New Invertebrate Containment Facility

Weed biocontrol in New Zealand relies on importing potential agents from overseas and we can only do this if we have a secure containment facility in which to house them. Under containment we can safely confirm the identification of the species, conduct host-specificity testing, and check that the agent is disease- and parasite-free, without the risk of them escaping into the outside environment. The containment facility we have been using at Lincoln is 30 years old and coming to the end of its useful life, so in September 2009 we set about building a new one.

The old facility was becoming increasingly inefficient and unreliable. When an audit was conducted on power usage at the Lincoln site the containment facility accounted for about one-third of the total electricity used! The age of the electrical and plumbing systems meant that things often broke down. There was an instance where one of the air-conditioning units failed over a weekend and one of the rearing rooms heated up to over 40°C. All the insects in the room were baked! It had become too expensive to run the old facility and maintain it to the standard we, and the regulations, required.

In New Zealand the standards required of containment facilities are specified by the Ministry of Agriculture and Forestry (MAF) through the Hazardous Substances and New Organisms (HSNO) Act 1996 and the Biosecurity Act 1993. Standards are specified for different levels of physical containment (PC) depending on what is to be housed. PC1 is for containing plants, PC2 is for invertebrates, and PC3 and PC4 are for microorganisms, including diseases. As the PC number increases so does the degree of precautions that must be taken to ensure the organisms can’t escape. “Our new facility, like the old one, is PC2, which means there must be no opening windows, all gaps or cavities must be sealed, and there must be fine mesh on all exit points – such as for plumbing or air conditioning – so that when all the doors are closed inside of the building is a ‘seamless sealed cavity’,” said Hugh Gourlay, who manages the facility. The doors are also magnetically sealed all around. In addition to the legislated requirements we have included four extra features in the new facility, which means it exceeds PC2 minimum requirements. Firstly, there is an airlock, so that the outer door must be closed before the inner one into the PC2 area can be opened. Before leaving the facility all waste water is steam-treated and all air in the ventilation system goes through a sealed HEPA filter (filtration system for microorganisms). Finally, a negative-pressure airflow is created by blowing air into the corridors and internal spaces and sucking it out of the containment rooms – this provides a slight draught into rooms so that nothing will be accidentally blown out their doors.

The new facility was built with resource efficiency in mind. (1) The builders kept landfill waste to a minimum by sorting waste and recycling it where possible. (2) The building is made from “Kingspan” insulated panels supported by a structural steel framework. The panels consist of two outer layers of aluminium with a polystyrene-type core. “‘Kingspan’ has great insulation qualities so less energy is used to keep the rooms at a constant temperature,” said Gordon Burrow, the site manager at Lincoln. It is also fire resistant. (3) Rainwater is collected from the roof and held in retaining tanks for use within the facility. (4) Right throughout the construction records were kept of the amount of water, power, and materials used, as well as vehicle use. From this the amount of carbon emissions created by building the facility can be calculated. Knowing how much was produced means that in the future we could offset these emissions.
There are 13 rooms, one office, and three labs (two in containment and one not). Five of the rooms are dedicated PC2, four are for rearing insects once these are no longer required to be contained, and the remaining four can be either. These four rooms have two doors so that you can lock one and then enter from either containment or the rearing area. “It is great to have this flexibility as our needs change,” said Hugh. Another novel feature is that two rooms have glass ceilings and therefore natural light. Over the years we have found that some insects require cues from changing light levels to initiate parts of their life cycle, such as mating or hibernation, and these are lost under artificial light. We have also installed special light bulbs that are better at mimicking natural-light wave lengths. “They do a very good job, but are so bright that I might have to start wearing sunglasses inside,” said Helen Parish, who rears many of the insects.

The Environmental Risk Management Authority (ERMA) and MAF Biosecurity are responsible for auditing and licensing containment facilities in New Zealand. Every 6–12 months an auditor from MAF will visit the facility to check that it is up to scratch.

The facility must also provide secure containment in the face of a natural disaster or fire. Coinciding with the building of the new containment facility, our Lincoln site recently got a new diesel generator. This generator provides back-up power to the facility (and other essential services at Lincoln) in case of power cuts. Being built of, and containing mostly fire-resistant materials, the new facility is at low risk of fire; and the local fire service knows about the need to avoid breaching containment so would not break down any doors in the event of fire.

Before any insects are moved in we need to confirm that everything is running properly, so the first new resident will be Hugh. There may also be some residual chemicals and smells, which the insects might not like, to get rid of. The first insect residents should move in to their new home in June. Species presently residing in containment will move over first, such as the four tradescantia beetles (Neolema ogloblini, N. abbreviata, Lema basicostata and Buckibrotica cinctipennis), woolly nightshade lacebug (Gargaphia decoris) and two potential moth plant agents (Colapsis argentinensis and Rhysomatus diversicolor). Any new species imported from overseas will then take up residence as they arrive.

The new facility was opened on 29 April by the Minister of Conservation Kate Wilkinson and will be known as the David Miller Invertebrate Containment Facility. Dr Miller was a prominent figure in New Zealand entomology and an early driving force for biocontrol of weeds research (see below).

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Dr David Miller, CBE (1890–1973)

One of the founders of professional entomology in New Zealand, Scottish born David Miller was internationally respected for his work on biocontrol. He initially worked on forest and timber insects, which led to the establishment of the Forest Biological Research Station at Nelson in 1929. The success of the New Zealand timber industry is largely due to research Dr Miller initiated on methods of timber preservation, particularly the control of insect pests. During the Depression he campaigned vigorously for backing from primary producer organisations, local bodies and banks to maintain the services of his highly skilled entomological staff. Under his guidance New Zealand had become a world leader in the field of biocontrol by the late 1930s, and he helped to establish the Commonwealth Institute of Biological Control as well as the Commonwealth Institute of Entomology in London. Dr Miller became the director of the Cawthron Institute in 1956. He retired in 1959, but continued to work and publish. Dr Miller was the author of many technical papers and books, including a Catalogue of the Diptera of the New Zealand Sub-region (1950), Insect People of the Maori (1952), Native Insects (1955), Bibliography of New Zealand Entomology (1956), and Common Insects in New Zealand (1971). He was made a Fellow of the Royal Society of New Zealand and of the Royal Entomological Society of London, he received the Hutton Memorial Medal, and in 1958 was awarded a CBE.
Ginger Project Heats Up

Surveys led by CABI Europe–UK for potential biocontrol agents for weedy ginger species (*Hedychium* spp.) have been continuing in India. Gaining the necessary permission to undertake this work has been enormously challenging, especially with respect to taking any biological material out of India. But with a collaborative Memorandum of Understanding (MOU) now in place and an application for a Material Transfer Agreement (MTA) awaiting final approval, these difficulties appear close to being resolved.

After a brief exploratory survey in 2008, two further surveys were undertaken in 2009 in the state of Sikkim, before and after the monsoon. *Hedychium* spp. were again found to have a large number of natural enemies — many of which are likely to be new to science, attacking all stages of the plants. “The biocontrol of ginger appears to have great potential,” claims Djami Djeddour of CABI. Some of the most promising natural enemies of interest found so far are described below.

A rust fungus (identified as a species of *Puccinia*) was found on kahili ginger (*H. gardnerianum*) seedlings. No infected adult plants were found, and it has been suggested that the white powder commonly seen on the undersides of adult leaves could hold some antifungal properties. *Puccinia* rusts have a long and successful history as biocontrol agents as they tend to be highly co-evolved and host specific. *Mycosphaerella* leaf spots were also found, on both kahili and white ginger (*H. coronarium*). Species of *Mycosphaerella* cause extensive economic losses on many crops, such as bananas, but many are known to be highly specific, so the leaf spot found on ginger will also be investigated further if it continues to show promise in the field.

“In the native range we often found kahili ginger plants with arrested stem elongation and consequently no flowering; in some cases more than 60% of tips were affected,” reports Djami. This damage appears to be caused by a suite of chloropid flies. Both young and mature tips were affected. Fly larvae were also found feeding on the seeds. Since kahili ginger has a high rate of abortion of fruit and seed in its native range, researching the role that these flies play in this will be a priority. The identities of all the flies found also need to be confirmed.

The most striking insect found so far is a large (1.5–2 cm long), handsome, red and black weevil (tentatively identified as a *Prodioctes* sp. by Indian taxonomists). The adults were seen chewing voraciously on leaves, stems, seed capsules and flowers of kahili, white and yellow ginger (*H. flavescens*). Identifying the mode of larval attack and confirming the level of specificity of the weevil will be a valuable step towards prioritisation of agents for the project. A number of beetles found attacking the flower heads may also show promise, but further study is needed to establish this.

Gregarious lepidopteran larvae were found en masse, skeletonising the leaves of kahili and yellow ginger. A wide assortment of other lepidopteran larvae were also found including a very large geometrid (8 cm long), small and large hairy caterpillars, a case moth, shoot and fruit capsule borers, and leaf cutters and folders, all of which still need to be reared through to adults so they can be identified.

So there is plenty of work to be going on with! Molecular studies to clarify how the plants found in India compare with the weed populations needing to be controlled in New Zealand and Hawai‘i will be undertaken once the MTA is signed. This will hopefully help match the introduced weeds with their exact area of origin and focus the search for those potential biocontrol agents that are likely to be most effective here and in Hawai‘i.

This project is funded by the National Weed Biocontrol Collective (New Zealand), and The Nature Conservancy of Hawai‘i.

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More to Seed-feeders than Meets the Eye

Part I - Broom Seed Size

Recent research shows that seed quality may play a role in explaining why broom (Cytisus scoparius) is such an invasive pest in its introduced range yet just a normal plant at home. Modelling predicts that three factors could explain why broom is a weed in exotic habitats: disturbance, longevity of broom plants, and ability to recolonise after the death of the parent plant. In France and the UK (native range) disturbance is important for seedling establishment and seedlings rarely take root in the shade of an undisturbed broom canopy. In contrast, in New Zealand and Australia (introduced range) seedlings frequently establish under mature plants. Longevity does not appear to contribute to weediness because broom plants live for similar lengths of time in both the native and introduced range. Differences in seed production are also unlikely to be the cause. Modelling has shown that reducing seed rain by up to 90% has little effect on overall broom populations. So differences in the ability of broom seedlings to establish in undisturbed sites, such as beneath their parent plants, explains why broom does better in the introduced range. New seedlings that grow up through mature plants contribute to a self-sustaining broom patch that will persist in the landscape. So what is causing this difference?

Research into seed size has found that broom seeds are on average about 30% heavier in the introduced range. Data from a variety of other plant species show that seed size influences the early growth and survival of seedlings. Preliminary work on broom agrees, showing that seedlings from large seeds survive better in shaded conditions than ones from small seeds.

We suspect that the difference in broom seed size between the native and introduced range might be due to seed-feeding natural enemies. Recent work shows that a close size relationship exists between the broom seed beetle (Bruchidius villosus) and broom seeds (see graph). “We collected and measured seed and beetles from 14 sites in New Zealand and found that beetle size is positively correlated with broom seed size. Big seeds produce big beetles!” confirmed Quentin Paynter. As beetle larvae feed internally on one broom seed each, it makes sense that small seeds might not have the food value or space to support the development of a large beetle. Being a big beetle has some advantages. In an experiment we found that the largest females laid on average about 30–40% more eggs than the smallest females. Preliminary work has also shown that small beetles do not survive as well over winter as large ones.

So it would seem that the broom seed beetles will do better on large-seeded broom plants thus creating a selection pressure for plants that produce small seeds. This pressure works through patches of broom that are predominantly small-seeded supporting far fewer beetles because of the lower quality (small) seed than patches of large-seeded plants. The small-seeded plants continue to set lots of seed and over time their small-seeded progeny are most likely to persist and reproduce. We are currently investigating whether the broom seed beetle has been present in New Zealand long enough to see a reduction in seed size yet. If our theory is right it will happen and contribute to reducing the invasiveness of broom by selecting for plants with smaller seeds and, therefore, less competitive seedlings.

There is an additional factor affecting broom seed set in New Zealand, the varroa mite (Varroa destructor). We have already reported that introduced bees are the only pollinators of the weed here and that without them broom would...
not be invasive (see Bees Busted, Issue 38). The varroa mite is killing off feral honeybees (Apis mellifera) and thus reducing the abundance of pollinators available to fertilise broom flowers. A likely consequence is a decline in broom pollination and seed set. Simulating the impacts of the broom seed beetle alone on broom coverage shows that a high level of seed destruction is required to significantly reduce broom infestations. When the effects of the varroa mite are added in, a much lower level of seed destruction is needed to reduce broom cover. “This shows that the broom seed beetle alone could have a major impact on broom,” said Quentin. However, commercial beehives are treated against varroa in many parts of the country allowing the presence of broom pollinators to be maintained. Management of beehives including restrictions on their location, such as not within Department of Conservation land, could be a beneficial part of an integrated broom management programme at many sites in New Zealand.

The impacts on broom described above are primarily due to one seed-feeding biocontrol agent and of course there are five more targeting the foliage. The newest of these is the gall mite (Aceria genistae), which has taken off well in Canterbury. The tiny mite was first released at Lincoln in November 2007 (New Agent Update, Issue 47) and broom plants at release sites in North Canterbury and Lincoln are already showing leaf loss and stem-tip dieback due to the number and size of galls that have formed. Releases were also made at sites in Te Anau, Southland, Bay of Plenty, and Wellington, and will continue. Curiously, the mites seem to establish readily on some broom plants but less well on others, even within the same release site. We are not sure why this should be but perhaps it is a case of a high level of host-specificity where the mites are specific to a particular strain of broom.

This project was funded by the Foundation for Research, Science and Technology under the Beating Weeds II programme.

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Part II - Gorse Flowering Strategies

Gorse (Ulex europaeus) has proved to be a difficult and long-term target for biocontrol in New Zealand. Recent work looking into the relationships between gorse flowering strategies, seed-feeding biocontrol agents and pollinators is giving us a better understanding of why we have not been as successful as we had hoped in reducing seed production by this invasive weed.

The timing of gorse flowering is variable and this is the case both in its native range and in countries where it has been introduced, including New Zealand. Some plants flower primarily in winter while others just flower in spring. A study of gorse flowering phenology in its native range suggests that the two co-existing flowering types are the result of opposing selection pressures. Seed predation, which occurs only in spring, selects for winter-flowering. However, cold winter temperatures may cause seed pods to freeze and abort the seed, which selects for flowering in the spring.

In New Zealand it appears that the present pattern of gorse flowering has not always been the case. Anecdotal evidence shows that gorse was predominantly spring-flowering when the gorse seed weevil (Exapion ulicis) was first released in the 1930s, and now it commonly flowers in winter. Could it be that gorse flowering phenology has changed in response to the gorse seed weevil?

In New Zealand the gorse seed weevil destroys up to 99% of the spring seed crop but has no impact on seed produced later in the year. The gorse pod moth (Cydia succedana) was introduced in the 1990s in the hopes that it would damage seed set in autumn. This has not proven to be the case as the moth is not well synchronised with gorse flowering. Studies have shown that adult moths are active and laying eggs from November through to March and so have built up to high numbers just as gorse finishes spring-flowering and only the tail-end of seed pods is present. For winter-flowering gorse there is an even worse match. Most pods are therefore produced when moth and weevil activity is low or non-existent (see graph). So there is definitely
an advantage to gorse flowering during the winter in New Zealand. “The gorse pod moth may in fact only persist in New Zealand because it can attack non-target weedy plants, such as Lotus and Lupinus species, which flower in the summer,” said Quentin Paynter. It will be interesting to see if the pod moth can adapt to become better synchronised with gorse flowering in New Zealand.

In terms of winter temperature, studies at a spring-flowering gorse site near Rotorua and a winter-flowering site near Christchurch showed similar temperatures at both (mean maximum 12–13°C, mean minimum 3–4°C). This indicates that cold winter temperatures do not always explain the presence of spring-flowering and other factors must also be involved.

One of these other factors is pollinator-limitation. Like broom, gorse is exclusively pollinated by exotic bees in New Zealand. Honeybees (Apis mellifera) are only active when the temperature is over 10°C and bumblebees (Bombus spp.) hibernate over winter, only occasionally emerging to forage on sunny days. Consequently, the amount of seed that winter-flowering gorse can produce is limited by the number of bees that are active. In contrast, pollinator activity is unlikely to restrict the amount of seed produced by spring-flowering plants. Data on spring pollinator abundance at two gorse sites showed around four times higher levels at the winter-flowering site than the spring-flowering site. “We can assume from this that gorse growing at the site with low pollinator numbers in spring is more likely to be pollinator-limited in winter, and therefore selecting for spring flowering, than the site with abundant pollinators in spring,” said Quentin. If pollinator numbers are high enough over winter, seed set then will exceed spring seed set (when seed-feeders are active) and make it worthwhile for plants to flower in winter. Spring-flowering gorse probably persists in New Zealand because pollination is so low in winter, meaning that winter seed set is lower than spring seed set, even though seed-feeders destroy a high proportion of spring seeds. As with broom, the presence of the varroa mite (Varroa destructor) is sure to have an impact on gorse pollination. By killing off wild honeybees, winter pollinator activity will be further reduced and in areas of already low pollinator numbers perhaps to below a threshold which makes it worthwhile flowering in winter at all. The higher the proportion of gorse that flowers in the spring, the higher the proportion available for the seed weevil to attack.

Unlike the broom seed beetle (Bruchidius villosus) our gorse seed-feeders are unlikely to affect seed quality in terms of size (see Part I - Broom Seed Size). Both the weevil and moth larvae feed on seed from the outside and each individual can damage more than one seed. Therefore weevil and moth sizes are not going to be constrained by seed size and so neither agent is likely to be exerting pressure on gorse to favour small or large seeds.

While seed-feeding biocontrol agents are having an impact on gorse reproduction in the long term, we still need to increase the pressure on the weed from foliage-feeders. On this note, the gorse colonial hard shoot moth (Pempelia genistella) is finally starting to boom in North Canterbury. Although the moth has been widely released, establishment has only occurred so far at sites in Canterbury. Moths released near the Wairau and Hurunui rivers in 2004 have done extremely well. A recent visit by Hugh Gourlay and Environment Canterbury staff found the moths established over a wide area at both sites and causing obvious damage to some plants.

This project was funded by the Foundation for Research, Science and Technology under the Beating Weeds II programme.

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Winter is a quiet time of year as most biocontrol agents become dormant or hide away during the next few months. However, if you need some fresh air you can still:

- Check nodding thistle crown weevil (*Trichosirocalus horridus*) release sites. Most weevils begin to lay eggs in the autumn and the damage they cause becomes most obvious later in the winter. Look for black frass in the crown and for leaves that have lost their prickliness. To find adult weevils look carefully on the undersides of leaves; they can often be successfully harvested and shifted around as late as June. While the crown weevil prefers nodding thistle (*Carduus nutans*), you may find signs of attack on other species of thistles too, especially Scotch (*Cirsium vulgare*) and cotton (*Onopordum acanthium*) thistles.
- Check ragwort flea beetle (*Longitarsus jacobaeae*) release sites. The beetles can be shifted around if present in good numbers.
- Make sure all the paperwork relating to release sites is up to date. If you have been shifting agents around, we would be interested to know about this (send information to Lynley Hayes: hayesl@landcareresearch.co.nz).

Things To Do This Winter

We are running our annual one-day workshop in June to fill you in on the latest weeds research in New Zealand, but this time you will be able to choose between two concurrent sessions, as we have expanded the workshop to include talks on mammal pests too.

Biosecurity Bonanza

**WHEN:** 9th June 2010  9:30am-4:00pm

**WHERE:** Commodore Airport Hotel, 449 Memorial Ave, Christchurch

**HOW:**
Free workshop, catering provided, limited to 120 people: To view the agenda or secure a place visit:www.landcareresearch.co.nz/news/conferences/biosecuritybonanza/
If you do not have computer access please phone Andrea Airey 03 3219 618.