

# Disruption of an exotic mutualism can improve management of an invasive plant: varroa mite, honeybees and biological control of Scotch broom *Cytisus scoparius* in New Zealand

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## Summary

1. A seed-feeding biocontrol agent *Bruchidius villosus* was released in New Zealand (NZ) to control the invasive European shrub, broom *Cytisus scoparius*, in 1988 but it was subsequently considered unable to destroy sufficient seed to suppress broom populations. We hypothesized that an invasive mite *Varroa destructor*, which has caused honeybee decline in NZ, may cause pollinator limitation, so that the additional impact of *B. villosus* might now reach thresholds for population suppression.
2. We performed manipulative pollination treatments and broad-scale surveys of pollination, seed rain and seed destruction by *B. villosus* to investigate how pollinator limitation and biocontrol interact throughout the NZ range of broom.
3. The effect of reduced pollination in combination with seed-destruction was explored using a population model parameterized for NZ populations.
4. Broom seed rain ranged from 59 to 21 416 seeds m<sup>-2</sup> from 2004 to 2008, and was closely correlated with visitation frequency of honeybees and bumblebees. Infestation of broom seeds by *B. villosus* is expected to eventually reach 73% (the average rate observed at the localities adjacent to early release sites).
5. The model demonstrated that 73% seed destruction, combined with an absence of honeybee pollination, could cause broom extinction at many sites and, where broom persists, reduce the intensity of treatment required to control broom by conventional means.
6. Nevertheless, seed rain was predicted to be sufficient to maintain broom invasions over many sites in NZ, even in the presence of the varroa mite and *B. villosus*, largely due to the continued presence of commercial beehives that are treated for varroa mite infestation.
7. *Synthesis and applications.* Reduced pollination through absence of honeybees can reduce broom seed set to levels at which biocontrol can be more effective. To capitalize on the impact of the varroa mite on feral honeybees, improved management of commercial beehives (for example, withdrawal of licences for beekeepers to locate hives on Department of Conservation land) could be used as part of a successful integrated broom management programme at many sites in NZ.

**Key-words:** biological control, bumblebee, honeybee, integrated weed management, invasion, varroa mite, weed

## Introduction

Interactions between non-indigenous species can be facilitative or detrimental (Simberloff & Von Holle 1999). For example,

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invasive pollinator mutualisms (e.g. Barthell *et al.* 2001) may promote weed invasions, while deliberately introduced biocontrol agents may have a detrimental impact on introduced weed populations (Thomas & Reid 2007). The relationships between multiple non-indigenous species may be complex, particularly when there are both facilitative and detrimental associations.

An example of a non-indigenous species that interacts with both facilitative and detrimental non-indigenous species is the European shrub Scotch broom, henceforth broom, *Cytisus scoparius* (L.) Link (Fabaceae), which is a serious weed in temperate Australia, New Zealand (NZ) and the USA (Paynter *et al.* 1996). Broom flowers must be 'triggered' by a pollinator for the keel petals to open and deposit pollen on the visiting insect (Stout 2000). Broom flowers were solely opened by bees (family Apidae) in the USA, Europe, Japan and Australia (Parker 1997; Stout 2000; Suzuki 2000; Simpson, Gross, & Silberbauer 2005). As in Australia (Simpson, Gross & Silberbauer 2005), broom seed set in NZ may also depend on exotic pollinators: no native bees belong to the family Apidae in NZ, but honeybees *Apis mellifera* L. and four species of bumblebee *Bombus* spp. have been introduced from Europe (Donovan 2007). A survey of NZ beekeepers reported that 55% of respondents used broom as a pollen forage source for their honeybee hives (Jarvis *et al.* 2006). Bumblebees (*Bombus terrestris*; Hanley & Goulson 2003) and native bees of the family Halictidae (Primack 1983) also visit broom flowers in NZ, but the relative importance of these pollinators was unknown. This also has relevance to Australia, where there is motivation to import *B. terrestris* as pollinators for the horticultural industry, despite the risk that they may increase pollination and, therefore, the abundance of exotic weeds (Stokes, Buckley & Sheppard 2006).

In NZ, broom fecundity is reduced by an exotic seed-feeding biological control agent *Bruchidius villosus* F., which was first released in 1988 (Syrett *et al.* 2000). Paynter *et al.* (1996) argued that *B. villosus* is unlikely to reduce fecundity sufficiently to affect broom populations, but should reduce the

rate that broom invades. Sheppard *et al.* (2002) subsequently concluded that seed-feeders may suppress broom if seed destruction is very high (e.g. 97% seed destruction at an improved pasture site). However, the exotic varroa mite *Varroa destructor* Anderson and Trueman was detected parasitizing honeybees near Auckland on the North Island of NZ in 2000 (Zhang 2000). The varroa mite may reduce broom pollination, due to the death of both feral and managed apiaries (Donovan 2007), potentially enhancing the impact of *B. villosus* by decreasing seed set to levels where the additional impact of *B. villosus* is more likely to reach thresholds for population suppression.

We, therefore, performed manipulative experiments and surveys to address the following questions:

1. Does broom rely on introduced bees for pollination in NZ and, if so, what are the relative contributions made by honeybees and bumblebees to seed set?
2. Could the varroa mite reduce broom pollination rates and seed set, by reducing honeybee abundance?
3. What is the current impact of the seed-feeding biocontrol agent *B. villosus* on NZ broom populations?

Finally, we use a previously developed population occupancy model parameterized for NZ populations to explore the potential for reduced pollination due to the varroa mite to improve the effects of biocontrol.

## Materials and methods

### MANIPULATIVE EXPERIMENT TO TEST FOR RELIANCE ON POLLINATORS

This experiment was performed at Palmerston North and at Lincoln, on the North and South Islands of NZ, respectively (see Table 1) during the summer of 2004/5 and repeated at Palmerston North in 2005/6.

Ten mature and flowering 'focal' broom plants were selected at each site. Four stems per plant were randomly assigned to four

**Table 1.** Characteristics of the broom populations sampled in the 2004–2006 survey

Site	Location	Altitude (m)	Habitat
<i>North Island sites</i>			
Te Matai Road	37°57'S, 176°09'E	470	Scattered patches; forestry
Lake Rerewhakaiitu	38°17'S, 176°30'E	440	Large dense infestation; wasteland
Tongariro National Park	39°21'S, 175°43'E	1030	Large dense infestation; pasture, feral bees had recently disappeared (N. Singers, pers. comm.)
Palmerston North	40°19'S, 175°45'E	100	c. 4 ha patchy stand, wasteland.
<i>South Island sites</i>			
Hanmer	42°34'S, 172°51'E	370	Patchy riparian infestation (several kilometres long and approximately 30–100-m deep). 16 beehives were present within 20-m of the sampled area in 2004/5, but moved to a site c. 4 km away in 2005/6
Landsdowne Valley	43°37'S, 172°35'E	40	c. 3 ha of patchy broom and gorse; wasteland.
Lincoln	43°38'S, 172°28'E	15	c. 0.2 ha dense broom patch surrounded by 16 beehives; wasteland.

treatments (see below). On each stem, a minimum of 15 mature but ‘untriggered’ flowers were labelled at fortnightly intervals and their fate (i.e. aborted or pod formed) was recorded at weekly intervals until all flowers had produced pods or been aborted. The treatments allocated were as follows:

1. *Sleeve*: stem sleeved with fine (approximately 0.2-mm) cloth mesh to exclude potential pollinators.
2. *Control*: no treatment, to allow natural pollination.
3. *Hand pollination*: To test for pollinator limitation, flowers were pollinated using a small paintbrush dipped in outcross pollen collected from plants growing > 10-m distant to swab the stigmas of hand-triggered flowers.
4. *Hand pollination plus sleeve*: a control for treatment 1, flowers were pollinated, as for treatment 3, but then sleeved as in treatment 1, to test whether a lack of pods in treatment 1 might be due to the sleeve treatment, rather than an absence of pollinators.

When pods matured, the number of seeds inside each pod was counted, so that the number of seeds per flower could be calculated.

## BROAD-SCALE SURVEYS

### *Pollination rates, seed rain and pollinator activity 2004–2006*

We estimated the proportions of flowers that were triggered by pollinators and measured seed rain at several localities, so that pollination rates could be related to plant fecundity. Data were collected from Lincoln, Palmerston North and Hanmer in the 2004/5 flowering season (late September–early January). In the 2005/6 flowering season, we expanded this survey to seven sites from the central North Island of NZ to the Canterbury Plains in the South Island of NZ that ranged from close to sea level to over 1000-m in altitude (Table 1).

The proportion of flowers triggered by pollinators was estimated according to Parker (1997): At each site, we selected 10 ‘focal’ broom plants at the start of the flowering season and marked a single branch per ‘focal’ plant for subsequent flower counts. Seed rain was estimated at each site using ten *c.* 5-cm-deep, plastic-sided, circular (*c.* 24-cm diameter) litter trays that had a base of plastic mesh sufficiently fine to trap broom seeds. These were placed onto the soil surface beneath the broom plants (one per focal plant). At fortnightly intervals throughout the flowering season:

1. The number of ‘untriggered’ and ‘triggered’ flowers on each selected branch was counted. The overall proportion of triggered flowers, over the whole season, at each site was calculated by taking the ratio of triggered flowers to total flowers after these were summed over all censuses for each plant and then averaged for all ten focal plants.
2. Litter was removed from each litter tray and seeds were counted until all seed pods had dehisced. Aborted flowers were also counted at eight sites (the exceptions were Te Matai road and Lake Rerewhakaitu), which enabled the number of seeds per flower to be calculated for those sites.
3. One of the 10 ‘focal’ plants was selected at random and an observer sat within *c.* 2-m of the plant and recorded visiting insects. Observations were performed for 10 min every hour; over an entire day from dawn (*c.* 7:00 a.m.) to dusk (*c.* 7:00 p.m.) at Lincoln and between 10:00 a.m. and 4:00 p.m. at Palmerston North. At the remaining sites, plants were observed for a minimum of 30 min (in discrete 10 min periods, generally between the hours of 10:00 a.m. and 4:00 p.m.). All potential pollinators that visited broom flowers were recorded, identified (to species, if possible, otherwise to genus or family) and, as in

Simpson *et al.* (2005), a tally of the following pollinator behaviours was recorded:

- a) Untriggered to triggered (U → T). Pollinator landing on an untriggered ‘virgin’ flower, triggering it and foraging.
- b) Triggered to triggered (T → T). Foraging from a previously triggered flower.

### *Biocontrol attack rates and seed rain 2007/8*

The proportions of seeds infested with *B. villosus* were recorded in the summer of 2007/8 in a total of 14 broom stands across a range of habitats on the North and South Islands of NZ (see Table S1, Supporting information). Seed rain was measured at 13 of these sites. Ten mature ‘focal’ plants were haphazardly selected per site and seed rain measured as described for the 2004–2006 sites. Fifteen mature closed pods from each focal plant were collected and stored at room temperature to determine the number of seeds per pod and the proportion of seeds infested with *B. villosus*. Infested seeds were first identified by the presence of an exit hole made by an emerging beetle; apparently uninfested seeds were then crushed, using a scalpel, to identify seeds containing an unemerged beetle.

## ANALYSIS

R 2.9.0 (R Development Core Team) and the nlme library were used for analyses. For all analyses of the broad-scale surveys where multiple years at the same site were present we included site as a random effect in a general linear mixed effects model (unless otherwise stated). For the manipulative experiment an ANOVA was used on the general linear mixed effects model as the design was orthogonal. For the survey data likelihood ratio tests between nested models were conducted to assess significance of fixed effects (using maximum likelihood). Parameter estimates for the best model were obtained using restricted maximum likelihood. General rather than generalized linear mixed effects models were used for proportion and count data due to convergence problems; where necessary transformations were used to ensure assumptions of normality of errors and constancy of variance were justified.

### *Manipulative experiment*

A general linear mixed effects model and ANOVA were used, with a random effect of plant which acted as a blocking effect as all treatments were applied within a single plant, to determine if the proportion of flowers that produced fruit (pods) and the number of seeds produced per pod varied according to the fixed effects of site and treatment for the first year (two sites) and for treatment only in the second year (one site). Proportions were arc-sine transformed prior to analysis to normalize errors. The identity of each focal plant was declared as a random effect.

### *Broad-scale surveys*

General linear mixed effects models were used to determine if the mean rate at which flowers were triggered per minute (square root transformed) varied according to pollinator species. A similar analysis was performed for visits to previously triggered flowers. The proportion of triggered flowers (arcsine transformed) was modelled using the rate that flowers were visited by pollinators (log transformed for linearity). Seed set (log(seeds per flower)) was modelled using the proportion of flowers that were triggered.

Log(seed rain) was modelled as a function of the rate at which flowers were visited by the essential pollinator species (species capable of opening broom flowers – honeybees and bumblebees). Correlation between bumblebee visits (flowers visited per minute) and honeybee visits was tested to investigate whether honeybees and bumblebees potentially competed for resources. Correlation between seed-destruction and seed rain was tested to determine if square root (proportion seed loss) was correlated with seed rain. The effect of island (North or South) on seed rain was tested to determine whether presence of the varroa mite (North Island) directly affected seed rain: the varroa mite spread throughout the North Island of NZ by 2002 (Goodwin 2004) and was first recorded in the South Island of NZ at Nelson in 2006 (MAFBNZ 2008). By September 2008 infested hives had reached the Waimakariri District in north Canterbury, but low mite numbers were not yet impacting honeybee populations (M. Goodwin, pers. comm.). During the present study (2004–2008), therefore, the varroa mite was impacting honeybees throughout the North Island but absent or rare and not impacting honeybees in the South Island sites we surveyed.

#### MODELLING

The broom equilibrium occupancy model developed by Rees & Paynter (1997) was used to explore interactions between pollination and biocontrol under a range of NZ scenarios. The simulation model incorporates key aspects of broom biology including a seed bank, an age-structured established plant population, local density-dependent competition, asymmetric competition between adult plants and seedlings and local seed dispersal (Rees & Paynter 1997). Parameter values were based on published data (for NZ where possible, Table 2) and observed pollination and biocontrol rates from the field surveys were incorporated through a range of fecundity scenarios (assuming biocontrol is independent of seed production). The mean proportion of sites with broom over the last 200 years of each 500 year simulation was used to assess effects of biocontrol and pollination on the equilibrium occupancy of broom.

Predicted control outcomes were based on field data and simulations: we estimated the impact of small-scale disturbance (i.e.

herbicide applications, that kill individual broom plants) on broom populations (assuming intermediate germination and survival in the model,  $g = 0.05$ ,  $s = 0.1$ ) under seed rain scenarios from the 2004–2006 broad-scale survey sites (Table 1). The impact of biocontrol was estimated using the mean biocontrol rate at sites that were in close proximity to beetle release sites (see Results, Table S1, Supporting information; 73%). The potential reduction in fecundity due to the varroa mite was estimated by using the correlation between seed rain vs. pollination rate by honeybees and bumblebees (see Results), predicted with bumblebee pollination visits only to simulate removal of honeybees from the system (by the varroa mite and/or by moving commercial hives).

## Results

#### MANIPULATIVE EXPERIMENT

In the first year the proportion of pods produced per flower significantly varied according to the site:treatment interaction ( $F_{3,63} = 5.0$ ,  $P < 0.004$ ). Natural pollination rates were higher at Lincoln than Palmerston North (18% at Lincoln vs. 6% of flowers produced pods in the control treatment at Palmerston North). At both sites, less than 3% of flowers in the sleeve treatment produced pods, compared with approx. 18–31% of flowers in the hand-pollinated treatments (where sleeving had no effect). Hand pollination had no impact at Lincoln, but resulted in an increase from 6% to 31% in the proportion of flowers that produced pods at Palmerston North (Fig. 1). Similar results were obtained for Palmerston North in the second year, where pollination increased from 9% to 53% with hand pollination (treatment  $F_{3,27} = 69.25$ ,  $P < 0.0001$ ). The number of seeds per seed pod did not vary between sites but did vary according to treatment ( $F_{3,48} = 3.6$ ,  $P < 0.03$  (2004/5);  $F_{3,21} = 4.9$ ,  $P < 0.01$  (2005/6)), with pods in the sleeved treatment producing fewer seeds than the other treatments (4.7 for sleeved treatment vs. 6.5 for other treatments).

**Table 2.** Parameter values used in the broom equilibrium occupancy model

Parameter	Value/s used	Reference/s
$d$ , Seed bank decay rate	0.45	Mean of Sheppard <i>et al.</i> (2002) and Paynter <i>et al.</i> (1998)
$A_{\min}$ , Minimum age for reproduction	2	Williams (1981)
$A_{\max}$ , Max plant age	15	Williams (1981) and Paynter <i>et al.</i> (2003) (approximate $x$ -intercept of NZ model of number of plants $\sim$ age)
$f_h$ , Proportion of seeds retained in parental site	0.6	Rees & Paynter (1997); Buckley (pers. obs.)
$P_{so}$ , Probability a site is suitable for recolonisation after plant death	1	Sheppard <i>et al.</i> (2002)
$P_{dist}$ , Probability of disturbance	0 to 1	n/a
$G$ , Germination rate	0.001, 0.05, 0.2	Range from Paynter <i>et al.</i> (1996), Paynter <i>et al.</i> (1998) and Sheppard <i>et al.</i> (2002)
$s$ , Seedling survival	0.005, 0.1, 0.43	Range from Paynter <i>et al.</i> (1998) and Sheppard <i>et al.</i> (2002)
$F$ , Viable seed rain (seeds $m^{-2}$ )	0–21 416	Current study

BROAD-SCALE SURVEYS

We recorded pollinators making 3337 visits to broom flowers during 2080 min of observations (Table S2, Supporting information). The most common pollinators were honeybees (*A. mellifera*: 70% of all visits), followed by bumblebees (*B. terrestris*: 17.1%) and the native bee *Lasioglossum sordidum* Smith (10.2%). Other insects including hoverflies (Syrphidae) and *B. villosus* were responsible for the remaining 2.7% of visits. Honeybees were only absent from Hanmer in 2005/6, when a beekeeper moved hives to another locality, and Tongariro National Park (Table S3, Supporting information). Bumblebees were absent from Hanmer in 2004/5 and Lake Rerewhakaitu, while the native bee *L. sordidum* was observed at five of the seven sites (Table S3, Supporting information).

Of the floral visits observed, 21.7% were to untriggered flowers that were subsequently triggered by the pollinator. Only honeybees and bumblebees triggered flowers (*L. sordidum* and hoverflies foraged from previously triggered flowers only). Bumblebees triggered a higher proportion of the flowers they visited than honeybees (Table S2, Supporting information). However, honeybees were more abundant than bumblebees and they triggered the most flowers (see below).

POLLINATION RATES, SEED RAIN AND POLLINATOR ACTIVITY

The rate that flowers were triggered (square-root transformed) varied significantly according to the pollinator:year interaction ( $LR_1 = 6.5, P < 0.02$ ). Honeybees triggered significantly more flowers per minute than bumblebees in 2004/5 but not in 2005/6 (Fig. 2a,b). Visits to previously triggered flowers also varied significantly between the three main pollinators, ( $LR_2 = 15.8, P < 0.001$ ), but year was not significant and honeybee visitation rates in both years were much higher than bumblebee and native bee rates (Fig. 2c).

The average proportion of triggered flowers was just under 40% and varied by an order of magnitude, ranging from less than 8% of flowers at Hanmer (in 2005/6) to c. 80% at Lincoln (Table S4, Supporting information). The proportion of triggered flowers was closely correlated with visits by effective pollinators (the number of visits that triggered flowers plus the number of visits to already triggered flowers per minute by honeybees and bumblebees) ( $LR_1 = 11.5, P < 0.001$ , Fig. 3a). The mean number of seeds produced per flower at each site was closely correlated with the proportion of flowers that were triggered ( $LR_1 = 5.6, P < 0.02$ ; Fig. 3b).

Broom seed rain, which averaged 3846 seeds  $m^{-2}$ , ranged from just 59 to 14 443 seeds  $m^{-2}$  in the 2004–2006 surveys (Table S3, Supporting information) and was strongly correlated with the overall pollination rate by honeybees and bumblebees ( $LR_1 = 25.0, P < 0.0001$  Fig. 4). Bumblebee visitation rate was uncorrelated with honeybee visitation rate ( $LR_1 = 1.14, P < 0.3$ ).

SEED RAIN AND SEED-DESTRUCTION RATES 2007/8

The seed rain in 2007/8 was similar to the previous field seasons and ranged from just 175 seeds  $m^{-2}$  to 21 416 seeds  $m^{-2}$ . *Bruchidius villosus* was well established at 11 of the 14 sites studied (Table S1, Supporting information). Seed infestation averaged 46% overall, ranging between 0% and 49% on the North Island and between 7% and 84% at South Island sites. An average infestation rate of 73% was observed at localities close to release sites (Table S1, Supporting information). Viable seed rain (after biocontrol) ranged between 87 and 9069 seeds  $m^{-2}$ . Seed- destruction (square root transformed) and seed rain were uncorrelated ( $LR_1 = 0.29, P < 0.6$ ).

Over all seasons (2004–2008), seed rain averaged 4175 seeds  $m^{-2}$  and was somewhat lower in the North Island (varroa mite present, mean seed rain 2131 seeds  $m^{-2}$ ) than in the South Island (varroa mite absent, mean seed rain 5747 seeds  $m^{-2}$ ), however the difference was not statistically significant ( $LR_1 = 2.19, P < 0.2$ ).

MODELLING

Our simulations indicate that, assuming intermediate seedling germination and survival (5% germination and 10% survival, respectively; Table 2) broom abundance in NZ populations is unlikely to decline without additional control effort unless viable seed rain is brought below 70 seeds  $m^{-2}$ . With the exception, therefore, of Tongariro National Park, where honeybees were absent, seed rain exceeded this threshold at all sites during both the 2007/8 and the initial 2004–2006 surveys (Tables S3 & S4, Supporting information). Therefore, apart from Tongariro National Park, broom pollination was high enough to drive broom invasion, even in the North Island sites where the varroa mite was present. Moreover, only very high levels of disturbance were predicted to successfully control most broom populations (Table S4, Supporting information).

Our simulations also indicate that a 73% reduction in seed set due to *B. villosus* is insufficient to affect broom occupancy at the 2004–2006 study sites, although it should reduce the

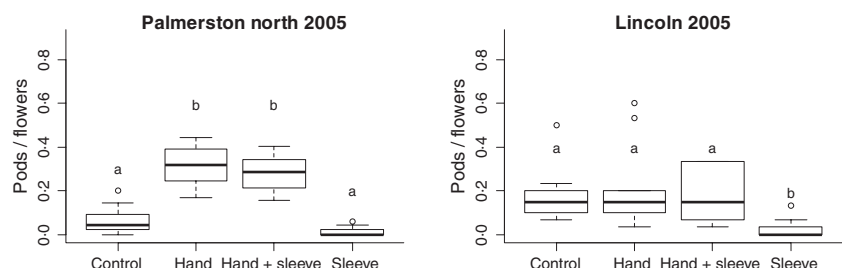
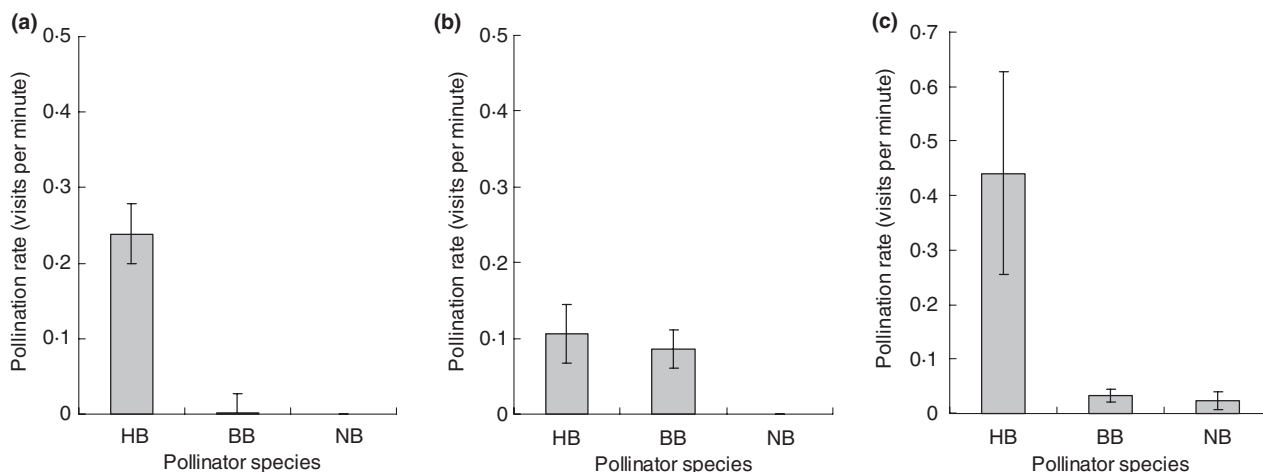


Fig. 1. Proportion of flowers that produced pods at Palmerston North and Lincoln in 2005. Within each site, a different letter between columns indicates a significant difference between the proportion of flowers that formed pods (Tukey HSD).



**Fig. 2.** The mean number of floral visits per minute ( $\pm$  SE) by the three most common pollinator species recorded during the 2004–2006 broad-scale surveys: (a) Mean number of untriggered flowers triggered (U  $\rightarrow$  T) in 2004/5 and (b) 2005/6, and (c) visits to flowers that had previously been triggered (T  $\rightarrow$  T; both seasons combined). HB = honeybee *Apis mellifera*; BB = bumblebee *Bombus terrestris*; NB = native bee *Lasiosglossum sordidum*.

threshold disturbance levels required to achieve control (Table S4, Supporting information; Fig. 5).

When we modelled the potential impact of an absence of honeybees, however, broom was predicted to persist at only four of the seven 2004–2006 study sites. Furthermore, an absence of honeybees and 73% seed destruction by *B. villosus* was predicted to result in eradication of broom from all but two of the 2004–2006 study sites, and to greatly reduce the threshold disturbance required for control at those remaining sites (Table S4, Supporting information; Fig. 5).

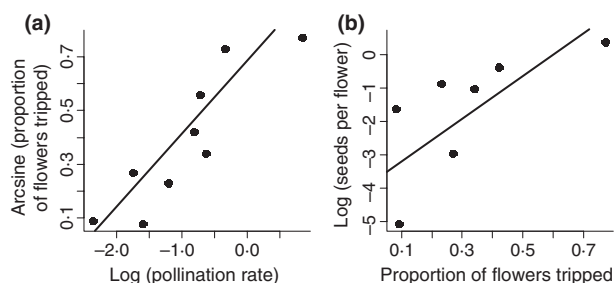
## Discussion

### CONTRIBUTIONS OF BUMBLEBEES AND HONEYBEES TO BROOM POLLINATION

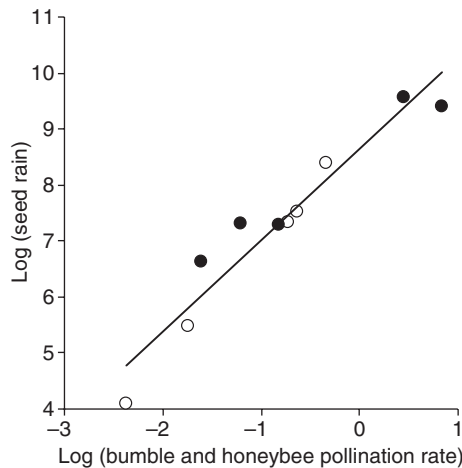
Our study concurs with past studies (Parker 1997; Stout 2000; Suzuki 2000; Simpson *et al.* 2005), showing that apid bees are essential broom pollinators. As in Australia (Simpson *et al.* 2005), introduced bees and broom form an invasive pollinator mutualism in NZ. Honeybees were clearly the most important

pollinators due to their higher abundance, although we may have slightly underestimated bumblebee pollination because pollinator observations were conducted between 10:00 and 16:00 h at most sites: At Lincoln, where observations were conducted from dawn to dusk, bumblebee activity relative to honeybees was highest in the early morning and late evening (Fig. S1, Supporting information).

Pleasants (1981) documented an increase in bumblebees, following honeybee decline in Colorado USA that was attributed to competitive release. Similarly, the appearance of bumblebees following honeybee removal at Hanmer in 2005/6 (Table S2, Supporting information) may have been due to competitive release, although their presence only partially compensated for the absence of honeybees. Moreover, we found no evidence that bumblebee and honeybee abundance were negatively correlated in 2004 to 2006. Therefore, given the relative abundance of honeybees and bumblebees, one might predict that broom seed rain should decline if the varroa mite reduces honeybee abundance in NZ. Our seed rain data, however, were inconclusive. Over all field seasons, seed rain in the North Island of NZ was only 37% of that in the South Island, but this difference was not statistically significant. Before the varroa mite established, feral hives were estimated to account for approximately one third of NZ honeybee hives (200 000 feral vs. 400 000 managed hives; Donovan 2007). Beekeepers control varroa mites using acaricides, so that broom plants nearby commercial hives are unlikely to experience pollination reduction to the same extent as those pollinated largely by feral honeybees. The varroa mite has apparently had a major impact at Tongariro National Park (Table S4, Supporting information), where commercial hives are forbidden, (Murphy & Robertson 2000). However, the presence of commercial beehives continues to drive broom pollination over much of the North Island of NZ, which has an overall density of 1.57 commercial beehives per km<sup>2</sup>, compared with 0.9 hives per km<sup>2</sup> on the South Island (Anon 2009). The varroa mite may have a bigger impact on broom pollina-



**Fig. 3.** (a) Proportion of flowers triggered vs. pollination rate (floral visits per minute; honeybees and bumblebees combined); (b) Seeds produced per flower vs. proportion of flowers triggered by pollinators recorded during the 2004–2006 broad-scale surveys. Pollination rate = number of visits that triggered flowers + number of visits to previously triggered flowers per minute.



**Fig. 4.** Log(seed rain (seeds  $m^{-2}$ )) vs. log(pollination rate by honeybees and bumblebees combined) for all sites in the 2004–2006 broad-scale surveys (North Island sites = open circles, South Island sites = filled circles; site as a random effect). Pollination rate = visits that triggered flowers + visits to previously triggered flowers per minute. Fixed effects:  $y \sim 8.66 + 1.68x$ . Random effect: sd about the intercept due to site = 0.29, residual sd = 0.32.

tion when it reaches the South Island high country, where most broom infestations occur (Jarvis *et al.* 2006) and most honeybees are feral (Simpson 2003).

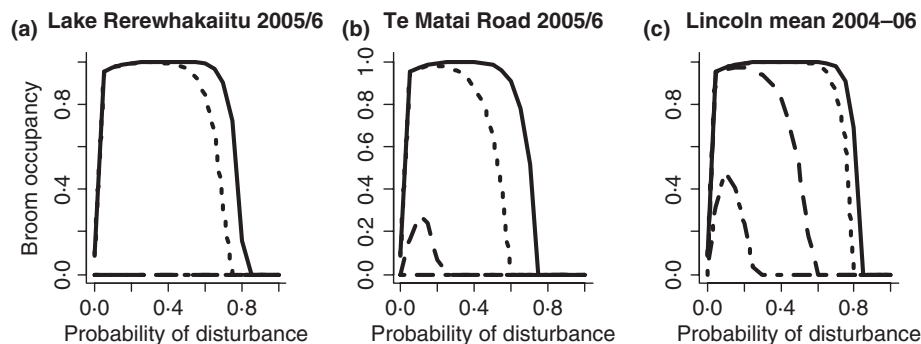
IMPACT OF THE SEED-FEEDING BIOCONTROL AGENT

*Bruchidius villosus* was well established at most sites. Although low attack was recorded at several upland sites, this most likely reflects past release effort: Harman (1999) compared the synchronization of *B. villosus* with broom flowering at lowland (early flowering) and upland (late flowering) sites and concluded that *B. villosus* is adapted to the variable phenology of its host. *Bruchidius villosus* was ini-

tially established at a few lowland sites in the South Island and only become sufficiently abundant for field collection and widespread nationwide redistribution to commence in the 1993–1994 field season (Syrett *et al.* 2000). Releases continued until 2002 and *B. villosus* is still expanding in range and abundance in NZ (L.M. Hayes, pers. comm.). Therefore, there is no reason to suppose that, nationwide, average infestation rates will not eventually reach *c.* 73% (the average rate observed at localities adjacent to release sites; Table S1, Supporting information). Moreover, no evidence of a relationship between seed rain and proportion seed loss was found (Fig. S2, Supporting information), indicating that seed-destruction by *B. villosus* was independent of seed rain.

MODELLING THE IMPACTS OF REDUCED POLLINATION AND BIOCONTROL

Previous manipulative experiments demonstrated that broom fecundity was pollinator-limited in both the USA and Australia (Parker 1997; Simpson *et al.* 2005). Similarly, the significant *c.* 5-fold increase in pod production following our hand pollination treatments demonstrated that fecundity of broom plants was pollinator limited at Palmerston North. Although there may have been pollinator saturation at Lincoln, where hand pollination did not result in increased fecundity, the broad-scale surveys revealed that the huge variation in broom seed set in NZ was explained by the visitation rate of introduced pollinators, implying that fecundity was pollinator limited at many sites. Nevertheless, although broom seed rain was generally rather lower (Tables S3 & S4, Supporting information) than that assumed typical by Rees & Paynter (1997; 22 400 seeds per plant, equivalent to *c.* 9000 seeds  $m^{-2}$ ) our models concur with Rees & Paynter (1997) by predicting that *B. villosus* alone is unlikely to control broom populations in NZ. In contrast, our models predict that an absence of honeybees could drive some broom populations to extinction, while an



**Fig. 5.** Simulated broom occupancy at three sites in New Zealand under four seed rain scenarios: (1) honeybee pollination, no biocontrol (solid line); (2) honeybee pollination and 73% seed destruction by *Bruchidius villosus* (dotted line); (3) no honeybee pollination, no biocontrol (dashed line); and (4) no honeybee pollination and 73% seed destruction by *Bruchidius villosus* (dot and dash line). Seed rain without honeybees was estimated by using the linear mixed effects model of seed rain vs. honeybee plus bumblebee pollination rate (Fig. 4), parameterised with bumblebee pollination values only. (a) Lake Rerewhakaitu (North Island) 2005/6 (4437 seeds  $m^{-2}$  with HB; 0 seeds  $m^{-2}$  without HB; 1198 seeds  $m^{-2}$  with HB + biocontrol; 0 seeds  $m^{-2}$  without HB + biocontrol). (b) Te Matai Road (North Island) 2005/6 (1542 seeds  $m^{-2}$  with HB; 84 seeds  $m^{-2}$  without HB; 416 with HB + biocontrol; 23 seeds  $m^{-2}$  without HB + biocontrol). (c) Lincoln (South Island) average of 2004/5 and 2005/6 (13301 seeds  $m^{-2}$  with HB; 349 seeds  $m^{-2}$  without HB; 3591 seeds  $m^{-2}$  with HB + biocontrol, 94 seeds  $m^{-2}$  without HB + biocontrol). Full parameter details for the simulation model are listed in Table 2.

absence of honeybees in combination with, *B. villosus* can further reduce seed rain to levels where the majority of broom populations go extinct (Table S4, Supporting information), indicating the pollinator disruption can enhance the impact of biocontrol.

#### IMPLICATIONS FOR MANAGING INVASIVE SCOTCH BROOM POPULATIONS

Our simulations demonstrate that broom seed rain is currently sufficient to drive broom invasions over much of NZ, even in the presence of the varroa mite and a seed-feeding biocontrol agent. Other management options are, therefore, required and improved management of commercial beehives (for example, through the withdrawal of licences for beekeepers to locate hives on Department of Conservation land) may be the key to successful integrated management of broom at many sites in NZ.

#### IMPLICATIONS FOR MANAGING THE VARROA MITE

The varroa mite is predicted to have substantial detrimental economic impacts on the NZ economy. In the South Island of NZ, 90% of this impact was assumed to fall on the pastoral industries, because clover nitrogen fixation may decline as a result of reduced pollination (Simpson 2003).

White clover *Trifolium repens* L. and ryegrass *Lolium perenne* L. form the basis of the NZ pastoral agricultural industry (Sarithchandra *et al.* 2001). Barrett & Silander (1992) found that, in US pastures, annual white clover seedling recruitment (3.5 seedlings m<sup>-2</sup>) was several orders of magnitude lower than vegetative recruitment (14 800 ramets m<sup>-2</sup>). If seedling establishment is equally unimportant in NZ, then it is inconceivable that white clover populations should become limited by the varroa mite. Moreover, red clover *Trifolium pratense* L. and subterranean clover *Trifolium subterraneum* L., which are planted in drier pastures than white clover (Johnstone & McLean 1987; Ledgard & Steele 1992) are, respectively, bumblebee pollinated (Hanley & Goulson 2003) and capable of self-fertilization (Khan *et al.* 1994). The costs of the varroa mite may, therefore, be significantly lower than anticipated. Moreover, honeybees largely rely on introduced plants for pollen and nectar in NZ (Hanley & Goulson 2003) and, as shown here, are major contributors to the problems caused by broom. The costs of the varroa mite in NZ may potentially be offset by previously unrecognized benefits arising from reduced weed pollination.

Due to a lack of evidence, the impact that honeybees can have on weed infestations in NZ has not been fully appreciated. For example, Lindsay (2004) recommended planting, or siting hives next to, a range of pollen sources, including major weeds such as thistles, blackberry, broom and gorse *Ulex europaeus* L. Gorse has a similar pollination mechanism to broom and may also rely on honeybee pollination (Hanley & Goulson 2003). MacFarlane, Grindell & Dugdale (1992) noted that on the Chatham Islands, where bumblebees were absent and feral honeybees were rare, substantial gorse seed set

only occurred in close proximity to beehives. More research and debate is required regarding varroa mite management options in NZ because an absence of feral honeybees presents an opportunity for better targeted use of managed honeybee hives: providing pollination where they are needed and exclusion from areas where they are not required and may cause weed problems. Management options that strive to breed varroa mite-resistant bees (e.g. Harbo & Harris 1999) may re-establish feral honeybee populations in NZ and squander that opportunity.

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#### References

- Anon (2009) *Beekeeping in New Zealand, Part 1*. Downloaded from: <http://www.nba.org.nz/docs/Part1.pdf>, Accessed 19 November 2009.
- Barrett, J.P. & Silander, J.A. (1992) Seedling recruitment limitation in white clover (*Trifolium repens*, Leguminosae). *American Journal of Botany*, **79**, 643–649.
- Barthell, J.F., Randall, J.M., Thorp, R.W. & Wenner, A.M. (2001) Promotion of seed set in yellow star-thistle by honeybees: evidence of an invasive mutualism. *Ecological Applications*, **11**, 1870–1883.
- Donovan, B.J. (2007) Apoidea (Insecta: Hymenoptera). *Fauna of New Zealand*, **57**, pp. 295. Manaaki Whenua Press, Landcare Research, Lincoln, New Zealand.
- Goodwin, M. (2004) Introduction and spread of varroa in New Zealand. *Bee World*, **85**, 26–28.
- Hanley, M.E. & Goulson, D. (2003) Introduced weeds pollinated by introduced bees: cause or effect? *Weed Biology and Management*, **3**, 204–212.
- Harbo, J.R. & Harris, J.W. (1999) Selecting honey bees for resistance to *Varroa jacobsoni*. *Apidologie*, **30**, 183–196.
- Harman, H.M. (1999) The effect of variability in the phenology of the reproductive stages of Scotch broom (*Cytisus scoparius*) on the synchronization of the life stages of broom seed beetle (*Bruchidius villosus*) in New Zealand. *Biological Control*, **15**, 228–234.
- Jarvis, P.J., Fowler, S.V., Paynter, Q. & Syrett, P. (2006) Predicting the economic benefits and costs of introducing new biological control agents for Scotch broom *Cytisus scoparius* into New Zealand. *Biological Control*, **39**, 135–164.
- Johnstone, G.R. & McLean, G.D. (1987) Virus disease of subterranean clover. *Annals of Applied Biology*, **110**, 421–440.
- Khan, M.R.I., Tabe, L.M., Heath, L.C., Spencer, D. & Higgins, T.J.V. (1994) Agrobacterium-mediated transformation of subterranean clover (*Trifolium subterraneum* L.). *Plant Physiology*, **105**, 81–88.
- Ledgard, S.F. & Steele, K.W. (1992) Biological nitrogen fixation in mixed legume/grass pastures. *Plant and Soil*, **141**, 137–153.
- Lindsay, F. (2004) Choosing a pollen site. *New Zealand Beekeeper*, **12**, 19.
- MacFarlane, R.P., Grindell, J.M. & Dugdale, J.S. (1992) Gorse on the Chatham Islands: seed formation, arthropod associates and control. *Proceedings of the 45th New Zealand Plant Protection Conference*, pp. 251–255.
- Ministry of Agriculture and Forestry Biosecurity New Zealand (MAFBNZ). Varroa mite website, accessed January 2009, last updated 7 October 2008.
- Murphy, C. & Robertson, A. (2000) Preliminary study of the effects of honey bees (*Apis mellifera*) in Tongariro National Park. *Science for Conservation*, **139**, 18.
- Parker, I.M. (1997) Pollinator limitation of *Cytisus scoparius* (Scotch broom), an invasive exotic shrub. *Ecology*, **78**, 1457–1470.
- Paynter, Q., Fowler, S.V., Hinz, H.L., Memmott, J., Shaw, R., Sheppard, A.W. & Syrett, P. (1996) Are seed-feeding insects of use for the biological control of broom? *Proceedings of the IX International Symposium on Biological Control of Weeds, 19–26 January 1996* (eds V.C.

- Moran & J.H. Hoffman), pp. 495–501. University of Cape Town, Stellenbosch, South Africa.
- Pleasants, J.M. (1981) Bumblebee response to variation in nectar availability. *Ecology*, **62**, 1648–1661.
- Primack, R.B. (1983) Insect pollination in the New Zealand mountain flora. *New Zealand Journal of Botany*, **21**, 317–333.
- Rees, M. & Paynter, Q. (1997) Biological control of Scotch broom: modelling the determinants of abundance and the potential impact of introduced insect herbivores. *Journal of Applied Ecology*, **34**, 1203–22.
- Sarathchandra, S.U., Ghani, A., Yeates, G.W., Burch, G. & Cox, N.R. (2001) Effect of nitrogen and phosphate fertilisers on microbial and nematode diversity in pasture soils. *Soil Biology and Biochemistry*, **33**, 953–964.
- Sheppard, A.W., Hodge, P., Paynter, Q. & Rees, M. (2002) Factors affecting invasion and persistence of broom *Cytisus scoparius* in Australia. *Journal of Applied Ecology*, **39**, 721–734.
- Simberloff, D. & Von Holle, B. (1999) Positive interactions of nonindigenous species: invasional meltdown? *Biological Invasions*, **1**, 21–32.
- Simpson, B. (2003) Summary of the assessments of the economic impact of varroa in the South Island of New Zealand. Biosecurity New Zealand, Ministry of Agriculture and Forestry, New Zealand.
- Simpson, S.R., Gross, C.L. & Silberbauer, L.X. (2005) Broom and honeybees in Australia: an alien liaison. *Plant Species Biology*, **7**, 541–548.
- Stokes, K.E., Buckley, Y.M. & Sheppard, A.W. (2006) A modelling approach to estimate the effect of exotic pollinators on exotic weed population dynamics: bumblebees and broom in Australia. *Diversity and Distributions*, **12**, 593–600.
- Stout, J.C. (2000) Does size matter? Bumblebee behaviour and the pollination of *Cytisus scoparius* L. (Fabaceae). *Apidologie*, **31**, 129–139.
- Suzuki, N. (2000) Pollinator limitation and resource limitation in the Scotch Broom, *Cytisus scoparius* (Leguminosae). *Plant Species Biology*, **15**, 187–193.
- Syrett, P., Sheat, J.J., Harman, H.M., Harris, R.J., Hayes, L.M. & Rose, E.A.F. (2000) Strategies for achieving widespread establishment of broom seed beetle, *Bruchidius villosus* (Coleoptera: Chrysomelidae), a biological control agent for broom, *Cytisus scoparius*, in New Zealand. *Proceedings of the X International Symposium on Biological Control of Weeds, 4–14 July 1999* (ed. N.R. Spencer), pp. 761–771. Montana State University, Bozeman, MT, USA.
- Thomas, M.B. & Reid, A.M. (2007) Are exotic natural enemies an effective way of controlling invasive plants? *Trends in Ecology and Evolution*, **22**, 447–453.
- Zhang, Z.-Q. (2000) Notes on *Varroa destructor* (Acari: Varroidae) parasitic on honeybees in New Zealand. *Systematic and Applied Acarology Special Publications*, **5**, 9–14.

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## Supporting Information

Additional Supporting Information may be found in the online version of this article:

**Table S1.** Mean seed rain and seed-destruction rates by the seed-feed-in biocontrol agent *B. villosus* at 14 sites across NZ.

**Table S2.** Summary of insects observed visiting *C. scoparius* flowers.

**Table S3.** Summary of pollination by the most common insect visitors to broom flowers.

**Table S4.** Percentage of flowers triggered and seed rain (seeds m<sup>-2</sup>) recorded during the 2004–2006 broad-scale surveys.

**Fig. S1.** Mean activity patterns of honeybees and bumblebees recorded at Lincoln.

**Fig. S2.** Seed-destruction vs. seed rain for 2007/8 study sites.

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