chloroclystis filata (Guenée)	Australian pug	herbivorous	native	15	R		
Declana floccosa Walker	forest semilooper	herbivorous	native	15	R		
Pseudocoremia suavis Butler	common forest looper	herbivorous	native	8,15,36,37	R	R	
unid. Geometridae		herbivorous		7,8,26,30,36	O	O	
Noctuidae	**armyworms, cutworms**						
unid. Noctuidae		herbivorous		15	R		
Psychidae	**bag moths**						
Liothula omnivora (Fereday)	common bag moth	herbivorous	native	5,8,15,18,24,25,38	O	R	
Scoriodyta sp.		herbivorous	native	18	R		
undescribed sp.	little log cabin bag moth	herbivorous – algal feeder		4	O		
Tortricidae	**leafrollers**						
Cnephasia jactatana Walker	black-lyre moth	Herbivorous	native	3	R		
Ctenopseustis obliquana (Walker)	brown-headed	Herbivorous	native	2,3,4,5,6,7,9,11,30,31,32	C	O	
Ctenopseustis obliquana or *C. herana*	leafroller	Herbivorous		12,13,14,15,16,17,18,19,22,24,26,27,34, 36,37,38,39,43	C	O	R
Epalxiphora axenana Meyrick	sharp-tipped bell moth	Herbivorous	native	3,18	R		
Epiphyas postvittana (Walker)	light-brown apple moth	Herbivorous	introduced	5,6,10,11	O		
Harmologa amplexana (Zeller)		Herbivorous	native	19	R		
Harmologa scoliastis Meyrick		Herbivorous	native	11	R		
Planotortrix sp.		Herbivorous	native	9,31,32	O	R	
Planotortrix notophaea Turner		Herbivorous	native	18,31,32,36	R	O	
Pyrgotis plagiatana Walker	painted wedge	Herbivorous	native	12,31,40	R	R	
unid. Tortricidae		Herbivorous		1,3,4,5,6,7,8,9,10,11,15,24,30,31,32,38, 39	C	C	

Insect species	Common name	Feeding site	Origin	Site	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
ORTHOPTERA	**crickets and grasshoppers**						
Anostostomatidae							
Hemideina crassidens (Blanchard)	Wellington tree weta	Omnivorous	native	18	R		
Hemideina thoracica White	Auckland tree weta	Omnivorous	native	5,6	R		
Raphidophoridae	**cave weta**						
unid. Raphidophoridae		Omnivorous		17,34	R	R	
Tettigoniidae	**long-horned grasshoppers**						
Caedicia simplex (Walker)	katydid	Herbivorous	native	3,5,7,8,10,11	O		
Conocephalus sp.	field grasshopper	Herbivorous	native	11,15,22	R		
Phasmatodea	**stick insects**						
Clitarchus hookeri (White)	smooth stick insect	Herbivorous	native	11	R		
THYSANOPERA	**thrips**						
Sub-Order Terebrantia							
Thripidae							
Thrips obscuratus (Crawford)	New Zealand flower thrips	Herbivorous	native	6,22,23	C		

NOTES:

BCA: biological control agent

refer to Fig 1 and Fig. 2 for site numbers

R: rare

O: occasional

C: common

A: abundant

Only one herbivorous species, the barberry aphid *Liosomaphis berberidis* (Aphididae), was classified as 'abundant' on *B. glaucocarpa*. A further 13 herbivorous species or taxonomic groupings were classed as 'common', 33 species were classed as 'occasional' and 62 as 'rare' on *B. glaucocarpa*. No herbivorous species were 'abundant' on *B. darwinii*, although the barberry aphid was one of two species classified as 'common', with 6 species classed as 'occasional'.

Foliage feeders: In general, foliar damage that could be attributed to invertebrate herbivory was minor. It was unusual to find a leaf that was greater than 10% consumed, and the overall amount of foliage that appeared to have been consumed or damaged by herbivores was estimated to be less than 5% at most sites.

The most obvious foliar damage appeared to be caused by the larvae of a range of moth species, especially leafroller (Tortricidae) larvae, one of the most well-represented herbivorous families, mostly native species, in this survey. Leafroller larvae were sometimes found still inside 'rolled' leaves on the plant, but more commonly they were collected from the beating tray after being dislodged from the plant. The brown-headed leafroller (*Ctenopseustis obliquana)*, a common horticultural pest with a wide range host range, was the most common moth species collected. A number of moth larvae, collected to rear through to adult for identification, died during rearing, and parasitoids emerged from some of them (listed in Appendix 1).

Beetles (Coleoptera) in general, and weevils (Curculionidae) in particular, were well represented in collections, with 39 herbivorous species of beetles, 16 of which were weevils, collected from *B. glaucocarpa* sites, most with broad host ranges. Only 9 coleopteran species were collected from *B. darwinii*. Judging by the total number of herbivorous beetles collected at each site and the fact that many of them were very small, the combined effect of herbivorous beetles would probably be minimal at most sites. However, at three *B. glaucocarpa* sites (Maramarua, Stonehurst Farm, and Tukituki Rd) the relatively large Fuller's rose weevil, *Asynonychus cervinus*, appeared to be quite numerous (classified as "common") and may have been consuming appreciable amounts of foliage. This introduced weevil has a wide host range, attacking citrus, kiwifruit, roses and many flowering plants through out New Zealand (Scott 1984).

Sap feeders: The sap-feeding barberry aphid, *Liosomaphis berberidis* (Aphididae), was the only invertebrate found during the survey to be classed as 'abundant'. The numbers of this aphid varied greatly between sites but it was widespread on both *B. darwinii* and *B. glaucocarpa* even though previously only being recorded from *B. glaucocarpa* (Dale & Maddison 1982). Some sites had very heavy infestations with noticeable amounts of sticky "honeydew" being produced, whereas at other sites few or none were collected. This could be related to parasitism and predation with outbreaks occurring during a temporary escape from parasites and predators. The barberry aphid is also found on *Mahonia* species (e.g., Oregon grape), another genus in the family Berberidaceae. Although other *Aphis* species were present, often in high numbers (e.g., *Drepanosiphum platanoidis* was classed as 'occasional' on *B. glaucocarpa)*, this may not be indicative of potential damage caused by such species as many are known to be host specific on other plant species. *D. platanoidis* feeds on sycamore, so these aphids are only present on *Berberis* sp. due to locally specific conditions, i.e. over-hanging sycamore trees.

The passion vine hopper, *Scolypopa australis* (Ricaniidae), was common on *B. glaucocarpa* in the North Island and northern South Island. For climatic reasons this accidentally introduced Australian species does not occur further south. A total of 36 species of sap-feeders were found during the survey on all *Berberis* species. The damage caused by sap-feeders, either directly by the removal of nutrients, or indirectly by puncturing the plant and possibly allowing entry of pathogens, is very difficult to quantify.

Stem borers: Longhorn beetle (Cerambycidae) adults of a number of species were commonly found on barberry, with 7 species on *B. glaucocarpa* and 2 species on *B. darwinii*. Longhorn beetle larvae are woodborers, often into dead wood, and their tunnels were commonly found in barberry branches (e.g., *Psilocnaeia* sp.), especially in older plants. Other invertebrates, including weta, earwigs, slaters and cockroaches, also inhabited many of the tunnels created by these beetles.

Seed feeders: Fruit or seed was collected from *B. darwinii* at 7 sites (Rotorua, Catlins Valley Road, Thompson's Bush, Manapouri, Kaitangata, Mouats Saddle, Tahakopa), and from *B. glaucocarpa* at 11sites (Lake Rerewhakaaitu, Rangiriri, Maramarua, Puhoi, Kawakawa Bay, Featherstone, Tukituki Road, Deadman's Island, Brancott Station, Stonehurst Farm, Kurow). No dedicated seed feeders were reared from the seed collections made. All invertebrates that emerged were generalist plant feeders, feeding on the fruit and only occasionally feeding on the seed: from *B. glaucocarpa* at Stonehurst Farm one leafroller (Tortricidae) larva was collected and 1 katydid nymph (*Caedicia simplex*); from Deadman's Island and Kurow one leafroller (Tortricidae) larva was collected from each site. From *B. darwinii* at Kaitangata 20 shield bug (Pentatomidae) nymphs emerged from an egg cluster amongst the seeds. All other sites sampled yielded only small numbers of Lathridiidae (mildew beetles) and Psocoptera (book lice), both fungal feeders, or no insects at all.

Little or no seed/fruit damage was observed at field sites also. Some leafroller (Tortricidae) feeding damage to fruits was observed at some sites, e.g., Deadman's Island and Kurow, but this was minor. Some external damage to the fleshy fruits was observed at Kaitangata but this was probably attributable to small bird (e.g., wax eye) feeding damage. However, the most important means of dispersal of *Berberis* species is probably through bird feeding.

Non-herbivorous invertebrates

Predatory, parasitic, saprophytic and fungivorous species (Appendix 1) may inhibit the establishment and dispersal of any biological control agents that may be introduced and so provides a valuable reference list when prioritising possible biological control agents. In total, 61 non-herbivorous species and 23 larger taxonomic groups were collected from the 3 barberry species. These were subdivided into feeding groups: fungivorous (26); predatory (18); parasitoid (12); omnivorous/algal feeders (9); and saprophytic (8). By far the largest group was the spiders (Araneida), possibly the most numerous group at all sites, and was classed as 'abundant' on both *B. glaucocarpa* and *B. darwinii*. The pollen/fungus feeding beetles in the family Cryptophagidae were the only other 'abundant' group with 15 other species/groups classed as 'common'.

6. Conclusions

Barberry was locally widespread and out of control at only a few of the sites visited. Sites in or neighbouring extensive native bush (e.g., Manapouri and Whisky Gully) offer the greatest immediate threat compared with manicured hedgerows bordering extensive grazed pasture. Hedged barberry remains controlled in size and spread with regular trimming, with both branching and seed production being limited. However, these species, once feral, are capable of moving into new territory, and with their tolerance of growing under canopy, both exotic and native, they represent a significant threat to bush and pasture alike (especially where grazing pressure is low).

Prospects appear good for a biological control programme directed at *B. darwinii* and *B. glaucocarpa*. Several fungi and invertebrate species are known to attack Berberis species in their native range (McGregor 2002) and some of these cause significant damage to the plant. Also, since *Berberis* is a large genus, the prospects for locating further potential biological control agents are good. Overseas, *Lanzia parasitica* has caused significant damage to *B. vulgaris*, while the cecidomyiid midge *Lasioptera* sp. and the sawfly *Arge berberidis* damage some *Berberis* spp. This suggests the potential for introducing agents that would reduce the vigour and production of viable seeds of barberry species in New Zealand is good. Other fungi and invertebrates also attack the plant, but the extent of damage they cause is not clear; nevertheless, the pool of potential agents is likely to be large, particularly in light of the genus' wide distribution.

There are no indigenous Berberidaceae in New Zealand and no commercially important exotic members of the family, with only a small number of ornamentals in the genus *Mahonia,* so prospective agents need not be restricted to attacking only the adventive species of *Berberis.* Several fungi and invertebrate species are known to specifically attack *Berberis* species and some of these cause significant damage to the plant.

6.1 Fungi

A high level of fungal colonisation was observed on both *B. glaucocarpa* and *B. darwinii*, as both species exhibited a wide range of disease symptoms on leaf, flower and fruit tissues. Disease damage on *B. glaucocarpa* was often widespread, particularly in areas where the plant was also experiencing water stress. As the disease symptoms were observed on both young leaves and reproductive tissues (flowers and fruit), these pathogens could significantly affect the growth and spread of the weed in some areas. Approximately 21 different fungal species were recovered on plates and a further 3 by direct methods. A number of fungal isolates were cosmopolitan saprophytes or secondary-weak pathogens and therefore are unlikely to have potential as either classical biocontrol agents or inundative mycoherbicides as they lack the required virulence to be considered as agents. However, five species of primary plant pathogens associated with leaf, flower and/or fruit disease symptoms, *Colletotrichum gloeosporioides, C. acutatum, Pestalotiopsis* sp., *Phomopsis* sp., and *Sclerotinia sclerotiorum*, may warrant further investigation.

Colletotrichum gloeosporioides was the predominant plant pathogen recorded in the survey and was also found consistently associated with the most severe symptoms recorded on barberry tissues. In New Zealand, *Colletotrichum gloeosporioides* is a common inhabitant of many plant species (di Menna & Parle 1970; Pennycook 1989; Young & Fletcher 1997). While *C. gloeosporioides* is widespread and recorded from many different hosts, host specific strains have been identified and developed as both classical and inundative biocontrol agents against a number of environmental and agricultural weed species. The first plant pathogen to be registered as a bioherbicide was a virulent strain of *C. gloeosporioides* (f. sp. *malvae*) for control of Malvaceae weed species such as round-leaved mallow (*Malva pusilla*) (Morin et al. 1996; Mortensen & Makowski 1992). In Canada, *Colletotrichum gloeosporioides* (f. sp. *hypericum*) is a highly virulent disease that infects all aerial parts of St John's Wort (*Hypericum perforatum*), causing lethal stem girdling lesions on seedlings and on stems of the basal rosette (Morrison et al. 1998). Hawaiian researchers have also introduced a strain of the fungus to Hawai'i and French Polynesia, resulting in the successful biocontrol of *Miconia calvescens* in both places (Killgore et al. 1997; Killgore 2004 pers. comm.). Additional weed biocontrol programmes using such *C. gloeosporioides* strains are underway currently for dodder (*Cuscuta australis*), northern jointvetch (*Aeschynomene virginica*), dwarf mistletoe (*Arceuthobium tsugense*), and *Hakea sericea* (Van Rooj & Wood 2003).

Sclerotinia sclerotiorum, although reported to cause many economically significant disease losses to crops, has also been formulated as an inundative mycoherbicide for the control of giant buttercup, *Ranunculus acris* (Verkaaik et al. 2004) and Californian thistle, *Cirsium arvense* (Hurrell et al. 2001; Harvey et al. 1994) in New Zealand. This fungus is also capable of causing disease to a number of other weed species such as ragwort, *Senecio jacobaea*, in New Zealand (Waipara et al. 1993). Although this fungus is plurivorous (unspecialised pathogen with a very wide host range), there is often minimal risk to non-target hosts when using this as mycoherbicide in non-cropping systems (Bourdot et al. 2001). Given recent developments in the formulation technology and risk assessments of using this fungus for weed biocontrol, the prospects of using *Sclerotinia* based products against barberry may be warranted if primary pathogenicity is confirmed at a later stage in the project.

Phomopsis species were also associated with minor shoot damage on gorse and broom (Johnston et al. 1995); however, no biocontrol potential for these isolates was observed during glasshouse inoculation trials (Johnston & Parkes 1994). *Phomopsis* species have a wide host range across other plant species (Pennycook 1989; Young & Fletcher 1997), with many associated with pre- and post-harvest diseases on a variety of crops such as curcubits, grapes and strawberries (Pennycook 1989; Farr et al. 2004).

To date there are no or few examples of using strains or species of either *C. acutatum, Pestalotiopsis* or *Phomopsis* for weed biocontrol, although *Phomopsis amaranthicola* is under development as a post-emergence bioherbicide against pigweeds (*Amaranthus* spp.) in Florida, USA (Morales-Payan et al. 2003). Further research is needed to elucidate the prospects of *C. acutatum* or *Pestalotiopsis,* as no successful overseas examples currently exist for developing these fungi as biocontrol agents.

The most promising species isolated were *Colletotrichum gloeosporioides*, *Sclerotinia sclerotiorum* and *Phomopsis* sp. As pathogenic host-specialised strains of these fungi have been reported in other weed biocontrol projects overseas, the feasibility of using them as inundative agents should be assessed by undertaking experimental 'Koch's postulates' pathogenicity trials to determine their virulence as primary pathogens.

Prospects and conclusions for barberry rust, *Puccinia graminis*

Our finding of *Puccinia graminis* on the flowers of *B. glaucocarpa* in New Zealand represents a new host/pathogen record for this country and may also be the first time the rust has been found on the flowers of any *Berberis* species.

As the rust has a complicated lifecycle involving economically important grasses and cereals, shows limited potential to infect and damage barberry, and is rare, it will not be sensible to investigate using this rust as a biocontrol agent for barberry in New Zealand.

Prospects and conclusions for *Aspergillus niger* and *Rhizopus* sp.

Although *Aspergillus niger* and *Rhizopus* were associated with serious berry fruit decay of *B. glaucocarpa*, both are cosmopolitan and widespread spoilage fungi that are unlikely to be useful as inundative biocontrol agents.

6.2 Invertebrates

With only one specialist invertebrate, *Liosomaphis berberidis* (Aphididae), found to be using *Berberis* species in New Zealand there remains a very real opportunity to successfully introduce invertebrate biocontrol agents to use the untapped feeding niches created by these plants.

McGregor (2002) reports heavy infestations of the aphid *L. berberidis* on *B. vulgaris* in Poland resulted in discoloration, curling and premature fall of leaves, as well as harmful and disfiguring fungal attack due to honeydew emissions (Jaskiewicz 1995, 1996). It also attacks *B. thunbergii* (Labanowski 1990). *L. berberidis* has been recorded in New Zealand only from *B. glaucocarpa* (Dale & Maddison 1982), but in this survey was also found to be present on *B darwinii*.

Effective seed dispersal by these *Berberis* species is a major reason for their invasiveness, so particular, but not exclusive, effort should be given to identifying biocontrol agents that attack flowers, fruits and seeds.

7. Recommendations

In the light of our conclusions that only one host-specific invertebrate herbivore (*Liosomaphis berberidis)* contributes minimal damage to *Berberis* species in New Zealand, and with no specialised pathogenic fungi known to be present on these weeds in New Zealand, we recommend:

1. A classical biocontrol programme for barberry should proceed along the following lines:
- Survey *B. darwinii* and other *Berberis* species in Chile and Argentina to identify prospective invertebrate and fungal biological control agents;
- Survey *B. glaucocarpa* and other *Berberis* species in the western Himalaya to identify prospective invertebrate and fungal biological control agents;
- Undertake host-range tests on plant species of importance to New Zealand for the most promising prospective invertebrates and fungal biological control agents;
- Host-specific invertebrates and pathogenic fungi should be introduced to New Zealand as classical biocontrol agents.

2. The inundative biocontrol potential of the *Colletotrichum gloeosporioides*, *Phomopsis* sp. and *Sclerotinia sclerotiorum* isolates associated with leaf, flower and fruit disease symptoms should be determined using inoculation trials to elucidate their taxonomy, epidemiology and pathogenicity.

3. The possible presence of bacterial or viral pathogens associated with leaf disease symptoms that were not associated with culturable fungal pathogens should be determined.

This follows the recommendations as made by McGregor (2002).

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10. Appendices

Appendix 1 Fungal specimen site numbers for *Berberis* species

Berberis glaucocarpa

Apata (Pathogen Specimen [Spec] No. 98)
Brancott Station (Spec. No. 100, 136)
Castaly River (Spec. No. 99)
Cheviot (Spec. No. 101, 140)
Deadman's Island (Spec. No. 111, 137)
Featherston (Spec. No. 135)
Handrail Creek (Spec. No. 109)
Kawakawa Bay (Spec. No. 130)
Kowhai River (Spec. No. 102, 139)
Kumara (Spec. No. 105)
Kurow (Spec. No. 112, 131)
Lake Rerewhakaaitu (Spec. No. 126)
Maramarua (Spec. No. 128)
Mt Peel Station (Spec. No. 115)
Oparara (Spec. No. 103)
Pelorus Bridge (Spec. No. 110)
Puhoi (Spec. No. 129)
Rai Valley (Spec. No. 104)
Rangiriri (Spec. No. 127)
Reedy Road (Spec. No. 108)
Stonehurst Farm (Spec. No. 107, 138)
Tapanui (Spec. No. 120)
Tukituki Road (Spec. No. 134)
Waimate North (Spec. No. 94)
Welcome Bay (Spec. No. 96)
Whatatiri (Spec. No. 95)
Woodstock (Spec. No. 106)

Berberis darwinii

Barberry Road (Spec. No. 123)
Catlins Valley Road (Spec. No. 122)
Kaitangata (Spec. No. 117)
Karori (Spec. No. 132)
Lower Hutt (Spec. No. 133)
Manapouri (Spec. No. 125)
Mouat's Saddle (Spec. No. 124)
Mt Peel Station (Spec. No. 113, 114)
Pukehihi (Spec. No. 116)
Rotorua (Spec. No. 97)
Tahakopa (Fauna only)
Thompson's Bush (Spec. No. 121)
Whisky Gully (Spec. No. 118)

Berberis vulgaris

Mt Iron (Spec. No. 119)

Appendix 2. Non-herbivorous invertebrates associated with *Berberis* species collected from sites throughout New Zealand during 2003/2004

Insect species	Common name	Feeding mode	Origin	Sites	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Phylum ARTHROPODA							
CRUSTACEA							
ISOPODA	**slaters**						
Unid. Isopoda		saprophytic		1,2,5,6,10,14	C		
Class ARACHNIDA							
ACARINA	**mites and ticks**						
Unid. Acarina				26,34,37,39	R	C	
Anystidae							
Anystis sp.	whirlygig mite	predatory	introduced	8,15,17,23	O		
Bdellidae							
Bdellodes sp.				13,17	R		
Oribatid mites							
Unid. Oribatid mites		fungivorous		13,15,16,17,23,24,34,38,39	C	O	
ARANEIDA	**spiders**						
Unid. Araneida		predatory		all sites	A	A	C
PSEUDOSCORPIONIDA	**false scorpions**						
Unid. Pseudoscorpiones		predatory		1,4,5,14,15,30	O	R	
HEXAPODA							
COLLEMBOLA	**springtails**						
unid. Collembola		saprophytic		11,12,27,36,38	R	O	
INSECTA	**insects**						
BLATTODEA	**cockroaches**						
Celatoblatta sp.		saprophytic	native	1,2,3,7,10,12,14,15,18,30,31	C	O	

Insect species	Common name	Feeding mode	Origin	Sites	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Parellipsidium sp.		saprophytic	native	2	R		
COLEOPTERA	**beetles**						
Aderidae							
Scraptogetus anthracinus Broun			native	26	R		
Anthicidae	**ant beetles**						
Anthicus minor Broun		saprophytic	native	3,17	R		
Anthribidae	**fungus weevils**						
Androporus discedens (Sharp)		fungivorous	native	3	R		
Etnalis spinicollis Sharp		fungivorous	native	12,14,15	R		
Dysnocryptus inflatus (Sharp)		fungivorous	native	6	R		
Phymatus sp.		fungivorous	native	10	R		
Sharpius brouni (Sharp)		fungivorous	native	3,4,6,8,9,14,15,30,32	O	R	
unid. Anthribidae		fungivorous		9	R		
Cantharidae	**soldier beetles**						
Malthodes pumilus (Brebisson)			native	12	R		
Carabidae	**ground beetles**						
Demetrida sp.		predatory	native	12	R		
Cleridae	**checkered beetles**						
Lemidia aptera (Sharp)		predatory	native	13	R		
Lemidia sp.		predatory	native	37		R	
Phymatophaea sp.		predatory	native	14,25	R		
Coccinellidae	**ladybirds**						
Adalia bipunctata (L.)	two-spotted lady bird	predatory	introduced	2,24,25,26	O		
Coccinella undecimpunctata L.	eleven-spotted lady bird	predatory	introduced BCA	15,17,24,25	O		
Halmus chalybeus (Boisduval)	steely-blue ladybird	predatory	introduced BCA	1,2,4,5,15	C		

Insect species	Common name	Feeding mode	Origin	Sites	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Rhyzobius acceptus (Broun)			native	32		R	
Rhyzobius fagus (Broun)		predatory	introduced	14,17	R		
Rhyzobius forstieri (Mulsant)		predatory	introduced BCA	22	R		
Rhyzobius sp.			native	2,4,5,8,13,15,17	C		
Corylophidae	**hooded beetles**						
Sericoderus sp.		fungivorous		14	R		
unid. Corylophidae				17	R		
Cryptophagidae	**cryptic beetles**						
Micrambina sp.		pollen/fungus feeder	native	15	R		
Paratomaria sp.		pollen/fungus feeder	native	15,17,19,26,39	O	R	
unid. Cryptophagidae		pollen/fungus feeder		most sites	A	C	
Erotylidae	**handsome fungus beetles**						
Loberus depressus (Sharp)		fungus/pollen feeder	native	23,39	R	R	
Lathridiidae	**mildew beetles**						
Aridius bifasciatus (Reitter)		fungivorous	introduced	39		R	
Corticaria sp.		fungivorous	introduced	26	R		
unid. Lathridiidae		fungivorous		1,5,8,9,11,15,16,17,24,26,30,32,34,39	C	O	
Melyridae							
unid. Melyridae			native	1,12,15,17,23,24,25,26,27,34,38,39	C	O	
Mycetophagidae	**fungus beetles**						
Litargus vestitus (Sharp)		fungivorous	introduced	5,10	R		
Nitidulidae	**sap beetles**						
Soronia hystrix Sharp		fungivorous	native	34		R	
Phalacridae							
unid. Phalacrid				34		R	

Insect species	Common name	Feeding mode	Origin	Sites	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Salpingidae	**bark mould beetles**						
Salpingus bilunatus Pascoe		fungivorous	native	1,4,5,8,9,10,15,19	C		
Salpingus perpunctatus Broun		fungivorous	native	9,30	R	R	
Salpingus sp.		fungivorous	native	1,10	R		
Scarabaeidae							
Ataenius picinus Harold		saprophytic	introduced	2	R		
Scirtidae	**marsh beetles**						
(formerly Helodidae)							
unid. Scirtidae		adults: pollen larvae: detritus	native	5,8,12,14,15,16,17,19,23,24,25,26,27,30,38,39	C	O	
Staphylinidae	**rove beetles**						
unid. Staphylinidae		predatory	native	9,10,11,15,17,18,19,22,25,34	O	R	
Trogossitidae	**cadelle beetles**						
Grynoma sp.		?predatory	native	26	R		
unidentified Trogossitidae			native	14,17	R		
DERMAPTERA	**earwigs**						
Forficula auricularia L.	European earwig	omnivorous	introduced	6,8,9,15	O		
DIPTERA	**flies**						
Tachinidae	bristle flies						
Pales funestra (Hutton)	New Zealand leafroller tachinid	parasitoid	native	12,24	R		
Trigonospila brevifacies (Hardy)	Australian leafroller tachinid	parasitoid	introduced BCA	3,5,11	O		
EPHEMOPTERA	**mayflies**						
unid. Ephemeroptera				13,17,38	R	R	
HEMIPTERA	**Bugs**						
Miridae	**mirid bugs**						

Insect species	Common name	Feeding mode	Origin	Sites	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Sejanus albisignatus (Knight)		predator/pollen feeder	native	8	R		
Pentatomidae	**shield bugs**						
Cermatulus nasalis (Westwood)	brown soldier bug	predatory	native	34		R	
Reduviidae	**assassin bugs**						
unid. Reduviidae		predatory		14,17,24,25,26	O		
HYMENOPTERA	**bees, wasps, ants**						
Braconidae	parasitic wasps						
Dolichogenidea sp.		parasitoid		3,5,6,15,18,32	O	R	
Dolichogenidea tasmanica Cameron		parasitoid	introduced BCA	11	R		
Meteorus cinctellus (Spinola)		parasitoid	introduced	12,42	R	R	
Meteorus pulchricornis Wesmael		parasitoid	introduced	26	R		
S.F. Microgasterinae							
unid.		parasitoid		31,36,39		R	
Formicidae	**Ants**						
Monomorium antarcticum (F. Smith)	southern ant	omnivorous	native	5,14,17	O		
Technomyrmex albipes (Smith)	white-footed house ant	omnivorous	introduced	1,2,4,5,8,17	C		
Ichneumonidae							
Diadegma sp.		parasitoid	native	30		R	
Glabridorsum stokesii (Cameron)		parasitoid	introduced BCA	5	R		
Phytodietus zealandicus (Ashmead)		parasitoid	native	23	R		
Xanthopimpla rhopaloceros Krieger	yellow-banded leafroller parasite	parasitoid	introduced BCA	6,9,11	R		
unid. Ichneumonidae				36		R	
Pteromalidae							
Trichomalopsis sp.		parasitoid		5	R		

Insect species	Common name	Feeding mode	Origin	Sites	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
LEPIDOPTERA	**moths and butterflies**						
(collected as larvae and reared to adult for identification)							
Tineidae	**Clothes moths**						
Opogona omoscopa (Meyrick)	dusky scuttler	saphrophytic	native	15	R		
MANTODEA	**praying mantids**						
Orthodera novaezealandiae (Colenso)	Praying mantis	predatory	native	11,14,17,25,26,43	O		R
NEUROPTERA	lacewings						
Hemerobiidae							
Micromus tasmaniae (Walker)	Tasmanian lacewing	predatory	introduced	1,3,5,7,8	C		
unid. Neuroptera				14,15,16,17,18,22,23,25	C		
ORTHOPTERA	**crickets and grasshoppers**						
Anostostomatidae							
Hemideina crassidens (Blanchard)	Wellington tree weta	omnivorous	native	18	R		
Hemideina thoracica White	Auckland tree weta	omnivorous	native	5,6	R		
Raphidophoridae	**cave weta**						
unid. Raphidophoridae		omnivorous		17,34	R	R	
PLECOPTERA	**stoneflies**						
Stenoperla sp.		algal feeders	native	12,26,33,34,38	R	R	
unid. Plecoptera				15,19,23,37	O	R	
PSOCOPTERA	**book lice**						
unid. Psocoptera		saprophytic and fungivorous		most sites	A	C	
THYSANOPERA	**thrips**						
Sub-order Tubulifera							
Phlaeothripidae							
Heptathrips cottieri (Mound and Walker)		fungivorous		12	R		

Insect species	Common name	Feeding mode	Origin	Sites	*B. glaucocarpa*	*B. darwinii*	*B. vulgaris*
Heptathrips tillyardi (Mound and Walker)		fungivorous		26	R		
Nesothrips propinquus (Bagnall)		fungivorous		16,17,24	O		
Nesothrips sp.		fungivorous		23	R		
unid. Tubulifera		fungivorous		11	O		

NOTES:

BCA: biological control agent

refer to maps for site numbers

R: rare

O: occasional

C: common

A: abundant