BIOLOGICAL CONTROL OF THE WOODWASP *Sirex noctilio* IN AUSTRALIA

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ABSTRACT

The woodwasp *Sirex noctilio* is the major pest of *Pinus radiata* plantations in Australia. An account of the introduction, spread, current status and development of the integrated pest management of this insect in Australia is provided together with a more detailed description of recent developments in the use of nematodes in its control.

RESUMEN

La avispa de la madera, *Sirex noctilio*, es una de las principales plagas de las plantaciones de *Pinus radiata* en Austrália. Un relato de la introducción, dispersión, estado actual y desarrollo del manejo integrado de este insecto es presentado juntamente con una descripción detallada del desarrollo en el uso de nematóides para su control.

RESUMO

A vespa-de-madeira, *Sirex noctilio*, é uma das principais pragas de plantações de *Pinus radiata* na Austrália. Um relato da introdução, dispersão, estado atual e desenvolvimento do manejo integrado deste inseto é apresentado, juntamente com uma descrição detalhada de desenvolvimentos no uso de nematóides para seu controle.

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INTRODUCTION

During the middle of the nineteenth century a number of exotic species of pine tree were introduced into Australia because of a shortage of endemic softwoods. One of these *Pinus radiata* D. Don, Monterey pine, originally from California, now forms the bulk of Australia's one million hectares of pine plantations and is also the main plantation species in New Zealand and Chile.

There are over 40 species of siricidae throughout the world most of which oviposit in dead, dying or damaged trees, but only one of these, *Sirex noctilio* (F.), from Europe is known to be capable of killing relatively healthy trees (although in Europe it generally confines its attacks to weakened or dying trees). It is this species, that was accidentally introduced into New Zealand during the early 1900's, the Australian island of Tasmania during the early 1950's and onto the mainland of Australia (near Melbourne) at the beginning of the 1960's. Unfortunately, it transpires that out of all soft wood trees, *P. radiata* is one of the most susceptible to *S. noctilio* (SPRADBERY 1973). Thus there was in Australia an unfortunate combination of the most virulent siricid from Europe, one of the most susceptible tree species from USA, an absence of natural enemies and a climate disposed to inevitable periodic droughts which stress trees and make them more susceptible to attack. The outcome of this "recipe for disaster" and how it has been dealt with in Australia forms the basis of this paper.

INTRODUCTION, SPREAD, DAMAGE AND PRESENT STATUS OF *Sirex*

*S. noctilio* (sirex) did little damage in New Zealand for nearly 50 years after its introduction there, but killed 20-30% of *P. radiata* trees in 120,000 ha of unthinned plantations between 1946 and 1951 (RAWLINGS 1955). Then in 1952 sirex was found in a *P. radiata* plantation at Pittwater near Hobart in Tasmania after its accidental introduction probably from New Zealand in a shipment of timber. Within a decade, sirex had killed some 40% of the trees in the Pittwater plantation. Whether sirex reached the mainland of Australia in another shipment of timber from New Zealand or via Tasmania is a matter of some debate but despite efforts to eradicate or at least contain it, it spread from the Melbourne area at a rate of about 20 to 30 km per year.

During the 1960's many pine shelterbelts on farmland were destroyed by sirex and some 10,000 trees were killed in various forests owned by a private company (NEUMANN et al. 1987). During the 1970's, a major infestation occurred in the Delatite plantation of east-central Victoria. In this 2000 ha plantation however, infestation developed from 10 sirex killed trees in 1972 to severe damage from 1976 - 1978 to a collapse of the sirex population in 1979 (MCKIMM & WALLS 1980) (Table 1). Up until 1979 losses from the entire plantation were equivalent to 12% of the total merchantable wood volume (MCKIMM & WALLS 1980) while in the worst affected stands merchantable volume was reduced by 48% (NEUMANN & MINKO 1981). This infestation is believed to have been associated with drought induced tree stress (NEUMANN et al. 1987). However, a major contributing factor was lack of control by nematodes which were not introduced until 1975 and then only sparsely. Nevertheless, by 1977, 52% of emerging sirex were parasitised by nematodes (Walls 1977) and at the time of the population collapse in this plantation, nematode parasitism was over 90%.
TABLE 1 Total tree mortality from 1972 - 1979 in the Delatite plantation as a result sirex infestation. (Compiled from NEUMANN et al. 1987)

<table>
<thead>
<tr>
<th>Silvicultural treatment</th>
<th>Area infested with sirex (Ha)</th>
<th>Tree Mortality* (1972-1979)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unthinned</td>
<td></td>
<td></td>
</tr>
<tr>
<td>25</td>
<td>77%</td>
<td></td>
</tr>
<tr>
<td>79</td>
<td>63%</td>
<td></td>
</tr>
<tr>
<td>379</td>
<td>35%</td>
<td></td>
</tr>
<tr>
<td>70</td>
<td>15%</td>
<td></td>
</tr>
<tr>
<td>390</td>
<td>0%</td>
<td></td>
</tr>
<tr>
<td>Thinned</td>
<td></td>
<td></td>
</tr>
<tr>
<td>78</td>
<td>26%</td>
<td></td>
</tr>
<tr>
<td>266</td>
<td>5%</td>
<td></td>
</tr>
</tbody>
</table>

* Accumulated percentage of mortality from 1972 to 1979.

Sirex took 17 years to reach the borders of the state of Victoria. During this time various control measures, including nematode and parasitoid liberations, were undertaken and as a result, apart from the one in the Delatite plantation, there were relatively few serious outbreaks of the pest. This led to complacency so that after sirex arrived in the "Green Triangle" forests of SW Victoria/SE South Australia during 1977 adequate protective measures were not taken. Although there were yearly liberations of the insect parasitoids *Italia leucospoides* (Hockerworth) and *Megasyrus nortoni* (Cresson) no serious attempt was made to introduce the nematode *Deladenus siricidicola* Bedding for nine years. By then it was almost too late because in 1987 1.8 million trees were killed by sirex (HAUGEN & UNDERDOWN 1990b) and during the next two years a further 3 million trees were killed.

The 113,000 hectares of *P. radiata* forest in the Green Triangle turned out to be a giant control plot showing that in the absence of control agents, sirex could kill up to 80% of trees in some areas. Fortunately, as a result of a $1.3m operation to introduce nematodes during 1987, up to 100% nematode parasitism was reached within two years and the sirex population crashed. However, this was not before millions of dollars worth of timber was lost and the quality of the timber of many of the remaining trees impaired (Underdown pers. comm.). On evidence from this outbreak, it has been calculated Underdown (pers. comm.), that in the absence of control agents, sirex had the potential to cause a A$1 billion to A$4 billion loss of timber in each rotation (every 30 years) of the total pine plantations in Australia.

Sirex has now spread to most forests in New South Wales and South Australia. Currently there are few serious outbreaks but these may well develop particularly in new areas in response to drought or other stress factors unless the efficacy of nematode control is improved (see below). Sirex has not yet reached West Australia and Queensland. Although West Australia has large forests of *P. radiata*, the main pine species in Queensland are *P. elliottii* and *P. caribea* which are doubtful susceptibility.

The one benefit of the Green Triangle outbreak was that it alerted foresters throughout Australia and hopefully elsewhere in the world to the importance of sirex as a severe pest of pine trees. As a consequence, renewed effort has been since then made to adequately deal with the problem on a national scale.

DEVELOPMENT AND USE OF INTEGRATED PEST MANAGEMENT

During the 1960's attempts were initially made to eradicate sirex from the mainland of Australia in "search and destroy" operations which involved cutting down and burning all sirex infested trees. Together with this there was considerable publicity about sirex and it was made mandatory for owners of pine trees to report sirex attack to the Forests Commission, Victoria. When sirex proved impossible to eradicate and because of the very great expense involved, "search and destroy" was restricted to the advancing front while attempts were made to
introduce parasitoids and nematodes into the "core area". At an early stage the possibility of using insecticides to control sirex was rejected as being too difficult and expensive.

COLLECTION OF BIOLOGICAL CONTROL AGENTS

While sirex was restricted to Tasmania, importation of biological control agents was limited to import of the parasitoids *R. persuasoria* and *I. leucospoide* from New Zealand. The Tasmanian Department of Agriculture liberated the former in 1957 and the latter in 1959 and 1960 in the plantation in Pittwater (ANON 1958, 1959, 1960). However, once sirex was discovered on the Australian mainland, a major program of collection of and research of these agents commenced. Two sirex units were established. One at Hobart airport was for the receipt of parasites and research on these and on sirex. The other at Silwood Park in England was the quarantine centre for collecting sirex infested material and parasitoids and nematode parasites from anywhere in the world that sirex and other sirecid species were to be found and for conducting intensive research on this material. In all, thousands of infected billets were collected from most countries in Europe, Turkey, North Africa, North America and Japan and a wealth of Rhysines, *Ibalia* species and parasitic nematodes were obtained from this material. At an early stage, the Commonwealth Institute of Biological Control, (CIBC) was also involved in collecting parasitoids from Canada, India and Pakistan. A total of 21 species of insect parasitoids (TAYLOR 1975) and 8 species of nematodes was eventually sent to Tasmania for final evaluation.

PARASITOIDS

Species of the ichneumonids, *Megarhyssa, Rhyssa*, and *Schlettererius* that have been introduced into Australia and the indigenous *Ceratonous tasmaniensis* Turner drill deep into the tree response to odours from the symbiotic fungus (MADDEN 1968), and paralyse well developed sirex larvae before ovipositing on them. The parasite larvae then feed as ectoparasites. *Ibalia* spp. oviposit down the original sirex oviposition hole to lay an egg within the developing egg of sirex and *Ibalia* larvae subsequently live endoparasitically for several months before finally killing the, by then, well developed sirex larva.

Of the 21 species of parasitoids imported into Tasmania the 10 species that could be reared successfully in sirex infested timber in the laboratory were liberated in the field once adequate numbers had been reared; originally attempts were made to establish all species in a forest in the north and another in the south of Tasmania to establish a "bank" for later distribution (TAYLOR 1976) but now that sirex is rare in these areas parasitoids other than *I. leucospoide* are absent. Four species of parasitoids were released annually in Victoria from 1970 to 1985 totalling 168,117 *I. leucospoide*, 23,397 *M. nortonii*, 6537 *R. persuasoria* and 1412 *R. hoferi* (with approximately equal numbers of males and females except for *R. hoferi*) (NEUMANN et al. 1987). *S. cinctipes* and *C. tasmaniensis* were released in much lower numbers in the laboratory on timber infested naturally in the field. However as sirex has become rarer in areas within reach of the rearing facilities it has become increasingly difficult to rear parasitoids.

Unfortunately, at least on the mainland of Australia, these parasitoids rarely kill a total of more than 30 - 40% of the sirex and are not particularly density dependent. *R. persuasoria* and *R. hoferi* appear to have completely failed to establish in Victoria although both bred successfully in captivity (NEUMANN et al. 1987) successful parasitoid, *Ibalia* spp., was limited to between 14.5% and 28.9% parasitism in various localities in Victoria and they have also failed in South Australia and New South Wales. Although *M. nortonii* produced high levels of parasitism in Tasmania (TAYLOR 1978) it accounted for less than 12% parasitism in Victoria (FRY 1981, NEUMANN and MOREY 1984) and the most successful parasitoid, *Ibalia* spp., was limited to between 14.5% and 28.9% parasitism in various localities in Victoria from 1977 to 1983 (NEUMANN et al. 1987, Table 2). The situation has been similar in South Australia (Underdown pers. comm.) and New South Wales (Eldridge pers. comm.).
Thus while several of these parasitoids are highly spectacular in appearance they are unlikely to contribute much to the biological control of sirex at least in mainland Australia.

**TABLE 2. Parasitism of Sirex noctilio by Ibalia spp. assessed from 1.8 m long P. radiata billets from seven Victorian plantations (After NEUMANN et al. 1987)**

<table>
<thead>
<tr>
<th>Year</th>
<th>Locality</th>
<th>N* of billets (approx.)</th>
<th>Estimated N* of available sirex</th>
<th>Total N* emerging Ibalia</th>
<th>Mortality of sirex due to Ibalia</th>
</tr>
</thead>
<tbody>
<tr>
<td>1977</td>
<td>Piries</td>
<td>760</td>
<td>40,805</td>
<td>8,718</td>
<td>21.4 %</td>
</tr>
<tr>
<td>1978</td>
<td>Delatite</td>
<td>110</td>
<td>6,178</td>
<td>1,319</td>
<td>21.3 %</td>
</tr>
<tr>
<td>1978</td>
<td>Cathedral Valley</td>
<td>740</td>
<td>56,908</td>
<td>8,230</td>
<td>14.5 %</td>
</tr>
<tr>
<td>1979</td>
<td>Delatite</td>
<td>770</td>
<td>95,727</td>
<td>24,813</td>
<td>25.9 %</td>
</tr>
<tr>
<td>1980</td>
<td>Warrenbayne</td>
<td>830</td>
<td>39,247</td>
<td>9,201</td>
<td>23.4 %</td>
</tr>
<tr>
<td>1981</td>
<td>Beechworth</td>
<td>1050</td>
<td>48,919</td>
<td>13,440</td>
<td>27.5 %</td>
</tr>
<tr>
<td>1982</td>
<td>Billywing</td>
<td>220</td>
<td>18,535</td>
<td>3,047</td>
<td>16.4 %</td>
</tr>
<tr>
<td>1982</td>
<td>Myrtleford</td>
<td>220</td>
<td>15,714</td>
<td>1,991</td>
<td>12.7 %</td>
</tr>
<tr>
<td>1983</td>
<td>Billywing</td>
<td>220</td>
<td>18,096</td>
<td>5,220</td>
<td>28.8 %</td>
</tr>
</tbody>
</table>

1 Estimates based on number of exit holes in billets
2 *Ibalia leucospoides* and *I. ensiger*

**NEMATODES**

Only *D. siricidicola*, of seven species of Deladenus (BEDDING 1968, 1975, AKHURST 1975a) found parasitising 31 siricid and parasitoid hosts from 31 tree species and 29 countries (BEDDING & AKHURST 1978) was found to be suitable for the control of sirex in Australia (BEDDING 1984). This nematode is strongly density dependent and can achieve levels of parasitism approaching 100%. Its unusual and complicated biology (BEDDING, 1967, 1972, Fig. 1) has been exploited for the biological control of sirex (BEDDING & AKHURST, 1974, BEDDING, 1979, 1984, 1992a).

*Deladenus* species have non-parasitic cycles feeding on the symbiotic fungus of siricids. This discovery enabled the storage of a library of cultures of different species and strains of *Deladenus* to be established and stored for evaluation as suitable biological control agents. In addition, the fungal feeding stage could be mass produced and liberated in the field.

*Deladenus* species were dissected from thousands of insects from hundreds of sources (BEDDING & AKHURST 1978). These nematodes would have died within a few days had it not been possible to culture them on the symbiotic fungi of their hosts. As it was, hundreds of monoxenic nematode cultures were readily established on plates, sub-cultured onto slopes in tubes and placed under refrigeration after initial establishment to build up a library of species and strains.

Hundreds of isolates of seven species of *Deladenus* were screened for potential to control sirex. However, most siricids are associated with *Amylostereum chailletti* and this is also the only fungus on which five species of *Deladenus* can feed (BEDDING & AKHURST 1978) whereas the symbiotic fungus of *Sirex noctilio* is *A. areolatum*. *Deladenus wilsoni* was able to feed on both species of *Amylostereum* but frequently parasitised the insect parasitoids *Rhysa* spp. and so this left only strains of *D. siricidicola* for further consideration. Many strains of this species did not fully sterilise Australian *S. noctilio* (BEDDING 1972) and were accordingly eliminated.
At the next stage of choosing a suitable strain, hundreds of randomly selected sirex infested logs were inoculated with various strains of *D. siricidicola*. Four of these (Corsican, Thasos, Sopron and New Zealand) produced levels of nearly 100% parasitism (BEDDING & AKHURST unpublished). It was expected that those sirex which could fly furthest and oviposit the most would be most suitable for distributing the nematode and it was found that sirex parasitised by the 198 strain from Sopron, Hungary were significantly larger than those parasitised by other species. Not only could these fly further but they also produced more eggs and more nematodes. Therefore, although some releases were made of the other strains, the bulk of releases in Australia have been of the 198 strain and this strain has also been liberated in Brazil (BEDDING & IEDE unpublished).

**National Strategy for Australia**

The massive outbreak of sirex in the "Green Triangle" altered forestry departments in all states to the importance of sirex, stimulated the formation of a new National Sirex Coordination Committee and led to new research work on sirex being undertaken. It also led to the formulation of a National Sirex Strategy for Australia (HAUGEN *et al.*, 1990) concerned with:

- national sirex coordination
- sirex awareness campaigns
- quarantine
- sirex detection
- sirex monitoring
- silvicultural control
- culture and supply of biological control agents
- nematode introduction
- parasitoid introduction
- evaluation of biological control agents
- research priorities

In conjunction with the strategy, the National Sirex Coordination Committee (NSCC) also produced a series of worksheets "in order to ensure a consistent and high standard for the various operations which form the National Sirex Control Strategy" (NSCC 1991). The national strategy will be updated as required and certainly recent developments concerning nematode control alone make this very necessary.

It is important to consider the strategy as a whole, but space does not permit further elaboration of most aspects here. It is generally agreed that nematodes stand out as by far the most important control agents and so, much of the remaining document concerns these.

**Nematode Control in Jeopardy**

A major problem has arisen in nematode control of sirex within infested regions treated in the last 5 years or more in both Australia and Brazil. This affects many of the latest developments with nematodes and is explained by BEDDING (1992a) as follows:

Although the massive outbreak of sirex in the "Green Triangle" was rapidly brought under control following an intensive treatment with nematodes, there were strong indications that the all important nematode control of sirex throughout Australia could be in jeopardy. A symptom of this was that whereas 20 years ago inoculations inevitably resulted in nearly 100% parasitism (BEDDING & AKHURST 1974), recent inoculation of trees produced levels of parasitism less than 25% (Underdown pers. comm.). Apparently there had been a steady
decline over several years but unfortunately it was only as a result of the “Green Triangle” outbreak that the importance of this was noticed.

A possible reason for the decline could have been progressive changes in inoculation techniques that were made during a series of personnel replacements. However, investigations by HAUGEN & UNDERDOWN, (in press), showed that this was not the problem.

It has now been determined that declining parasitism over several years in inoculated logs is almost certainly a result of a genetic change in the nematodes used. While use of the fungal feeding cycle for maintaining cultures and mass producing *D. siricidicola* has been an essential part of managing this nematode for biological control, it has also resulted in a major problem (BEDDDING 1972a, b).

The continual sub-culturing of *Deladenus* in the free-living form for over 20 years without intervention of the parasitic life cycle has led to the selection of a strain which rarely forms the parasitic stage. Even at high concentrations of CO₂ and low pH such cultures will now rarely produce infective females. It was initially hoped that, since in the wild *D. siricidicola* had a large number of free-living cycles with only one annual parasitic cycle, this would not be a problem. However, the situation in the field is more complicated than this. In the absence of *sirex* larvae, the CO₂ levels in the tree are usually not high enough and the pH not low enough to promote development of infective females. Consequently infective females will only be produced in the vicinity of *sirex* larvae, that is, where they have the opportunity to pass their genes on to a succeeding generation.

Thus there is a little or no selection against a predisposition to develop into infective females in the field, but the converse is true when *D. siricidicola* has been cultured artificially. The nematodes pass through repeated generations without intervention of a parasitic cycle (current cultures have been through hundreds of generations). Infective female nematodes produced in these cultures cannot reproduce because there is no host and so there has been a selection pressure against their production.

The production of low levels of parasitism in inoculated logs was unfortunate and costly (four times as many trees needed to be inoculated), but of far greater significance is what this means in terms of the ability of this defective strain of nematodes to control *sirex* populations. There is every reason to expect that nematode control with the defective strain may not occur until *sirex* infestations are severe (perhaps >10% tree death) whereas the original strain produced high levels of parasitism at very much lower tree death (probably <1%). Recent results from New South Wales and South Australia tend to confirm this (Eldridge & Underdown pers. comm.). The defective strain was almost certainly only effective in the “Green Triangle” because of the very high density of *sirex* infestation (up to 80% tree death) and highly intense liberation.

Since it is important to obtain control, with limited human input, long before tree deaths become economically significant, it was necessary to obtain a more effective strain of *D. siricidicola*. This was achieved by collecting *sirex* infested timber from the Kamona forest in Southdale, Tasmania where it was first liberated in 1970 but where no subsequent liberations had taken place (Bashford pers. comm.). Nematodes were found in and extracted from just one of only nine trees found, established in monoxenic culture on *A. areolatum*, and used for subsequent liberations while steps were taken to ensure that such a decline in infectivity did not recur.
LATEST DEVELOPMENTS IN THE LABORATORY CULTURE AND INOCULATION OF THE PARASITIC NEMATODE Deladenus siricidicola

LABORATORY CULTURE OF Deladenus

*D. siricidicola* is cultured in its fungal feeding cycle as described by BEDDING & AKHURST (1974). Firstly inoculum is produced by growing monoxenic cultures of the nematode together with the symbiotic fungus of sirex, *Amylostereum areolatum*, on potato dextrose agar plates. Then these cultures are used to inoculate autoclaved hydrated wheat in 500 ml Erlenmeyer flasks which are harvested with water 6 to 8 weeks afterwards. This technique has been used to produce *D. siricidicola* for field release since 1970 including the 40,000 million required during 1987-8 to combat the massive sirex infestations which occurred in the "Green Triangle".

Mass production of the "defective" (previously termed "Old") strain is relatively easy since it has been cultured under laboratory conditions for over 20 years. The newly isolated Kamona strain which should be used for all future liberations is however much more difficult to culture because it has a lower fecundity and is less able to hold back fungal growth. (Nematodes only grow and reproduce well on the advancing front of the fungus). There are the additional problems associated with ensuring prevention of the Kamona strain also eventually becoming "defective". The essential changes facilitating culturing the Kamona strain are described in the appendix (BEDDING & CROMPTON in preparation).

LIQUID NITROGEN STORAGE

Since it may not always be possible to re-isolate the "Kamona" strain from the field, it is obviously most important to preserve its genetic integrity indefinitely in 5% glycerol and slowly evaporating water from a suspension of *D. siricidicola* in 5% glycerol to 50% glycerol under sterile conditions prepared nematodes for indefinite storage in liquid nitrogen with over 75% survival after 18 months.

Many hundreds of vials of the Kamona strain have been stored in this way so that each year all cultures used for release will be re-initiated from this stock. At the same time, all sub-culturing will be conducted so as to eliminate or minimise production of infective females during artificial culture by avoiding lowering of pH and accumulation of carbon dioxide.

NEMATODE LIBERATION AND DISPERSAL

Parasitised female sirex introduce nematodes into fresh trees when they oviposit and this leads to nematode infection of the progeny of non-parasitised sirex ovipositing in the same tree. This is the main means by which nematodes are spread within a forest. However, although nematodes can sometimes be spread from forest to forest by sirex females (BEDDING 1979) this is too unreliable and may occur too late in the infestation to prevent a serious outbreak. Nematodes are therefore introduced as early as possible by artificially inoculating trees already infested by sirex at easily accessible, strategic points within a forest. As described by BEDDING & AKHURST (1974), important features of inoculation are cleanly cutting streaks to make inoculation holes, using as inoculum of nematodes suspended in gelatine foam, inoculating timber of adequate moisture content and using correct inoculum size and spacing.

BEDDING & AKHURST (1974, and unpublished data) showed that the better the distribution of inoculated timber throughout a forest area, the more rapidly high levels of nematode parasitism are likely to be induced throughout. However, this may be unnecessarily expensive if infestation levels of sirex are at very low levels (<1% trees infested). In this case, inoculation of trap trees (MADDEN & IRVINE 1971, NEUMANN et al. 1982, 1989) located every 100 metres along forest boundaries (HAUGEN et al. 1990a) can be quite adequate since a high proportion of the sirex ovipositing during the next season will have come from these trap
trees and will therefore be parasitised. Where levels of sirex are any higher than this though, sirex emerging from trap trees will represent only a small part of the population and it is important to inoculate sirex infested trees scattered throughout the plantation.

In areas of the "Green Triangle" which had up to 80% sirex infestation, every sirex-infested tree in each fifth row was injected with nematodes (20% of all trees and a total of 106,000 trees inoculated). This led to high levels of parasitism during the next year after that accompanied by a dramatic decline in the sirex population (Underdown pers. comm.).

**Liberation of the Kamona Strain**

At this stage, *D. siricidicola* has been liberated over nearly 500,000 hectares of pine forest in southern Australia. Throughout the large forests of the states of New South Wales and South Australia and in Brazil, all the nematodes have been liberated recently and have therefore (prior to 1991) been of the "Defective" strain. Liberations in Victoria and Tasmania have been made since the early 1970’s and it is not yet known when cultures started to become defective but it will be important to find out in order to know where the Kamona strain should be introduced in an effort to replace the Defective strain. When the Kamona strain is liberated into areas where there is already Defective strain it will also be important to know whether it has become fully established.

Since the Kamona and Defective strains have been derived from the same source strain, it is not possible to distinguish them morphologically and so research is currently being conducted (Curran pers. comm.) to develop DNA probes to separate them and monitor the spread of the field released Kamona strain of *D. siricidicola*. Success of this project would eventually allow for the rapid identification of the strain of nematode present in any parasitised sirex or woodchips sample. Where Kamona strain was found to be absent there would obviously be a need to introduce it. The most easily prepared probes are of repetitive DNA and such probes can be used early in the program to determine whether or not pure Kamona or pure Defective strain is present by testing with probes both for Defective strain and for Kamona strain. However, since these probes will not be of that portion of DNA responsible for infectivity, crossbred populations may be positive for the probe but still be defective in infectivity. Eventually it may be possible (Curran pers. comm.), after finding the precise stage at which juvenile *D. siricidicola* are induced to switch their development, to identify the proteins or messenger RNA mediating this switch. These could then be used to make probes which would be specific for infectivity genes.

The ability of *Dendrobatena* to form infective females on culture plates may well mirror infectivity in the field. However, more exact tests are being developed which may be able to determine whether nematodes from particular areas, liberated at some time in the past or from mixed populations of the Defective and Kamona strains, are likely to be adequately infective or not. So far it has been determined that incorporating 0.2% lactic acid into culture plates and then maintaining cultures of *A. areolatum* and *D. siricidicola* on these in an atmosphere of 20% carbon dioxide, results in levels of infective female production ranging from 1-2% in the Defective strain to 100% in the Kamona strain and 60% in the crosses (BEDDING & CROMPTON in prep.). Eventually it should be possible to develop more precise tests to screen nematodes from the field to determine whether they are infective enough or whether further introductions of Kamona strains need to be made.

**Conclusion**

An understanding of what is required to manage sirex has only just been arrived at forty years after the arrival of this most important pest of Australia's nearly 1 million hectares of exotic pine. This is partly because of the complexity and interactions of a whole range of factors that affect the situation. Also the pest itself is likely to cause devastating damage only spasmodically in response to particular combinations of environmental and other factors; this absence of serious damage for long periods of time can easily lead to complacency. A problem
is that because of the very long period (>10 years) over which particular radiata plantings can be susceptible to sirex it is almost inevitable that, at least once in the life of radiata forest, conditions will be favourable for a major sirex outbreak to develop if appropriate protective measures are not already in place. The Australian National Sirex Strategy (HAUGEN et al. 1990) describes what we believed these measures should be. In addition to this we now have the problem of ensuring that the Kamona strain of nematodes replaces the defective strain and unfortunately this is also true for Brazil.

This and other problems will of course be overcome and there is no reason why, provided the situation is continually monitored, that this nematode should not be used for hundreds of years to come to control the very serious pest, S. noctilio, not only in Australia but in many other places in the world to where this insect will doubtless eventually spread.

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