# Predicting the likely success of biological control of hawkweeds in New Zealand

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#### **Summary**

Five species of hawkweed (Hieracium pilosella, H. praealtum, H. caespitosum, H. aurantiacum and H. lepidulum), originating from Europe, are invasive in native grassland ecosystems in New Zealand, displacing desirable environmental and agricultural species. A rust fungus and five insect species have been identified for introduction as complementary biological control agents. The rust fungus Puccinia hieracii var. piloselloidarum is now widely established as the result of an accidental introduction, but attacks only some forms of H. pilosella. Two insect species, a plume moth, Oxyptilus pilosellae, and a gall wasp, Aulacidea subterminalis, have already been released in the field, and two syrphid flies (Cheilosia urbana and C. psilophthalma), and a gall midge (Macrolabis pilosellae) are expected to be released shortly. The six control agents should complement each other because they attack different parts of the plant, and between them are predicted to affect all five weedy hawkweeds. All six agents are likely to achieve significant levels of damage on H. pilosella, three of them will target H. praealtum, four of them should suppress H. caespitosum and H. aurantiacum. Only one insect species, the root-feeding C. urbana, is predicted to be as damaging to H. lepidulum as to H. pilosella, so further control agents are likely to be needed to suppress this nonstoloniferous hawkweed.

# Introduction

A biological control programme for Hieracium spp. (Asteraceae) (hawkweeds) was initiated in 1992 because the invasiveness of this group of plants was identified as an important factor contributing to the desertification of large areas of tussock grassland in the South Island hill- and high-country of New Zealand (Syrett et al. 1996). Representatives of both subgenera Pilosella (stoloniferous) and Hieracii (nonstoloniferous) species are invasive in New Zealand. One possible biological control agent, a strain of the rust fungus Puccinia hieracii (Röhl.) H.Mart. var. piloselloidarum (Probst) Jørst., was accidentally introduced and has established in New Zealand on H. pilosella (Morin and Syrett 1996). Not all H. pilosella plants in New Zealand appear to be susceptible to this strain of the rust so two further strains have been imported for widespread release (T.A. Jenkins, personal communication).

Five of the nine adventive Hieracium species in New Zealand are regarded as problem weeds: H. pilosella L. (mouse-ear hawkweed) is the most widely established, and has been in New Zealand the longest, since 1878; H. praealtum Gochnat (king devil) is also widespread, but less abundant; while H. caespitosum Dumort. (field hawkweed) and H. lepidulum (Stenstr.) Omang (tussock hawkweed) are more recent arrivals (1940 and 1946 respectively) with more limited distributions (Webb et al. 1988). Hieracium aurantiacum L. (orange hawkweed) has only recently been regarded as weedy in New Zealand, although it has been here since 1911 and is also a problem weed in North America (Wilson et al. 1997).

A survey was conducted throughout the Hieracium-infested areas of New Zealand (Figure 1) for insects feeding on these plants with the aim of identifying phytophagous species that were already causing damage (Syrett and Smith 1998). Seventy six species of native and exotic insects were found, including both polyphagous and oligophagous species, but there were none of the specialist European Hieraciumfeeding insects. There were no species causing significant damage that would make the introduction of any potential biological control agent redundant.

Surveys were conducted in Europe to identify those host-specific insects that had potential for introduction into New Zealand as biological control agents for Hieracium species (Jordan 1993, Syrett and Sárospataki 1993). Four species were selected for further study: a plume moth, Oxyptilus pilosellae Zeller (Pterophoridae), whose larvae feed in the crown of the rosette plant; Aulacidea subterminalis Niblett (Cynipidae), which galls stolons; Cheilosia urbana (Meigen) (= C. praecox (Zetterstedt)) (Syrphidae), which feeds externally on roots; and Macrolabis pilosellae (Binnie) (Cecidomyiidae), which produces galls on leaves and stems as well as at stolon tips (Grosskopf 1995, 1996). A further Hieraciumfeeding syrphid, Cheilosia psilophthalma (Becker), that feeds on the above-ground parts of the plant and complements the activity of C. urbana, was later identified,

and added to the suite of potential agents (Grosskopf 1996).

First-instar larvae of O. pilosellae hatch after 2-3 weeks, and over winter at this stage, or rarely as second-instar larvae. In spring, larvae are found in a loose web at the centre of the rosette crown. By feeding at the growing points, larvae deform and severely stunt the growth of leaves and stolons. Flowering is reduced, and heavily infested rosettes may die. Levels of parasitism of up to 35% in fieldcollected larvae of O. pilosellae have been observed in Europe (Grosskopf 1997). The gall wasp, A. subterminalis, is parthenogenic and only females are produced. Eggs are laid into stolon tips, and larvae develop within the tip, inducing formation of a gall. The wasp larvae spend winter inside the galls, pupating in spring so that new-generation adult wasps emerge in early summer. Stolons infested with gall wasps produce deformed daughter rosettes, or form galls instead of new plants. Plant nutrients are diverted from forming daughter rosettes to producing galls so that vegetative reproduction may be severely impeded (Grosskopf 1995,

The predicted host range of the five insect species to be established in New Zealand was determined experimentally, and from these data, and the known feeding behaviour of the species, possible comparative levels of damage to weedy Hieracium species were predicted. A simulation experiment was conducted to measure the impact of two levels of depression of Hieracium species on vegetation composition of hawkweed-infested grassland. The field release of two species, O. pilosellae and A. subterminalis, has been approved, but the outcome of an application for release of the other three insects is still awaited. A summary of the host range testing, preliminary results from the simulation experiment, and rearing and release of O. pilosellae and A. subterminalis are reported here.

#### Methods

# Host specificity tests

A similar number of plant species were selected for host specificity tests with all five insects. They included all Hieracium species naturalized in New Zealand (except H. pollichiae Schulz.-Bip.), from both subgenera Pilosella and Hieracium, species chosen from other genera in the tribe Lactuceae (including native New Zealand species that would not previously have been exposed to the insects), economically important members of other tribes in the family Asteraceae, and a selection of more distantly related representatives of families containing important cultivated plants and native New Zealand plants. Host plants of close relatives of the proposed biological control agents were also included. Tests were conducted by CABI Bioscience at Delémont, Switzerland, and under containment by Landcare Research at Lincoln.

No-choice larval feeding and development tests, single-choice larval feeding tests, and multiple-choice oviposition tests were conducted with the plume moth, O. pilosellae (Syrett et al. 1997). Larval development tests were carried out on potted plants. Pots were embedded in the ground, covered with individual gauze bags, and examined after 10 months when all stages of the plume moth were retrieved.

For the gall wasp, A. subterminalis, all the tests were combined oviposition and larval development tests, because once the egg has been placed by the female wasp, the larva hatches and feeds inside the developing gall, and is unable to transfer to another feeding site or host plant (Syrett et al. 1998). Oviposition was induced in gauze cages containing four randomly allocated test plants with a single H. pilosella plant. Female wasps were released into each cage for four days. When gall development was complete, numbers of galls, adult wasps, and parasitoids were recorded. All ungalled plants were dissected to identify eggs or immature stages of the wasp.

No-choice development tests, and single-choice oviposition tests, were carried out both with the root-feeding hover fly, C. urbana, and the crown-feeding hover fly, C. psilophthalma. An open field test was also carried out with C. urbana. In nochoice larval development tests newly emerged larvae were transferred into leaf axils of each test plant. Plants were individually caged in gauze bags for several days until larvae had established themselves in an appropriate feeding site. Potted plants were then embedded in soil, and maintained for four months when all pots were checked for the presence of larvae. Larvae were extracted, and placed individually in vials with soil until they pupated. Numbers pupating successfully, and numbers of adults emerging the following spring, were recorded.

For the tests with the gall midge, M. pilosellae, newly emerged female flies were placed with male flies within a gauze bag covering each test plant. Plants were checked weekly for signs of gall development, and once this was observed, plants were placed in gauze cages. Adults were collected and counted as they emerged.

Simulated biological control of hawkweed In 1993 three study sites were selected in hawkweed-infested regions of the South Island: a tall-tussock community (Chionochloa rubra grassland, with bare ground, and invading H. pilosella, H. praealtum, and H. lepidulum) at Lindis Pass; a short-tussock community (Festuca novaezelandiae with native turf grasses and

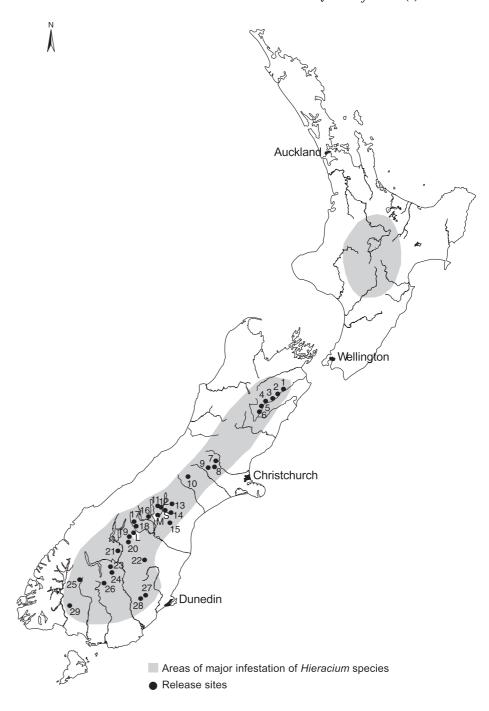


Figure 1. Distribution of weedy *Hieracium* species (hawkweeds) in New Zealand, and sites where the biological control agents O. pilosellae and A. subterminalis have been released.

Gall wasp release sites: 1 = Muller,  $2^* = Molesworth$ ,  $3^* = Molesworth$ , 4 = Molesworth, 5 = Molesworth, 6\* = Molesworth, 7 = Castle Hill, 8\* = Castle Hill, 9 = Ryton, 10 = Clent Hills, 11 = Balmoral, 12 = Balmoral, 13 = Glenrock, 14\* = Grampians, 15 = Stoney Creek, 16 = Ben Ohau, 17 = Ben Avon, 18\* = Ben Avon, 19 = Geordie Hills, 20\* = Geordie Hills, 21 = Mt. Pisa, 22 = Michael's Peak, 23 = Nokomai, 24 = Nokomai, 25 = Mt. Nicholas, 26 = Lorn Peak, 27 = Goulburn, 28\* = Goulburn, 29 = Mt. Linton Stations.

\*sites where galled plants were planted out.

Plume moth release site: 13 = Glenrock Station.

Simulated biological control experimental sites: S = Sawdon Station, **M** = Maryburn Station, and L = Lindis Pass Reserve.

herbs, some matagouri (Discaria toumatou) and invading H. pilosella, H. praealtum, and H. caespitosum) at Maryburn Station in the Mackenzie Basin; and a severely degraded site (sparse F. novae-zelandiae, native herbs and turf grasses, with extensive cover of H. pilosella and substantial amounts of bare ground) at Sawdon Station (Figure 1). In order to reduce variability in the results sub-plots were selected within each treatment plot so that hawkweed cover was in the range 40-60%. Two treatments were used to suppress hawkweeds: glyphosate herbicide was handpainted onto all, or 50%, of hawkweed leaves and stolons in each treatment subplot. Treatment sub-plots were small (0.1  $\times$  0.1 m) so that 50 and 100% suppression represented a realistic simulation of effective biological control at this scale. We reapplied the treatments as necessary to maintain hawkweed cover at 0 and 50% of initial levels respectively. Visual estimates of vegetation type and cover in treatment sub-plots and control plots were made once a year from 1993 to 1999 by a single skilled observer (C.M.). The 1993 measurements were taken before the treatments were applied.

# Rearing and release of O. pilosellae and A. subterminalis

Up to 20 newly emerged adult moths of O. pilosellae were placed with eight-potted H. pilosella plants in a clear plastic box within a controlled-temperature room under long days and moderate temperatures. Moths were provided with a bouquet of nectar-producing flowers, cotton wool soaked in a dilute solution of honey-water, and pollen as a food source. A second method was tested in which moths were transferred to polyester fibre mesh cages maintained in a glasshouse where they were subjected to natural daylight. They were provided with similar food sources. Eggs were left on the potted plants so that emerging larvae would search out suitable over-wintering sites on the undersides of leaves. Plants were maintained through the winter, so that larvae continued their development in the spring, and newgeneration adult moths emerged from cocooned pupae on the underside of leaves. The first field release, of 32 adult moths, was made on 26 March 1999 at Glenrock Station in the Mackenzie Basin.

Adult gall wasps (*A. subterminalis*), newly emerged from galls, were placed with potted *H. pilosella* plants in either clear plastic boxes or polyester fibre mesh cages in either controlled-temperature rooms under artificial lighting, or in a glasshouse. Cotton wool soaked in a dilute solution of honey-water and pollen was provided as food for the wasps. Hawkweed plants with developing galls were maintained under spring/summer conditions for 3–4 months, when some

galls were harvested and placed into an artificial winter for 5–6 months, and the rest left to develop naturally on the plant in a shade house under winter/spring conditions. The first two field releases, each of 100 adult wasps, were made on 11 February 1999 on Balmoral and Glenrock Stations, in the Mackenzie Basin. A further 27 releases were made at sites throughout the South Island between November 1999 and February 2000 (Figure 1). Eighteen releases comprised 80–150 adult wasps, and nine consisted of 3–6 plants bearing a total of 15–40 galls. The plants were grown in pots and transplanted in the field.

#### **Results**

#### Host specificity tests

Normal larval feeding by the plume moth, *O. pilosellae*, was observed only on plants within the genus *Hieracium*, and *H. pilosella* was preferred over other *Hieracium* species (Table 1). In no-choice feeding tests a small number of late-instar larvae were able to complete development on the New Zealand native plants *Sonchus kirkii* and *Embergeria grandifolia* (Lactuceae), but in single-choice feeding tests these plants were less preferred than *H. pilosella*.

Galls of *A. subterminalis* were produced on plants of *H. pilosella* and *H. aurantiacum*, but not on any other plant species. Wasps developed through to adult on both *H. pilosella* and *H. aurantiacum* (Table 2).

In no-choice larval development tests larvae of the root feeding hover fly, *C. urbana*, were found on all *Hieracium* species except *H. murorum* (Table 3), but plants

outside the genus were not attacked. In single-choice oviposition tests eggs were laid on all *Hieracium* species, but not on any of the other test plants. Very few larvae established in the open field test, and most were on *H. aurantiacum*, *H. praealtum* and *H. pilosella*. A proportion of all above ground-feeding hover fly larvae, *C. psilophthalma*, developed to adult on all *Hieracium* species in no-choice larval development tests (Table 4). No larvae, pupae, or feeding marks were found on any test plants outside the genus. Fewer larvae developed on non-stoloniferous *Hieracium* species (Table 4).

Gall development, and emergence of new-generation adult gall midges of *M. pilosellae* occurred only on *Hieracium* species within the sub-genus *Pilosella*. Two species within this group, *H. aurantiacum* and *H.* × *stoloniflorum*, were unacceptable hosts in tests (Table 5).

From results of the host range tests, summarized here from Grosskopf (1996, 1997) and Grosskopf and Hassler (1998), the host range of all five insect species was predicted (Table 6).

Simulated biological control of hawkweed Preliminary analyses were conducted using data averaged over all three sites. Except for 1993 (prior to treatment), the treatment plots showed a significant increase in bare ground and litter (P<0.001) over the controls. This was not surprising since treatments were designed to remove a substantial proportion of vegetation, and removed hawkweed was not rapidly replaced by other plants. Some 100%

Table 1. Acceptability of *Hieracium* spp. to *Oxyptilus pilosellae* in no-choice larval development tests conducted in Switzerland.

Hieracium species	No. L1 <sup>A</sup> larvae placed on plants	% larvae developed to adul	
Sub-genus Pilosella			
H. pilosella EUR	195	27.2	
H. pilosella NZ	72	38.9	
H. aurantiacum	70	7.1	
H. caespitosum EUR	65	4.6	
H. caespitosum USA	40	20.0	
H. praealtum	68	2.9	
Sub-genus Hieracium			
H. lepidulum	70	5.7	
H. murorum	60	1.7	
H. sabaudum	70	5.7	

<sup>&</sup>lt;sup>A</sup> First-instar larvae.

Table 2. Acceptability of *Hieracium pilosella* and *H. aurantiacum* to *Aulacidea subterminalis* (Cynipidae) in multi-choice oviposition and larval development tests conducted in Switzerland.

Plant species	No. plants offered	% plants attacked	No. galls/ plant	No. wasps/ plant		% developed
					plant	to adult
H. pilosella EUR	76	97	9.1	4.4	2.3	81
H. pilosella NZ	14	71	11.0	8.1	2.4	77
H. aurantiacum	4	50	15.5	12.5	2.5	88

treated plots had less than 50% vegetation cover one year after treatment. From 1993 to 1995 there was significantly higher cover of vegetation excluding hawkweeds in control plots than in treated plots (P<0.001 for all three years). This occurred because treatment plots were not randomly selected, and had higher initial levels of hawkweed than the controls.

However, from 1996 to 1998 vegetation cover excluding hawkweeds was 10-30% higher in treated plots than in control plots, although these differences were not significant. The levels of vegetation cover excluding hawkweeds for both 50% and 100% treatments increased with time, while values for control plots did not change significantly.

Table 3. Acceptability of Hieracium species to Cheilosia urbana (Syrphidae) in no-choice larval development tests conducted in Switzerland.

Test plant species	No. of L1 <sup>A</sup> transferred	% developed to mature larvae	% developed to pupae	% developed to adult
Sub-genus Pilosella				
H. pilosella EUR	160	63	54	50
H. pilosella NZ	45	56	44	44
H. aurantiacum	30	70	63	60
H. caespitosum EUR	35	57	54	46
H. caespitosum NZ	35	57	54	43
H. caespitosum USA	30	87	83	80
H. praealtum	35	57	49	46
H. × stoloniflorum	50	28	16	12
Sub-genus Hieracium				
H. argillaceum	40	13	2.5	2.5
H. lepidulum	30	53	40	33
H. murorum	40	_	_	_
H. sabaudum	30	47	40	40

<sup>&</sup>lt;sup>A</sup> First instar larvae.

Table 4. Acceptability of Hieracium species to Cheilosia psilophthalma (Syrphidae) in no-choice larval development tests conducted in Switzerland.

Test plant species	No. of L1 <sup>A</sup> transferred	% developed to mature larvae	% developed to pupae	% developed to adult
Sub-genus Pilosella				
H. pilosella EUR	95	35	30	23
H. pilosella NZ	30	33	33	33
H. aurantiacum	65	45	43	42
H. caespitosum EUR	25	36	36	36
H. caespitosum NZ	20	25	20	15
H. caespitosum USA	25	48	40	32
H. praealtum	50	44	44	42
H. × stoloniflorum	30	33	33	33
Sub-genus Hieracium				
H. argillaceum	30	3	3	3
H. lepidulum	60	5	3	2
H. murorum	45	9	9	4
H. sabaudum	45	16	13	11

<sup>&</sup>lt;sup>A</sup> First instar larvae.

Table 5. Acceptability of stoloniferous Hieracium species (sub-genus Pilosella) to Macrolabis pilosellae (Cecidomyiidae) in combined no-choice oviposition and larval development tests conducted in Switzerland.

Test plant species (sub-genus <i>Pilosella</i> )	Number of plants		Number of female flies	Number of adult flies	
	offered	produced galls	incubated	emerged	
Hieracium pilosella EUR	38	36	105	417	
H. pilosella NZ	11	9	33	114	
H. aurantiacum	12	0	34	0	
H. caespitosum EUR	6	6	17	124	
H. caespitosum USA	6	6	15	53	
H. praealtum	6	5	16	27	
H. × stoloniflorum	12	2	36	0	

### Rearing and release of O. pilosellae and A. subterminalis

Plume moths did not oviposit in cages under artificial lighting. Under glasshouse conditions, eggs were found on the fine hairs on the upper side of H. pilosella leaves. Small numbers of hatching larvae developed through to adult, allowing one field release to be made, but this insect has proved difficult to rear under artificial conditions and further work is required to develop a successful method. No larvae, or new-generation adult moths, have yet been recovered from the field. Gall wasps oviposited successfully under all conditions, and females survived for up to 14 days. Oviposition scars, usually on the underside of stolons, a few millimetres from the stolon tip, were apparent after 3-4 days. Gall formation was apparent after 2-3 weeks, as the stolons began to swell. Not all oviposition attempts led to gall development. Galls were recovered from two of the six sites checked in April 2000; the remaining sites will be checked in 2001.

#### Discussion

The rust fungus P. hieracii var. piloselloidarum is widely established on H. pilosella in New Zealand, and is often conspicuous in spring and autumn (T.A. Jenkins, personal communication). Two insects (O. pilosellae and A. subterminalis) have already been released and galls of the latter have established under natural conditions. In the simulated biological control trial, preliminary indications are that when hawkweeds are suppressed, they are replaced only slowly by other species. Vegetation other than hawkweeds increased over time in hawkweed suppressed plots, while that in the control plots remained relatively stable. Increases in hawkweed are believed to be a result of natural invasion facilitated by management practices (Johnstone et al. 1999), although others have suggested they increase only in degraded systems (Treskonova 1992). If introduced biological control agents suppress hawkweeds effectively they may be replaced by more desirable vegetation.

Observations in Europe showed that both the insect biological control agents released in New Zealand have potential to suppress some species of hawkweeds (Grosskopf, personal observation). Although the plume moth has not been found on species of Hieracium other than H. pilosella in Europe, host tests indicated that it might attack two of the weedy species (H. caespitosum and H. lepidulum) established in New Zealand (Table 1). From life cycle information reported in Europe (Grosskopf 1995, Syrett et al. 1997) we predict that plume moth eggs will be found on H. pilosella in the field in New Zealand from January to March. Because there are

Table 6. Predicted level of attack on five weedy species of *Hieracium* by one pathogen, and five insect, biological control agents.

Hawkweed species	Puccinia hieracii var. piloselloidarum	Oxyptilus pilosellae	Aulacidea subterminalis	Cheilosia urbana	Cheilosia psilophthalma	Macrolabis pilosellae
H. pilosella	<b>/</b> /	<b>//</b>	<b>//</b>	11	<b>//</b>	<b>//</b>
H. praealtum	-	✓	-	<b>//</b>	<b>√</b> √	<b>√</b> √
H. caespitosum	-	<b>//</b>	-	<b>//</b>	<b>//</b>	<b>//</b>
H. aurantiacum	-	✓	<b>//</b>	<b>//</b>	<b>√</b> √	
H. lepidulum	-	✓	_	<b>//</b>	✓	-

✓✓ equivalent level of attack to that on *H. pilosella*. ✓ minor level of attack compared to that on *H. pilosella*.

no specialized pterophorid parasitoids in New Zealand, it is likely that these moths may attain higher populations here (Syrett et al. 1997). Provided rearing problems can be surmounted, we expect that *O. pilosellae* will establish throughout the range of *H. pilosella* in New Zealand, but may be adversely affected in areas subject to summer drought.

In Europe galls of the wasp A. subterminalis have been recorded only from H. pilosella (Syrett et al. 1998), but laboratory host tests showed that it could perform equally well on H. aurantiacum. Newly developing galls should be found in New Zealand from late January onwards. Rates of parasitism of 36% have been observed in Europe, indicating that wasps may also attain higher populations here in New Zealand. There are no native New Zealand cynipid wasps, so specialized parasitoids are also absent. We predict that A. subterminalis should establish throughout the ranges of H. pilosella and H. aurantiacum in New Zealand, but populations may be limited under conditions when plants produce few stolons.

We expect that these two insects, together with the already established rust fungus, may significantly reduce the spread and impact of H. pilosella, and perhaps have some impact on H. caespitosum and H. aurantiacum. Three other insects still under study, C. urbana, C. psilophthalma, and M. pilosellae, should complement the activity of the two already released because, apart from attacking different parts of the plant, and exerting pressure at different times in the season, host range tests indicate that they will feed on a broader range of Hieracium species (Table 6). However, it is likely that further agents will be needed to suppress the nonstoloniferous H. lepidulum.

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