WHAT'S NEW IN Biological Control of Ueeds?

Delivering mikania rust to the Cook Islands

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Landcare Research Manaaki Whenua They might be small, but the beetles released to control tradescantia (*Tradescantia fluminensis*) are already starting to make a big impression on their host plant here in New Zealand. Tradescantia, which is native to south-east Brazil, is so widespread in New Zealand that manual or chemical control on a large scale is simply not feasible. The plant smothers the ground, preventing native tree regeneration, and is a real nuisance for home gardeners, as well as causing allergic responses in dogs. Even though it is still early days, the little Brazilian beetles appear to be establishing readily and quickly doing damage, described as 'spectacular' in some places!

Over the past few years, three beetle species have been released widely throughout the North Island and at some sites in the South Island, where tradescantia is less of a problem. Releases of the tradescantia leaf beetle (*Neolema ogloblini*), aka 'Shiny', got underway first in 2011, followed by the tradescantia stem beetle (*Lema basicostata*), aka 'Knobbly', in 2012, and finally 'Stripy', the tradescantia tip beetle (*Neolema abbreviata*) in 2013. Until recently the advice has been to release only a single species at each site. "There are several reasons for this approach, even though the beetles will happily coexist on the same plants and

eventually we want them all working together," explained Simon Fowler, who has been leading the project. "Initially we wanted to ensure that the beetles had the best possible opportunity to establish and so it was best to keep them apart. There were also technical reasons such as the staggered availability of beetles for release and the ease of monitoring their individual performance in terms of damage and survival," Simon added.

In March, Landcare Research board members and senior leadership were treated to a tour of weed biocontrol release sites in Northland. As well as seeing successful biocontrol of ragwort (*Jacobaea vulgaris*) and mist flower (*Ageratina riparia*), and participating in the first release of the lantana leaf rust (*Prospodium tuberculatum*) in New Zealand, they saw first-hand early signs of success against tradescantia, which is considered to be one of our worst environmental weeds. After just 4 years the tradescantia leaf beetle population has exploded at one of the earliest release sites in Kerikeri, with thousands of beetles clearly visible and doing obvious damage. As their name suggests, the larvae of the leaf beetles graze on the foliage, skeletonising the leaves, whereas the adults create notches on the side of the leaves. "Higher levels of damage than we expected this



Leaf beetle damage at Kerikeri.

early on were clearly visible and the beetles were so numerous that it has already been possible to collect and distribute some to other sites where the weed is also a problem," said Simon. "The beetles often drop to the ground when they are disturbed so we use a 'pooter' to suck them up or just catch them by hand," explained Jenny Dymock, a local scientist who has been assisting the Northland Regional Council with the release, monitoring and redistribution of the beetles. "We have a long list of people waiting patiently to get some of the beetles and we will prioritise sites that are going to be safe from things like herbicide use, hand weeding and flooding," Jenny said.

At least four tradescantia beetle release sites visited by Landcare Research staff this year have been sprayed, which is clearly not ideal, but fortunately in each case the beetles have survived on remaining foliage. Tip beetles also appear to have survived, but only just, following a major flood last winter near Kaeo in Northland. Successful biocontrol of tradescantia is likely to prove more difficult at sites that are regularly flooded, washing the beetles and their host plant away. "For this reason we also sought and gained Environmental Protection Authority approval to release a yellow leaf spot fungus Kordyana sp., which may prove better in these situations and should also complement the impact of the beetles," said Lindsay Smith, who has devoted a significant amount of time to rearing the beetles and ensuring that they were disease-free before they were released. "We are keeping the fungus 'up our sleeve' until we have quantified the effect of the beetles alone and can decide whether we need to release it here in New Zealand," Lindsay added. In the meantime the fungus has been taken up by Louise Morin from CSIRO, who is looking to see if it might be suitable for controlling tradescantia in Australia. We have also recently supplied Australian researchers with leaf beetles and South African researchers with stem beetles for studies to see if they might be suitable to release in their countries. Closer to home, the Wellington Botanic Gardens have set up their own tradescantia beetle rearing programme and a display to raise public awareness of tradescantia biocontrol.

Also showing early promise in the field is the stem beetle. Stem beetles released at Welcome Bay near Tauranga have already caused significant damage to tradescantia, according to Andrew Blayney from the Bay of Plenty Regional Council. "Although we don't often see the beetles or their larvae, after only 3 years they have reduced the coverage of the plant to bare-ground in places at the release site," said Andrew enthusiastically. Stem beetle adults are harder to see in the field than leaf beetles as they behave more cryptically. The larvae are also harder to see, being inside the stems, but there is no mistaking the damaged stems. Andrew is hoping to transfer some of the stem beetles to other sites next year and he also has a long list of gardeners keen to give the beetles a try!

Since we have always anticipated big things from the tradescantia beetles, a programme to monitor their impacts was put in place at the outset. Landcare Research, along with some interested councils, have set up plots to make detailed measurements, while others are making simpler measurements of beetle damage and impact on the weed as part of the new assessment protocol adopted by the National Biocontrol Collective. We also hope to team up with university staff and get some students involved in more detailed monitoring projects. "Some of the things we are interested in investigating include whether there are ecological benefits that can be directly attributed to the biocontrol of tradescantia, such as implications for native plant regeneration, improved ecosystem services in the form of nutrient cycling, increases in native invertebrate biodiversity and possibly other biota as well," said Simon.

This project is funded by the Department of Conservation, National Biocontrol Collective, and the Ministry of Employment, Business and Innovation as part of Landcare Research's Beating Weeds Programme.

CONTACT: Simon Fowler fowlers@landcareresearch.co.nz



Some of the people who released the green thistle beetle (*Cassida rubiginosa*) a few years ago to control Californian thistle (*Cirsium arvense*) on their properties, have been surprised by the levels of damage to this 'prickly customer' this summer.

The green thistle beetle was brought to New Zealand in 2006 by Landcare Research, on behalf of the Californian Thistle Action Group (CTAG), to try to combat a range of thistles, but in particular Californian thistles. Californian thistle is a perennial weed that has been here for over 100 years, during which time it has become widespread throughout New Zealand. This weed is notoriously difficult to control because of its persistent root system. Using herbicide on thistles is problematic because of impacts on other desirable pasture plants and because of the sheer extent of infestations in both lowland and high-country areas.

Many of the earliest releases of green thistle beetles were made in the Southland/Otago region where CTAG is based, but they have now been released widely throughout New Zealand. As with most insects, it is the larval stage that does most of the damage to plants. The younger larvae begin feeding on the lower sides of the leaves and then when they are a bit bigger they graduate to feeding on the top side of the leaf. "The larvae have a trick to deter predators," said Simon Fowler. "They make a protective canopy out of discarded skin and excrement to carry on their back which seems to deter predators such as birds." The adult beetles live for about 80 weeks if conditions are good, which is quite a long life in beetle terms. During this time the females will lay up to 1000 eggs, making them excellent candidates for biocontrol. Early reports suggested 90% establishment at the original release sites, with sites with shelterbelts or plantations nearby (which act as an overwintering refuge for the beetles) seeming to be favoured.

Previously, painstaking research by Graeme Bourdot's team at AgResearch has revealed that Californian thistle roots don't persist for more than one year. Instead, new roots grow each year, surviving the winter and then sending up new shoots the following season to form the above ground foliage. The roots that have overwintered then die, and new roots are formed to live through the next winter. The AgResearch team found that the amount of above ground foliage is directly proportional to the size of the new root material produced each season. Thus if the amount of green foliage is reduced in one season, the root formation will also be reduced and there will be proportionally fewer resources for the plant the next spring. "This information gives us a clue to use biocontrol agents that are foliage feeders rather root feeders," says Simon Fowler. "Previous attempts to find root feeders have failed and we now realise that this is because the roots completely replace themselves each year, preventing an insect from completing its lifecycle on them," he added. "So we are now concentrating on agents that limit the amount of foliage on the plants, so that there are less vigorous roots and vegetation produced the following year," Simon said. Mowing can be used successfully to the same end, but is not feasible on all terrain.

Some of the earliest release sites are now showing the great potential of the green thistle beetle. Harvey Phillips from the Greater Wellington Regional Council reported a huge outbreak



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Close up of a defoliated thistle.

this summer at a release site near Masterton, where 65 beetles were released in 2008. "There was a visible feeding front, behind which you could easily see where the beetle larvae had demolished the thistle plants," Harvey said. Harvey estimates some stems had as many as 300 beetles on them and he has collected and distributed around 30,000 beetles already from this site.

Similar reports come from Neil Gallagher from Horizons Regional Council, who has been involved with the project since 2008, when he released 120 adult beetles on a farm in the Pohangina Valley. Since then the beetles have dispersed right across the 278-acre farm and are so plentiful that some have already been harvested and redistributed to other farms in the district. "An article in our local newspaper reporting the success of the project resulted in a flood of enquires from other landowners wanting to know where to get some of these little green guys," said Neil. "As a result, a further 16 releases were made this summer in Manawatu, Tararua, Palmerston North, Wanganui hill country, Taihape, and Taumarunui," Neil explained.

Meanwhile, AgResearch scientist Mike Cripps is leading a Sustainable Farming Fund project to monitor the spread of the green thistle beetle and assess their impact on thistle populations. Mike is very encouraged by the heavy defoliation of thistles in some regions, but said that the next step is to get some quantitative data to demonstrate their true impact. "The key will be to determine how much the above ground defoliation by the green thistle beetle limits the growth of thistles in the following season," Mike explained. The project will also enable Mike to promote the use of the beetles to control thistles at field days held at release sites. Mike said that the long-term benefits of having self-sustaining green thistle beetle populations included increased farm productivity, a reduction in the reliance on costly herbicides, and improved engagement with landowners on the benefits of using biocontrol technology to manage weeds at a landscape scale.

If you have a green thistle beetle site that is also doing well we would like to hear from you.

CONTACT: Simon Fowler

fowlers@landcareresearch.co.nz Mike Cripps mike.cripps@agresearch.co.nz

Pampas Project Makes Progress

Biocontrol for pampas (*Cortaderia* spp.) has always appeared to be rather a long shot. "No potential agents were known at the outset of the project and we needed to find agents with a high level of specificity to avoid harming our closely-related native toetoe *Austroderia* spp.," said Lynley Hayes, who leads this project. Pathogens can sometime be so highly host-specific that strains may attack only limited genotypes of a host plant. This is true for four rust fungi of interest to New Zealand, blackberry rust (*Phragmidium violaceum*), Chilean needle grass rust (*Uromyces pencanus*), hieracium rust (*Puccinia hieracii* var. *piloselloidarum*), and lantana leaf rust (*Prospodium tuberculatum*). With such tightly host-specific agents, to avoid host-pathogen incompatibility, it is critical to survey genotypes of the weed in the native range that match what is to be biocontrolled here.

So the first task was to determine where in South America our New Zealand pampas comes from so we could search in the right places. We quickly discovered that current *Cortaderia* taxonomy doesn't resolve the genetic complexity in the group, with molecular studies quickly showing that *C. jubata* and *C. selloana* from Argentina were not the same as plant material in New Zealand of the same name. Molecular studies allowed us to quickly match our *C. jubata* with material in southern Ecuador, but a match for *C. selloana* proved much trickier. With a bit of luck, and the help of a number of South American collaborators, we sourced and genotyped more material from Argentina, Uruguay, Brazil and Chile, and eventually found a match in Chile. However, to complicate matters further, we also determined that some New Zealand pampas was neither *C. jubata* nor *C. selloana*, but an entity we commonly encountered in South American material that we can't accurately name because of the state of *Cortaderia* taxonomy. Initially we thought this material was rare and recommended these populations be eradicated, but the more we looked, the more we found, and so the project scope has widened to seek biocontrol agents for this plant as well."

Surveys in South America revealed only two potential control agents worthy of further study; a fungus and an insect. A black smut fungus that damages the flowerheads, reducing seeding, was found in Ecuador on *C. jubata* and in Chile on another *Cortaderia* species. Sequencing has shown the Ecuadorian smut is a 100% match with the published strain of *Ustilago quitensis*, and the Chilean smut is a 98% match. So the two smuts are likely to be the same species, but it is unclear whether they are different strains with different host preferences, and testing is required to resolve this.

However, so little is known about this black smut that we are not even sure how to infect plants with it. The floral plumes emerge already infected and, based on what is known about other similar pathogens, it may be that infection occurs when seeds germinate, creating a systemic infection that only becomes apparent at flowering time. Dr Charlie Barnes (Pontificia Universidad Católica del Ecuador) has two students looking into aspects of this further. In the meantime, with the help of Dr Hernan Norambuena, in Chile, we have imported the Chilean smut into containment and are observing plants that we have attempted to inoculate in several ways. Once we have a reliable method for infecting pampas plants host-testing will be able to get underway.

The other potential agent is a delphacid planthopper that attacks the leaves. Our Chilean collaborators suspected they had found two similar planthopper species and sent us specimens for sequencing. We confirmed the identity of the more common of the two as *Saccharosydne subandina*, which is reported to have too wide a host-range to have potential as a biocontrol agent. We also confirmed that the less commonly found, planthopper is a novel, un-named species, so no information is available about it at all. With Hernan's help we have recently imported some of the newly discovered planthoppers into containment for further study. "The planthoppers proved to be extremely fragile to ship, requiring three attempts before we had enough live specimens to form a useful starter colony," reported Hugh Gourlay. However, once settled in, the planthoppers quickly set about producing



Pampas planthoppers, adult and juvenile inset.

offspring and causing obvious damage, and there were sufficient numbers to allow host-testing to get underway within weeks. As an acid test, i.e. 'fail this and it is all over', planthoppers were initially exposed to two native toetoe, *Austroderia richardii* and *A. splendens*. Fortunately, these did not appear to be suitable hosts, so more comprehensive host-testing of other critical species (including the other pampas entities) is now underway.

Another key investigation was to see if the imported planthoppers, like other similar species, were associated with any phytoplasma, which are specialized bacteria that cause plant disease and something to be wary of. Relatively little is known about phytoplasma diseases in New Zealand, except where they have caused serious problems to native species like cabbage tree (Cordyline australis) and flax (Phormium tenax) and economically important exotic species like potatoes and celery. "We have used molecular tools, to explore the phytoplasma status of the planthoppers that did not survive the shipping," explained molecular technician Caroline Mitchell. Intriguingly they returned a positive result and 99% match for a bunch of phytoplasmas that relate to Candidatus phytoplasma australiense, the organism responsible for disease in the native plants mentioned above, and thought to occur only in New Zealand and Australia. It will be important now to explore the phytoplasma status of Cortaderia and Austroderia in New Zealand, as part of understanding potential risks posed by the planthopper. If a latent phytoplasma is found in Cortaderia and the planthopper proves highly hostspecific, this could be a damaging combination against pampas.

> Funding provided for this project by the Sustainable Farming Fund is drawing to a close. "This project has achieved a lot more than it originally set out to do," concluded Lynley. Further funding from a variety of sources will be sought to allow studies of the black smut and the planthopper to be completed, and to perhaps gain permission to release them if they prove suitable.

> This project is funded by the National Pampas Biocontrol Initiative through a grant from the Ministry for Primary Industries' Sustainable Farming Fund (11/049), supported by a number of co-funders, including the National Biocontrol Collective.

CONTACT: Lynley Hayes hayesl@landcareresearch.co.nz

Darwin's Barberry Weevil Released

There is finally some relief in sight for farmers and other land managers who have been struggling to control a Chilean plant, Darwin's barberry (*Berberis darwinii*). The thorny evergreen weed generates a huge number of fruit and seeds that are fed on by birds, allowing the plant to quickly spread, reducing pasture quality and native plant regeneration. Over the past few years, Lindsay Smith has been working to develop insect biocontrol agents that could limit the plant's reproductive ability. The Environment Protection Authority approved the release of two insect agents in 2012, a seed-feeding weevil (*Berberidicola exaratus*) and a flower-feeding weevil (*Anthonomus kuscheli*), but rearing them for release has proved unexpectedly difficult.

"We have discovered that barberry plants do not thrive in pots, particularly inside our containment facility under artificial conditions, and they often abort their flowers and fruit and produce little new growth, all of which the weevils need," explained Lindsay. When it became apparent that these plant issues could not easily be overcome to allow mass-rearing, direct field release options were explored. Our Chilean collaborator, Hernan Norambuena, sent a large shipment of both weevils in spring 2013 when they had overwintered and were ready to begin egg-laying. "At this point we discovered that both species were infected with microsporidia parasitic fungi, something we had not encountered with earlier shipments, ruling out any possibility of direct field releases. So we explored whether we could identify disease-free populations of the weevils in Chile," explained Lindsay. Material collected by Chantal Probst during pathogen surveys in Chile a couple of months later confirmed some disease-free sites, so last spring Hernan collected weevils from these sites and shipped them to New Zealand. It was therefore a blow to discover infection in these shipments too.

We then hypothesised that the microsporidian diseases might become prevalent during winter, when the weevils hibernate closely together, but not be passed on via the eggs to offspring (which become infected later on). To test this we asked Hernan to send us seed weevils from Chile as larvae inside infested fruits proved to be disease-free. We originally planned to hold the subsequent new adults in cages over the winter and field release them this coming spring. However, because the new adults were not doing well, perhaps due to the lack of new growth on our sulky potted plants, we decided to release them instead into a field cage where they had a better chance of finding what they need." So in April, with the help of Randall Milne (Environment Southland) we released 70 seed-feeding weevils onto Darwin's barberry plants in Southland, where the plant is a particularly bad problem. To prevent immediate dispersal the weevils were initially caged for a couple of weeks. More weevils will be released at this



Randall Milne making the first ever release of the Darwin's barberry seed weevil.

site when they are available. "This is the first time anywhere in the world this species has been released as a biocontrol agent, and it is great to have finally overcome the challenges to get to this point," confirmed Lindsay.

Now that we have a suitable method we are planning to import an even larger quantity of infested fruit next spring, with the aim of making more direct field releases. The priority is to get the seed weevil established and then to develop direct release methods for the flower weevil also, if subsequent monitoring shows that it is needed. Meanwhile efforts to find a pathogen that could attack the barberry bushes themselves are continuing. Previously two rusts and an unidentified pathogen were identified in Chile as potential biocontrol agents. Further surveys in 2013 by Chantal confirmed rust as the most promising prospect, and last spring we imported some samples for identification. "Molecular studies confirmed that we had found the species of greatest interest for further study (Puccinia berberidis-darwinii)," said Chantal. "This rust is likely to be highly host-specific and, now that the identity of the rust has been confirmed, we have been able to get a permit that will allow us to import the rust into containment for life-cycle studies and host-specificity testing, as soon as we are able to get a new shipment from Chile.

This project is funded by the National Biocontrol Collective.

CONTACT: Lindsay Smith

smithl@landcareresearch.co.nz

In 2014 New Zealand's Ministry of Foreign Affairs and Trade (MFAT) agreed to fund a 5-year project to develop weed biocontol for the Cook Islands. The first agent to be developed under this programme has now been approved for release and successfully delivered to the Cook Islands, where it will be mass-produced and released. The rust fungus Puccinia spegazzinii originates from South America and has recently been established against mile-a-minute (Mikania micrantha) in other Pacific Islands: Papua New Guinea (PNG), Fiji, and Vanuatu. Mile-a-minute, as its name suggests, is a rapidly growing vine, and has become a major weed throughout Asia and the South Pacific. In tropical conditions mile-a-minute is said to grow as fast as 80 to 90 mm in 24 hours. A weed of agricultural, environmental and urban areas, mile-a-minute will grow pretty much anywhere except in heavy shade. It produces thousands of lightweight barbed seeds that are easily spread by wind, people and animals, and plants can reshoot from broken stems. Crops can be guickly smothered and killed, threatening livelihoods of subsistence farmers.

The rust was discovered and developed by CABI Europe-UK and was initially released in India, China and Taiwan in the mid 2000s, although it only established in the latter where a new, more aggressive strain of the rust was released. The rust is highly host-specific and has a life cycle of 15-21 days. Tiny white spots appear on the leaf surface about 6 days after inoculation and these infections continue to grown and develop pustules, turning yellow by 11 days. The rust pustules erupt on the lower leaf surface, petioles and stems, maturing orange-brown in colour around 4-6 days later. These infections lead to cankering on the stems and death of leaves, weakening the vine.

With the help of plant pathologist Sarah Dodd, currently based in Port Vila, we were able to import rust-infected plants from Vanuatu into our containment facility and use these to infect mile-a-minute plants imported from the Cook Islands. Sarah reports that 2 years after the rust was released in Vanuatu the vine has been noticeably reduced there. Monitoring of the impact of the rust in PNG undertaken by Michael Day (Biosecurity Queensland) and Anna Kawi (National Agricultural Research Institute, PNG) also showed a reduction in plant density there following rust establishment. After permission to release the rust in the Cook Islands was received, Quentin Paynter, who leads the project, and plant pathologist Chantal Probst flew to Rarotonga with some infected plants. Following clearance at the airport there was a short ceremony to formally hand the plants over to the Cook Island's Ministry of Agriculture. The infected plants were then placed in a shadehouse amongst uninfected plants that, once infected, will be planted out in areas where mile-a-minute infestations are particularly bad. Experience elsewhere has shown this to be an effective method provided there is adequate water and humidity. The establishment success and impact of the rust in the Cook Islands will be monitored.

Meanwhile approval has also just been granted to release a rust fungus against cockleburr (aka Noogoora burr; *Xanthium strumarium* sp. agg) in the Cook Islands. "We imported *Puccinia xanthii* from Australia, where it has been successfully used against Noogoora burr, in our pathogen facility at Tamaki and undertook some work to confirm that it is safe to release in the Cook Islands and that populations there are susceptible. We plan to deliver the Noogoora burr rust to the Cook Islands in early June," confirmed Quentin.

Also a molecular study of peltate morning glory (*Merremia peltata*) is underway to try and determine how and when this plant colonised the Pacific region. There are conflicting views about whether this invasive vine is native or introduced to various islands, which needs to be resolved before any further steps could be taken to develop biocontrol for this target. Thanks to those from around the Pacific who have provided samples already from Palau, Micronesia and the Solomon Islands. However, we would welcome many more samples from the region – please get in touch if you are able to help.

Many thanks to MFAT for providing the funds for this project through its International Development Fund. ACIAR providing the funding that allowed the rust to be released in Fiji and Papua New Guinea. Australian Government funding allowed the release of the rust in Vanuatu.

CONTACT: Quentin Paynter paynterg@landcareresearch.co.nz

www.landcareresearch.co.nz

Editor: Lynley Hayes Any enquiries to Lynley Hayes

Thanks to: Christine Bezar Layout: Cissy Pan Contributions: Alison Evans



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