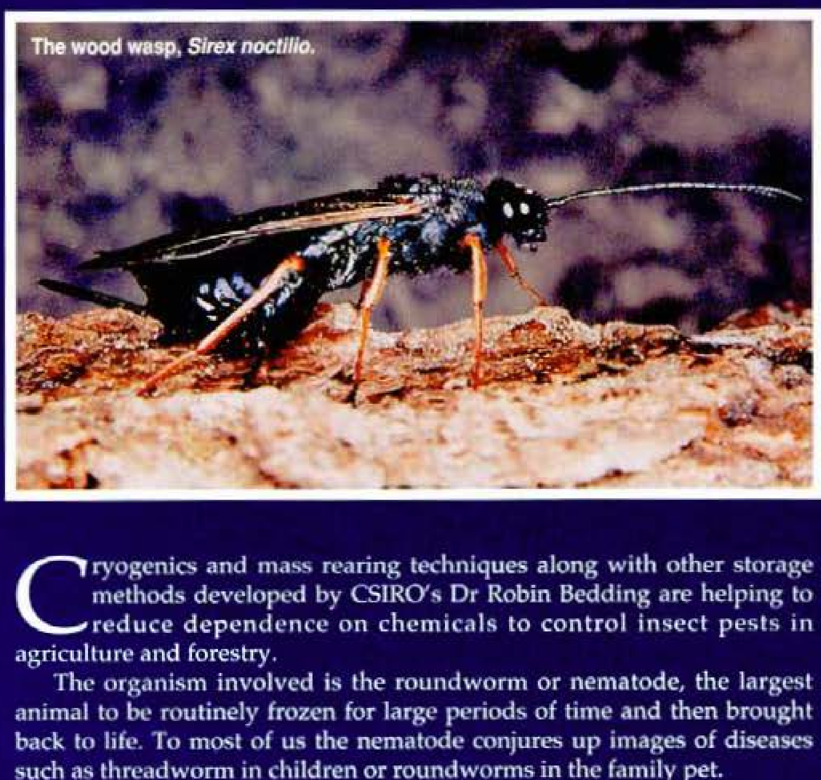
Cool work with wasps and worms


The wood wasp, *Sirex noctilio*.

The use of nematodes to control insect pests has been pioneered by Dr Robin Bedding. His work at the Division of Entomology has fostered a new export industry. Barry Kuch highlights one of the success stories.

Cryogenics and mass rearing techniques along with other storage methods developed by CSIRO's Dr Robin Bedding are helping to reduce dependence on chemicals to control insect pests in agriculture and forestry.

The organism involved is the roundworm or nematode, the largest animal to be routinely frozen for large periods of time and then brought back to life. To most of us the nematode conjures up images of diseases such as threadworm in children or roundworms in the family pet.

Indeed it is true that nematodes surround us in their billions. An apple decomposing on the ground of a typical backyard may yield between 50 000 and 90 000 nematodes belonging to several different species, while many millions of nematodes may be found in a square metre of mud scooped from the floor of Victoria's Westernport Bay.

Roughly speaking, nematodes are either free living or parasitic, although many species display both forms in their life cycle. The parasitic forms are so numerous that virtually no animal or plant is free of their attention.

It is this parasitism which brought them to the attention of Bedding, who for the past 20 years has sought to match parasitic nematodes to specific pests. His greatest success to date, of significant economic importance to Australia, has been the control of the wood wasp *Sirex noctilio*. ⇨

The sirex wood wasp was accidentally introduced to Australia in the 1950s. With no natural predators, it vigorously attacked softwood plantations, notably radiata pines. This introduced pine from North America accounts for the bulk of softwood production, with more than one million hectares under plantation.

The devastating effect of sirex was illustrated in South Australia in 1987. An outbreak of sirex in 50 000 ha of plantations resulted in the death of almost five million trees, valued at millions of dollars. It has been estimated that the uncontrolled spread of sirex over a 30-year plantation rotation period has the potential for a $1-$4 billion loss of timber.

The nematode used to control sirex is *Deladenus siricidicola*. This nematode can achieve parasitism in sirex of close to 100%. It does so by infecting the gonads of the adult wasp.

Although *Deladenus* does not sterilise males, it sterilises females either by suppressing egg development, or by entering and disrupting the eggs. The sirex female can still lay these eggs and in doing so spreads the nematodes to new trees. In the South Australian example, once the nematode had been introduced, the sirex population crashed within two years.

One of the unusual characteristics of this nematode is that it also occurs as a free-living form. This version of the nematode may repeatedly occur through a number of generations.

The free-living form of *Deladenus* feeds solely on symbiotic fungus within the tree introduced by sirex to feed their own larvae. Under certain conditions of high carbon dioxide levels and low pH, the free-living nematode juveniles will grow into the parasitic form and, as adults, invade the body cavity of sirex larvae. Once present in the sirex, they reproduce hundreds of juvenile parasitic nematodes which migrate to the mature wasp's reproductive organs.

When this generation is liberated by the mature sirex it produces nematodes which revert to the free-living form. This generation and subsequent generations remain free-living within the tree until nematode populations reach the vicinity of a sirex larva which again favours the development of the parasitic form of the nematode.

What prompted the development of cryogenics and mass-rearing technology in the fight against insect pests? It came in response to a number of unforseen problems that arose in the use of these natural predators of sirex.

The first problem is the infectiveness of the nematodes which are released. Twenty years ago when a plantation was inoculated with *Deladenus*, a virtual 100% parasitism of sirex was guaranteed. Today, parasitism may be as low as 25%.

The continual sub-culturing of free-living nematodes on a fungal culture without the development of a parasitic form has created a twist to the story.

In the nematode, it has culminated in a loss of their genetic ability to develop into the parasitic form in the field. For this defective strain to be effective, there must be very high levels of infestation by sirex before parasitism is sufficiently high to result in satisfactory control. This is clearly undesirable because of the economic cost of the high levels of sirex causing damage to mature trees.

Massive tree damage in a South Australian pine plantation as a result of sirex attack during 1988.

Here cryogenics came into play. First Bedding and his team had to re-isolate the original strain they used 20 years ago. This proved to be a daunting task as most of the pine plantations in south-eastern Australia had already been inoculated with the defective strain of nematode.

After a detailed search of all forests to find individuals from the original strain (198 strain), a breakthrough occurred in the Kamona forest near Scottsdale in Tasmania. Here it was discovered that an inoculation of the 198 strain had occurred only once and the defective strain had never been introduced. Even so, only nine trees in the forest were found to contain the original strain.

This strain, (now called the Kamona strain), was collected from one of the trees and carefully cultured until large numbers were once again established.

To protect the genetic integrity of this strain, hundreds of vials of these nematodes have been snap-frozen at temperatures below −190°C in liquid nitrogen storage containers. Unlike other methods of storage, these nematodes will last well over 12 months with a greater than 75% survival rate. Each year a fresh vial of nematodes is removed from liquid nitrogen and nematodes cultured from it. In this way more than 3000 million of the new Kamona strain of nematodes have been produced for release in NSW, SA and Victoria during the past three years.

The second problem is that the natural spread of *Deladenus* by sirex may not be fast enough or over a sufficient spread to counter a severe outbreak of the wasp. To artificially enhance the effectiveness of the nematode, large numbers must be in ready supply to be inoculated at the optimum time. These large numbers of nematodes must be reared easily and economically to make their use viable.

In the South Australian outbreak, it was necessary to inoculate 20% of the infected trees with 50 000 million *Deladenus* nematodes to guarantee the spread of the nematode in the next year as the sirex emerged.

Export market

Bedding's success with nematodes has not been confined to *Deladenus*. He has developed the technology for producing, processing and storing a range of different 'entomopathogenic nematodes' (nematodes that penetrate and rapidly kill insects with the help of a special symbiotic bacterium). Already, hundreds of billions of these nematodes have been produced for the Australian and overseas market. The technology that makes possible this production has secured CSIRO significant royalties and research funding.

Today CSIRO has the largest collection of nematodes stored either in liquid nitrogen or as cultures. In China, the control of a major apple pest using Bedding's nematodes to protect an annual harvest worth $1.7 billion a year appears imminent.

Here in Australia, the nematodes *Steinernema feltiae* and *Heterorhabditis bacteriophora* have been successfully used to control the black currant borer and the black vine weevil respectively. Final grower trials are testing *Steinernema carpocapsae* to control the banana borer. Eventually this nematode may replace insecticides such as organophosphates and carbamates, which cost the banana industry $5 million a year and to which the borer is becoming increasingly resistant.

The local and overseas market for this technique of insect control will continue to grow. Bedding and his team have ensured that Australia is at the forefront in the development of environmentally sound and cheap alternatives to costly and increasingly ineffective insecticides.

It is comforting to know that should these tiny nematodes begin to lose their edge over their insect hosts help is only as far away as the freezer!

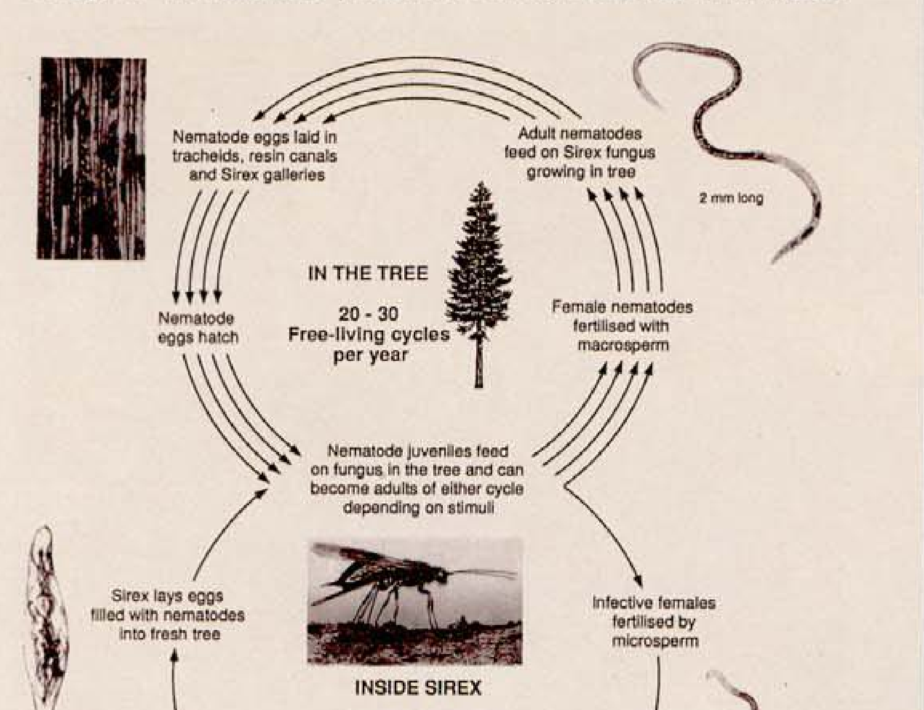
More about nematodes

Bedding RA (1990) Logistics and strategies for introducing entomopathogenic nematode technology into developing countries. In: *Entomopathogenic nematodes in biological control* (Eds R Caugler & HK Kaya) CRC Press, Boca Raton, Florida. 233-246.

Bedding RA (1992) Strategy to overcome the crisis in control of sirex by nematodes. *Australian Forest Grower*. Summer 1991/1922: 15-16.

Nematodes and the biological control of insect pests (1993) Eds RA Bedding RJ Akhurst & HK Kaya. CSIRO Publications.

Biology of the nematode parasite of sirex, *Deladenus siricidicola*.



Below: ***Deladenus siricidicola*** **can achieve parasitism in sirex of close to 100%. It does so by infecting the gonads of the adult wasp. Bottom: Dr Robin Bedding examines insect damage in China with a view to using entomopathogenic nematodes as a control technique.**