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Weed Biocontrol

WHAT'S NEW?

Contents

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COVER IMAGE:
Chilean flame creeper in flower.

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Chilean Flame Creeper Beetle Passes the Test

Chilean flame creeper (*Tropaeolum speciosum*) is a climbing vine that has become an important environmental weed in Southland, Otago and Canterbury. It is present in the lower regions of the North Island, but it is only considered a minor weed there. A biological control programme was initiated in 2021 following the serendipitous discovery of a leaf beetle (*Blaptea elguetai*) on Chilean flame creeper in Chile in 2019.

Following preliminary work on the beetle in Chile, our collaborator Hernán Norambuena travelled to New Zealand in November 2022 with a precious cargo of around 65 reproductive adult beetles and 450 eggs. Host range testing began immediately, and the results were mostly very promising. However, there remained a question about the potential risk to one species in the cabbage family (Brassicaceae), pak choi dark dragon (*Brassica chinensis*).

In the initial tests a few larvae nibbled on the leaves of pak choi, and one individual completed development to the adult stage. We then conducted an analysis using the Relative Performance Index developed by Senior Researcher Quentin Paynter. The index helps us predict whether low levels of feeding on non-target test species in the artificial testing arena are likely to translate to serious non-target feeding in the natural environment. A score of 0 means the candidate agent did poorly on the test (non-target) species compared to how it did on the target weed. A score of 1 means the candidate agent did equally well on the test species and on the target weed. A score below a certain threshold corresponds with no non-target feeding in the natural environment, whereas a score above the threshold means there is a realistic likelihood for non-target feeding to occur in the natural environment.

The score result from the initial testing for pak choi was below the threshold – but close to it. We decided that more testing was required to increase our confidence that this important vegetable was going to be safe. "Luckily, we discovered that the beetles remain active and reproductive if maintained at warm temperature, and do not require overwintering. This enabled us to run additional tests on pak choi with no delays," explained Ronny Groenteman, who oversees this programme. "We ran a great deal more replicates, and at the end of this set of tests we were confident that pak choi was not going to be a field host for the beetle," she added.

Robyn White, who conducted the additional testing, commented that "Only a handful of larvae attempted nibbling on pak choi. All but one larva died as first instar without feeding at all or after a small nibble, and only one larva made it to second instar before dying. No larvae made it beyond second instar on pak choi, while 42% of replicates on Chilean flame creeper had at least one larva make it to pre-pupal stage, and 22% of replicates had at least one larva make it to adult."

In the meantime, we had asked Hernán to complete two more tasks to give us further reassurance that brassica crops are safe from attack by the leaf beetle. Firstly, we asked him to search the Chilean grey literature, such as farming magazines and information pamphlets, to look for any reports of the beetle in association with brassica crops. Secondly, we asked him to visit field sites where unsprayed brassica crops are grown near Chilean flame creeper sites, where the beetles are also present.

Fortunately, both these tasks yielded the results we were hoping for. Hernán located numerous literature items associated with the main brassica crop in Chile, rape seed (*Brassica napus*; cultivated for oil), and other brassica crops such as cabbage and cauliflower. Despite brassica crops being grown in Chile frequently, even as far back as the 19th century, the literature did not mention the leaf beetle as a pest. "I have

Chilean flame creeper

also consulted with several entomologists, agronomists and growers of rape seed or other brassica crops, and they all confirmed they are not aware of beetles of the distinct shape and colours of the Chilean flame creeper beetle ever seen in association with these crops," explained Hernán.

In his field surveys Hernán visited large fields of unsprayed rape seed crops in Chile's Araucania region, as well as other smaller fields of *Brassica* crops such as cabbage, cauliflower, kale, arugula, and radish. He also surveyed field mustard plants including those found at the Chilean flame creeper site where the beetle was first detected in 2019. "The region of the surveys is currently under a state of emergency due to indigenous rights land disputes. Nevertheless, it was the most logical region to survey since there are sites with the highest early records of the leaf beetle, and is the largest rape seed cropping area" said Hernán.

The crops' distance from the nearest Chilean flame creeper with the leaf beetle ranged between 30 m and 25 km. The beetle was absent from all surveyed brassica crops. Other insects were present in those crops, such as honeybees, pests such as aphids, leaf miners, and the diamond back moth, and natural enemies such as parasitic wasps, hoverflies, and ladybird beetles. This survey serves as another level of evidence for the host specificity of the Chilean flame creeper leaf beetle.

With the host range testing, literature search, and field surveys all painting a consistent picture that the leaf beetle is host specific to species in the plant genus *Tropaeolum*, we are finally preparing for Māori and stakeholder consultation, and writing up the release application for the Environmental Protection Authority.

This project was funded by the Ministry for Primary Industries' Sustainable Food and Fibre Futures Fund [Grant S3F20095] and the National Biocontrol Collective.

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Welcome Luise Schulte

We are delighted to welcome Luise Schulte to the Biocontrol & Molecular Ecology team at MWLR. Luise joined MWLR in September 2023 as a researcher – plant pathology and is based at our Tamaki site in Auckland. She will mostly be working on plant pathogens as biocontrol agents for invasive weeds in New Zealand.

Luise has a background in molecular biology and environmental ecology, with experience in genetics, data analysis and fieldwork. She has spent time in Siberia, Burkina Faso, and Ivory Coast, working in multinational teams. She completed her PhD in 2022, at the Alfred Wegener Institute, Helmholtz Centre for Polar and Marine Research, in Potsdam, Germany. For her research she extracted ancient DNA, up to 50,000 years old, from lake sediment cores. This was used to unravel the glacial history of larch forests and how they colonised Siberia after the last glacial maximum. Following her PhD, Luise worked as a postdoctoral researcher at the Robert Koch Institute in Berlin, the public health institute of Germany. Here she contributed to a project analysing infectious diseases in West African countries and sequencing selected pathogens.

At MWLR Luise is currently working on the Chilean needle grass rust fungus (*Uromyces pencanus*) and is analysing data from the rural decision-makers surveys about future weeds of the productive sector. Luise is also developing informational material on weed biocontrol, using terminology and concepts from Māori world views to ensure they are meaningful and relevant to holders of te ao Māori. The purpose of these resources is to support Māori participation in decision-making processes and to facilitate more effective communication and engagement between councils and iwi and hapū involved in weed biocontrol consultations.

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Luise Schulte

Yellow Flag Iris Flea Beetle Touches Down

With the new year a new candidate biocontrol agent arrived at our insect containment facility at Lincoln. Angela Bownes's Christmas holiday in South Africa presented the opportunity for 34 adults of the yellow flag iris flea beetle (*Aphthona nonstriata*) to be hand-carried to New Zealand. The beetles are being reared by the Centre for Biocontrol at Rhodes University in South Africa and are undergoing evaluation for their potential as a biocontrol agent for yellow flag iris (*Iris pseudacorus*).

Yellow flag iris is native to Europe and northern Africa but was exported around the world because of its attractive yellow flowers. It became invasive in several regions in the southern hemisphere, including in New Zealand, Australia, Argentina, and South Africa. It can spread by seed and vegetative structures, forming thick networks of rhizomes in and around waterways. Yellow flag iris infestations negatively affect native biodiversity by displacing desirable plants, can alter the flow of waterways contributing to flooding, and invade paddocks and parks, disrupting agricultural and recreational activity. It is difficult to control due to the issues relating to herbicide use near water, and because manual removal is labour intensive and any overlooked rhizomes regenerate rapidly. In New Zealand yellow flag iris is a National Pest Plant Accord species, which means it is an unwanted organism under the Biosecurity Act 1993. Consequently, this plant cannot be propagated, distributed or sold in New Zealand.

We are fortunate that South Africa had already started developing a biocontrol programme for yellow flag iris. Rhodes University, along with Vrije Universiteit in Amsterdam, undertook surveys of yellow flag iris in parts of the native range and identified three candidate biocontrol agents. The flea beetle was regarded as the most promising option, and host specificity testing has largely been completed at Rhodes University, with encouraging results.


Yellow flag iris beetle


Yellow flag iris in waterway

The flea beetle is a tiny beetle that jumps when disturbed. The adults nibble on yellow flag iris leaves, preferring fresh tips. Eggs are laid in the soil and larvae feed on the rhizomes, causing extensive damage. Host specificity testing has shown that the flea beetle is an iris specialist, being able to feed and complete development on several species in the *Iris* genus.

New Zealand's closest native relatives to yellow flag iris are species in the genus *Libertia*. While *Libertia* species are phylogenetically distant from yellow flag iris, it was important to test representatives of this genus to confirm the flea beetle does not pose a risk to these native plants. Now that the flea beetle is being reared in containment at Lincoln we are confident we will be able to complete enough testing to determine whether it is sufficiently host specific for release in New Zealand.

This project is funded by the National Biocontrol Collective and the Ministry for Primary Industries' Sustainable Food and Fibre Futures Fund (Grant #20095) on multi-weed biocontrol.

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A Flying Start for a Bud-Galling Wasp

We are excited to report that our most recently introduced biocontrol agent, the Sydney golden wattle bud-galling wasp (*Trichilogaster acaciaelongifoliae*), has shown positive signs of establishment following its release last summer. This tiny wasp lays its eggs inside the flower and vegetative buds of Sydney golden wattle. Feeding by the wasp larvae leads to the formation of large growths (galls) instead of flowers (which would go on to produce seeds) and shoots (that increase plant biomass). The wasp was released at two sites in the Manawatū-Whanganui region in December 2022, shortly after gaining approval from the Environmental Protection Authority and the Ministry for Primary Industries to remove the wasps from containment.

In August 2023 enough time had passed to detect the development of galls at the release sites, and the team was pleasantly surprised to find three galls at their very first stop. "It was wonderful to see galls for the first time in New Zealand, suggesting a successful first release of the bud-galling wasp. This was a promising first sign that the wasp has great potential as a biocontrol agent for Sydney golden wattle here," said Angela Bownes, who made the first release of the wasp with Horizons Regional Council staff and Research Associate Richard Hill.

A second consignment of galls was collected and hand-carried to New Zealand in November 2023 by Fiona Impson, a world expert on weedy wattles from the University of Cape Town, South Africa. For biosecurity reasons it is not possible


First galls sighted on Sydney golden wattle at release site in early summer


Numerous emergence holes on an imported gall


Jack releasing wasps at Waitārere Beach

to remove the galls infested with wasp larvae and pupae from containment. Instead, freshly emerged wasps that have chewed their way out of the galls are collected daily for release. For each consignment, independent experts carry out rigorous testing, including identification of the wasp to confirm we are releasing the correct species, and disease testing to ensure the wasps are healthy and disease free, thus preventing the inadvertent release of any harmful organisms into the New Zealand environment.

Because the adult wasps only live for a few days, timing is critical to ensure these tests are completed in good time to gain MPI approval to remove the wasps from containment as quickly as possible. Galls in the second consignment from South Africa yielded 180 wasps, which were again released in Manawatū-Whanganui with the help of Horizons Regional Council staff Jack Keast and Robbie Sicely.

A third shipment of galls was planned in early December with the help of John Hoffman (University of Cape Town), but unfortunately this never made it to New Zealand due to issues with the transport of live insects by the only available airline at the time. "It turned out to be a very disappointing situation because we wanted to release more wasps in new areas this summer," said Arnaud Cartier, who has overseen the process to get the wasps out of containment since the start of this project.

To avoid such disappointment again, the team will opt to hand-carry galls to New Zealand next summer to ensure high numbers of wasps are available for release in other invaded regions such as Northland and Matakana Island.

This project is funded by the National Biocontrol Collective and the Ministry for Primary Industries' Sustainable Food and Fibre Futures Fund (Grant #20095) on multi-weed biocontrol.

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Variability in Herbivory Shows Patterns, and it Matters

Interactions between plants and herbivores are central to most ecosystems, but the high variability of these interactions makes it difficult to determine exactly how herbivores influence plant response to herbivory pressures. A group of researchers from Michigan State University set out to explore the factors that influence the variability in herbivory within plant populations: can any patterns be detected?

This was no small feat! The group designed a survey protocol that would be simple enough so that it could be replicated consistently by researchers around the world, yet informative and flexible enough to accommodate a variety of field situations that are different from the 'ideal standard' plant population.

Early in 2020 MWLR scientist Quentin Paynter was contacted by the Michigan researchers and invited to take part in this global initiative to represent New Zealand flora. "When I first read the study hypotheses and design, I could immediately see that these research questions would be useful for us to understand on a global scale, and that they would have implications for weed biocontrol," said Quentin. "Herbivory puts selection pressure on plants and how their defence mechanisms evolve, which is important for host specificity. Understanding patterns of variability in herbivory can potentially help us improve our predictions about biocontrol agent effectiveness," he added.

Quentin joined forces with colleague Ronny Groenteman, and together they planned a survey to represent New Zealand in what became the Herbivory Variability Network (HerbVar for short). "Although we think about weeds for most of our working day, we had to instead think about indigenous plants for this survey, because we were looking for natural patterns of herbivory," said Ronny. "Our herbivorous biocontrol agents are operating in a simplified enemy-free environment in New Zealand, and this could skew the variability patterns they display. That is why we focused on native ecosystems," she explained.


Surveying vegetable sheep

Each contributor was asked to survey at least five plant species according to the following hierarchical structure:

- at least one of the two focal species, the cosmopolitan dandelion (*Taraxacum officinale)* and broad-leaf plantain (*Plantago major)*
- at least one species (preferably more) belonging to at least one of the five focal plant families Apocynaceae, Asteraceae, Fabaceae, Rubiaceae, and Solanaceae
- any species that have not yet been sampled, preferably adding plant families that have not yet been sampled by other participants, and/or growth forms that have not yet been represented within the five focal plant families.

With our weird and wonderful New Zealand flora we were able to grant the survey a few funky representations, such as vegetable sheep (*Raoulia eximia*) and a tree daisy (*Olearia paniculata*). This last species was surveyed in collaboration with other researchers in the network, Lauren Walker (Ministry of Primary Industries) and Warwick Allen. Warwick has since joined the Ecosystems & Conservation Team here at MWLR.

We timed our surveys for autumn to maximise the chances of capturing footprints of herbivory. The autumn of 2020 was a bit more eventful than we anticipated, and we did most of our surveys in the week following the first confirmed case of Covid in New Zealand. The walking tracks used to access our selected plant populations, which would have been tourist highways at any other time, were all but abandoned and almost eerie, and we completed the surveys 2 days before the country went into its first lockdown. Talk about timing!

All in all, 127 research teams in 34 countries surveyed a total of 790 plant populations covering 503 plant species from 135 plant families. Survey sites spanned 116° of latitude across North America, Central America, South America, Europe, Africa, Asia, Australia and, of course, New Zealand. The numbers of species and sites continue to grow, with further surveys being added to the data set for future analyses.

So, did this epic effort find any meaningful patterns? It did indeed. Variability in herbivory is not random. Variability in statistical terms refers to the divergence of data from the mean. While mean herbivory was lacking any patterns in the data set, the variability told a different story.

Variability in herbivory within plant populations increases with increasing latitude. Plants at higher latitudes receive less herbivory on average, and this herbivory is concentrated on fewer individuals. This means that further away from the equator, variability in herbivory is greater. The reasons for this could be shorter growing seasons and less stable abiotic

Quentin Paynter and Aoife Neill carrying out survey work

conditions, which reduce the time available for herbivores to forage.

This latitudinal pattern of herbivory variability could help explain how herbivores have influenced global patterns of plant biodiversity. These plant biodiversity patterns failed to be explained by examining mean herbivory alone, because mean herbivory only showed weak correlation with latitude. Closer to the equator herbivory may help maintain high plant biodiversity, not only by being more intense, but also by being more consistent. This pattern could explain why plants further away from the equator exhibit defence mechanisms that only become induced by actual herbivory, as opposed to constantly active defences observed in plants closer to the equator.

The current study suggests that mean plant size affects the variability of herbivory. Variability in herbivory within plant populations was higher among small plants compared to large plants. Growth form (i.e. tree vs climber vs grass, etc.) was less important than mean size for determining how variable herbivory was. In contrast, mean herbivory showed no correlation with either mean plant size or plant growth form.

One explanation for this pattern of variability could be that larger plants are less likely to escape being discovered by herbivores, whereas smaller plants are more likely to either escape herbivory completely or become decimated by a small number of herbivory events, resulting in high variability. One key implication of this pattern is that we could expect larger plant species (and larger life-stages within a species) to evolve more towards a higher concentration of defences that are always present. Conversely, smaller species and smaller life stages should exhibit a lower concentration of defences, which only become induced when herbivores attack.

Finally, the study found that variability in herbivory within plant populations is phylogenetically structured. This means that plant species that are more closely related display similar levels of variability in herbivory within populations. Again, no such relationship was detected between mean herbivory and phylogenetic relatedness. This finding suggests that mean herbivory changes relatively rapidly in response to plant traits that are liable to rapid evolutionary changes (e.g. plant chemical defences), whereas variability in herbivory is more strongly determined by traits that are phylogenetically conserved. Characteristics such as geographical location and plant size, which the study found relate to variability, tend to be less likely to change rapidly.

"This finding – that within-population variability in herbivory seems to be driven by conserved plant traits – tells us that this variability is biologically informative, and is not random noise," concluded Quentin. "Understanding these patterns can inform weed target prioritisation as well as candidate agent prioritisation. For a start, we could look at existing weed biocontrol systems and compare patterns of variability in the native range compared to the introduced range. We could also start collecting data according to the HerbVar protocol for new weed targets as part of surveys in New Zealand and in the native range. Over time we expect these practices can improve our predictions and success rates."

We also now have the advantage of being part of an international network of researchers who are all trained in the survey methods, who could contribute to an international effort to study herbivory on some of our weeds in their native range. Some species of interest may have already been surveyed at least once in the existing data set, or can be made part of a branching add-on study, which the network encourages.

Funding: Quentin & Ronny's participation in this study was part of MWLR's Beating Weeds Programme, funded by the Ministry of Business, Innovation and Employment's Strategic Science Investment Fund.

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Gall of the Californian thistle gall fly

Autumn Activities

Gall-forming agents

- Check broom gall mite [*Aceria genistae*] sites for signs of galling. Very heavy galling, leading to the death of bushes, has been observed at many sites. Harvesting of galls is best undertaken from late spring to early summer, when predatory mites are less abundant.
- Check hieracium sites, and if you find large numbers of stolons galled by the hieracium gall wasp [*Aulacidea subterminalis*] you could harvest mature galls and release them at new sites. Look, also, for the range of deformities caused by the hieracium gall midge [*Macrolabis pilosellae*], but note that this agent is best redistributed by moving whole plants in the spring.
- Check nodding and Scotch thistle sites for gall flies [*Urophora solstitialis* and *U. stylata*]. Look for fluffy or odd-looking flowerheads that feel lumpy and hard when squeezed. Collect infested flowerheads and put them in an onion or wire-mesh bag. At new release sites hang the bags on fences, and over winter the galls will rot down, allowing adult flies to emerge in the spring.
- Check Californian thistle gall fly [*Urophora cardui*] release sites for swollen deformities on the plants. Once these galls have browned off they can be harvested and moved to new sites (where grazing animals will not be an issue), using the same technique as above.
- Look for swellings on giant reed [*Arundo donax*] stems caused by the giant reed gall wasps [*Tetramesa romana*]. These look like small corn cobs on large, vigorous stems, or like broadened, deformed shoot tips when side shoots are attacked. Please let us know if you find any, since establishment is only known from one site.

Honshu white admiral [*Limenitis glorifica*]

- Look for the adult butterflies at release sites, pale yellow eggs laid singly on the upper and lower surfaces of the leaves, and for the caterpillars. When small, the caterpillars are brown and found at the tips of leaves, where they construct pier-like extensions to the mid-rib. As they grow, the caterpillars turn green, with spiky, brown, horn-like protrusions.
- Unless you find lots of caterpillars, don't consider harvesting and redistribution. The butterflies are strong fliers and are likely to disperse quite rapidly without any assistance.

Privet lace bug [*Leptoypha hospita*]

- Examine the undersides of leaves for the adults and nymphs, especially leaves showing signs of bleaching.
- If large numbers are found, cut infested leaf material and put it in chilly bin or large paper rubbish bag, and tie or wedge this material into Chinese privet at new sites. Aim to shift at least 1,000 individuals to each new site.

Tradescantia leaf, stem and tip beetles [*Neolema ogloblini, Lema basicostata, N. abbreviata*]

- Look for the distinctive feeding damage and adults. For the leaf and tip beetles, look for the external-feeding larvae, which have a distinctive faecal shield on their backs.
- If you find them in good numbers, aim to collect and shift at least 100–200 beetles using a suction device or a small net. For stem beetles it might be easier to harvest infested material and wedge this into tradescantia at new sites (but make sure you have an exemption from MPI that allows you to do this).

Tradescantia yellow leaf spot [*Kordyana brasiliensis*]

- Look for the distinctive yellow spots on the upper surface of the leaves, with corresponding white spots underneath, especially after wet, humid weather.
- The fungus is likely to disperse readily via spores on air currents. If human-assisted distribution is needed in the future, again you will need permission from MPI to propagate and transport tradescantia plants. These plants can then be put out at sites where the fungus is present until they show signs of infection, and then planted out at new sites.

Tutsan moth [*Lathronympha strigana*]

- Look for the small orange adults flying about flowering tutsan plants. They have a similar look and corkscrew flight pattern to the gorse pod moth [*Cydia succedana*]. Look, also, for fruits infested with the larvae. Please let us know if you find any, as establishment is not yet confirmed.
- It will be too soon to consider harvesting and redistribution if you do find the moths.

Woolly nightshade lace bug [*Gargaphia decoris*]

- Check release sites by examining the undersides of leaves for the adults and nymphs, especially leaves showing signs of bleaching or black spotting around the margins.
- It is probably best to leave any harvesting until spring.

National Assessment Protocol

For those taking part in the National Assessment Protocol, autumn is the appropriate time to check for establishment and/or assess population damage levels for the species listed in the table below. You can find out more information about the protocol and instructions for each agent at: www.landcareresearch.co.nz/publications/books/biocontrol-of-weeds-book

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Target	When	Agents
Broom	Dec–April	Broom gall mite [*Aceria genistae*]
Lantana	March–May	Leaf rust [*Prospodium tuberculatum*] Blister rust [*Puccinia lantanae*]
Privet	Feb–April	Lace bug [*Leptoypha hospita*]
Tradescantia	Nov–April	Leaf beetle [*Neolema ogloblini*] Stem beetle [*Lema basicostata*] Tip beetle [*Neolema abbreviata*]
	Anytime	Yellow leaf spot fungus [*Kordyana brasiliensis*]
Woolly nightshade	Feb–April	Lace bug [*Gargaphia decoris*]