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CONTROL OF SIREX NOCTILIO (F.) WITH DELADENUS SIRICIDICOLA BEDDING

Part I - 1967 Field Trial

R. ZONDAG

Forest Research Institute, New Zealand Forest Service, Rotorua

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ABSTRACT

Four methods were used in a trial to establish **Deladenus siricidicola** Bedding, parasitic nematodes of **Sirex noctilio** (F.), in three forests in the South Island. All gave positive results. The most promising and the easiest for future use is to extract the **Deladenus** from wood, to place them in test tubes filled with wood chips and subsequently to insert the contents of the tubes in holes drilled in trees that have been successfully attacked by the woodwasp.

INTRODUCTION

The importance of the parasitic nematode of Sirex noctilio (Fabricius) described by Bedding (1968) as Deladenus siricidicola for the biological control of the woodwasp was realised soon after its discovery in 1962. The complete sterilisation of infected adults and mortality of heavily infected immature stages of the woodwasp make this organism a very effective controlling agent for reducing populations of Sirex in New Zealand (Zondag, 1969). High percentages (up to 96%) of infection of adults emerging from logs in North Island forests have been recorded on several occasions, but until the trial in 1967 no infection by these nematodes was ever found in Sirex obtained from South Island forests.

Study of the life history of *Deladenus siricidicola* suggested methods by which it could be disseminated.

The methods used were based on the fact that the nematodes, after insertion in trees attacked by nematode-free *Sirex*, would move through the timber and would mature, reproduce, and repeat the cycle. The insect-infective stage of the nematodes would then infiltrate the woodwasp larvae, wherein they would grow, mature, and their progeny sterilise the adult insects.

The most obvious method of disseminating the nematodes is by the release of adult female *Sirex*, since under natural conditions they are the only agents to insert the nematodes into the trees. Logs collected from trees successfully attacked by *Sirex* and showing a high percentage of nematode infection of the various stages of the woodwasp could be placed directly in forests, or in insectaries from which the females could be released in selected areas.

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The nematodes can, however, be introduced into *Sirex*-attacked trees by inoculating them. The nematodes could be obtained by extracting them from the wood, from *Sirex* adults of both sexes, or by mass rearing them on the *Sirex* fungal symbiont.

Adult Sirex can be readily obtained in the summer months by placing logs from Sirex-attacked trees in the insectaries; to obtain the adults in winter the logs are placed in cold storage from November and placed in temperature-controlled rooms in the winter months.

In the trial four methods were tested, with the aim of limiting the possible infection of *Sirex* to certain trees only. The nematodes had therefore to be inserted in the particular trees and the liberations of adults was not considered for the trial. The mass rearing of the nematodes has not been developed. It was planned to inoculate some trees in the winter with nematodes extracted from adults, but for some unexplained reason the number of *Sirex* emerging in winter was too low. A large number emerged in November and December too late to incorporate the method in the trial.

Three Forest Biology Observers in the South Island selected stands for the trial. Areas in Rai-Whangamoa State Forest in Nelson Conservancy, Omihi State Forest in Canterbury Conservancy, and Rankleburn State Forest in Southland Conservancy were chosen. No nematode infection of woodwasps has been found in these forests, in all of which some tree mortality associated with *Sirex* attack had been observed in the previous year. It was difficult to predict which trees, if any, would succumb to *Sirex* attack in 1967. For this reason, and to keep the trial within relatively small areas, it was necessary to precondition trees to *Sirex* attack.

Logs containing *Sirex* infected by nematodes were obtained from Kaingaroa and Esk State Forests in the North Island. Approximately 75% of the *Sirex* adults emerging from these logs were found to be infected by nematodes.

THE TRIAL

Preconditioning Trees to Sirex Attack

Of the several methods of preconditioning radiata pine to induce *Sirex* attack, the most promising under the circumstances was to prune standing trees to high levels and remove a 6 to 8in. ring of bark and cambium around the stem below the lowest remaining green branch.

Sirex attack on trees so treated during the flight season may occur within five days and continue for some time afterwards. The method has the great disadvantage that the trees are not always attacked and even when attacked the Sirex larvae and the symbiotic fungus often fails to develop in the lower parts of the trunk. The preconditioning could probably be improved by removing also a strip of bark and cambium near ground level to ensure the tree died quickly. This weakening had to be avoided in the trial, because it was intended to insert the nematodes in the standing trees.

Other methods of preconditioning pines to *Sirex* attack which have met with some success in the past, are to sever trees at ground level and leave them lodged against other trees, or to fell and prunes trees before the flight season. *Sirex* attack on trees so treated is unpredictable but is usually more successful when a moderate population of *Sirex* is present in the area.

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Methods

The following methods were used in the trial:

Method A. Nematode-infected Sirex females with the wings tied together were placed on trees attacked by the wild population of Sirex.

Method B. Nematodes extracted from Sirex adults (mostly males) were placed in holes drilled in trees attacked by the wild population.

Method C. Abdomens of nematode-infected Sirex adults (mostly males) were placed in holes drilled in trees attacked by the wild population.

Method D. Nematodes extracted from wood in which nematode-infected Sirex were found, were placed in holes drilled in trees attacked by the wild population.

A supply of Sirex and nematodes to carry out the work by the four methods was sent from the Forest Research Institute to the Forest Biology Observers after the trees had been preconditioned.

Because no differences between infected and uninfected insects can be detected by external examination, it was not certain that all the insects forwarded were infected.

Method A

No. 1

Sirex females recently emerged in the insectaries from logs originating from Kaingaroa and Esk State Forests had their wings tied together with thin nylon thread very close to the wing base but not so close that the muscles were under stress. The part of the wings above the knot was cut off. The females with the wings properly tied together lose their flight stimulus and generally remain on the tree. When the wings are cut off but not tied the flight stimulus is not impaired and, in trying to fly, the insects drop off the trees. The insects were forwarded to their destination on the same day by the quickest possible means. On arrival, the active females were placed on selected preconditioned trees, already attacked by the wild population.

After release on the trees the majority of such females usually oviposited straight away, but some remained sluggish for some time. Oviposition is generally confined below the ringbarked area of the preconditioned trees.

Method B

Sirex males and females were dissected after arrival at their destination. The testes and ovaries with eggs were removed, placed in water, and teased open to free the nematodes. More water was added at the forest site and the nematode suspension placed in $\frac{3}{8} - \frac{1}{2}$ in. holes drilled in trees already attacked by the wild population. The holes were then blocked with cellulose wadding after more water had again been added.

Method C

Sirex, on arrival at their destination, had the abdomen cut off. In the males the posterior black segments and in the females the ovipositor components and the last abdominal segments were also removed. The remains of the abdomens, containing the testes or the eggs, were then placed in 3in. holes drilled in trees which had already been attacked by the wild population, and squashed hard against the bottom of the hole. The membranes had to be ruptured to ensure that the nematodes in the testes and eggs dispersed. Some water was added to the holes, which were then stoppered with cellulose wadding.

Method D

Nematodes were extracted from logs as an aqueous suspension (Zondag, 1969). Surplus water was removed and the suspension was then examined for the presence of Deladenus siricidicola. If extractions yielded a large number of other species of nematodes commonly found in dead wood, the batches were rejected, even though D. siricidicola was also present. Batches containing 75% or more of D. siricidicola were placed in test tubes filled with wood chips, obtained by machine-planing a piece of Sirex-infected timber, crushing the shavings, and sieving them until ½-½in. pieces were obtained. More water was added to the test-tubes when necessary, so that the wood chips were moist. Each test-tube contained at least 300 nematodes, but the figures could be as high as 2,000. The test-tubes were held in cool storage until sufficient were obtained. Samples were examined at regular intervals to confirm that the nematodes were still alive. Tubes were kept no longer than two weeks before being forwarded for the trials.

In the field, the contents of the tubes were placed in $\frac{1}{2}$ -1in. holes drilled in trees attacked by *Sirex*. More water was added to the holes and the holes stoppered.

With Methods B and C, Sirex emerging from the logs in the insectaries can be placed in cool storage for a few days until a sufficient number are obtained. Insects dead on arrival at their destination were still used for both methods as the nematodes remain alive for at least 2 days after the death of the insects. With both methods males were generally used; only those females which were dead or unsuitable for liberation were dissected.

Methods A, B, and C were applied to each of the selected trees on three different days. The total number of *Sirex* used for each method varied in the different forests as did the number of holes drilled in each of the selected trees.

All the holes were drilled to near the centre of the tree or at least 3in. deep, in the preconditioned trees from about 6in. below the ringbarked area down to a height of 6-8ft from the ground.

The trees to which Method D was applied were treated only once. In these, whether standing or felled, holes were drilled about 1ft apart.

The number of *Sirex* used and the number of holes drilled in each of the trees from which samples were taken are recorded in Tables 1, 2, and 3.

The preconditioned trees in Rai-Whangamoa and Omihi State Forests did not show by September the characteristic foliage wilting, indicative of successful attack by Sirex and its fungal symbiont, although many oviposition holes were found. In both forests, however, several trees, not preconditioned, revealed obvious signs of successful Sirex attack and some of these were selected to apply Method D. To facilitate the insertion of the nematodes the selected trees were felled and the nematodes inserted into those parts of the trees where Sirex larvae could be located.

THE TRIAL AREAS AND TREATMENTS

Rai-Whangamoa State Forest (Nelson Conservancy)

The trial in this forest was carried out in Compartment 9, in which radiata pine was planted in 1952. The stand was low pruned to 6ft in 1961, and 120 stems per

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acre (s.p.a.) were high pruned to 18ft in November 1964. Thinning by poison (ammonium sulphamate) was done in February-March 1965. No tree mortality associated with backflash* of the poison occurred in the following year. The number of trees left in 1967 was approximately 250 per acre, with an average height of 56ft and a diameter range of 6 to 12in. at breast height. Sirex were present in the stand, as trees which did not immediately succumb to the poison application created a suitable breeding habitat.

Nineteen more or less suppressed trees were preconditioned on 31 January 1967. The trees were pruned up to 25ft and ringbarked at 18-23ft height.

Methods A, B, and C were all applied on 3, 16, and 25 March. Treatment A was used on three trees, treatment B on two trees, and treatment C on three trees. As hardly any definitely successful attacks by *Sirex* had occurred on these preconditioned trees, a naturally attacked tree was included for treatments B and C. Even in September, when Method D was carried out, hardly any wilting was observed on the remaining preconditioned trees in the same area. However, several untreated trees were dying after successful attack. Four of these trees were felled and Method D applied.

Even in November, when the samples were collected, many of the preconditioned trees had not died. Only a few showed wilting of the foliage, but not to the extent encountered in the naturally-attacked trees. Some of the preconditioned trees which showed some wilting were felled. Examination showed that the bark of these trees was still green at 15ft and sometimes higher; no *Sirex* larvae were found in the green sections.

Omihi State Forest (Canterbury Conservancy)

The trial was carried out in Compartment 1, in which radiata pine was planted in 1949. The stand was low pruned in 1957-58 and high pruned in 1960 and 1962. Thinning was done by poisoning the unwanted trees with ammonium sulphamate between September 1964 and January 1965. The poisoning resulted in severe mortality of the unpoisoned trees through backflash of the poison, followed by Sirex attack. By July 1966 it was found that approximately 50% of the unpoisoned trees were killed in parts of the area, and further mortality occurred in 1966. By January 1967 the stocking of the compartment was approximately 160 s.p.a., the average height was 55ft, and the diameter at breast height varied from 6 to 11in.

In the area allotted for the trial the average height and diameter of the trees were generally lower than the average for the stand. Because the trial area was on the steep slopes of a gully, treatment was extremely difficult.

Eighteen trees were preconditioned on 16 January 1967, by pruning to a height of 15-18ft, and ringbarking below the lowest green whorl. Most of these trees were attacked by *Sirex*, but many of them still had their green crown in November 1967 and others which showed signs of wilting had green bark at a height of 10ft.

Methods A, B, and C were all applied on 7, 16, and 22 February 1967, in standing trees. Three trees were treated by Method A, three trees by Method B, and two trees by Method C. Method D was carried out on 19 September 1967 on six naturally-attacked trees, which were felled before treatment.

^{*} Translocation of poison via root graft to unpoisoned trees.

Rankleburn State Forest (Southland Conservancy)

The trial was carried out in compartment 21 where radiata pine was planted in 1955. This stand, which carried approximately 700 trees per acre, was pruned in 1965. The average height of the trees was between 35 and 40ft and their diameter at breast height ranged from 4 to 8in. Scattered trees had died following *Sirex* attack in the previous year.

For the trial, 20 trees were preconditioned on 17 February 1967. The trees were pruned up to 35ft, much higher than in Rai-Whangamoa and Omihi State Forests. Only 1-3 whorls of green crown were generally left. Ringbarking was done at heights varying from 28 to 33ft.

Methods A, B, and C were all carried out on 24 February, 3 and 10 March. For each method three standing preconditioned trees were selected. Eight preconditioned standing trees were treated by Method D on 20 September.

Sirex attacked the preconditioned trees and the majority succumbed.

THE SAMPLE LOGS

Samples were collected from the trial areas in the third week of November 1967. The sample logs were all 3ft 8in. long and were collected from those parts of the trees in which *Sirex* larvae or their tunnels were present. The samples were taken at random below and above the ringbarked areas of the preconditioned trees. With treated naturally-attacked trees the samples were taken from parts in which no nematodes were inserted.

In Rai-Whangamoa and Omihi State Forests some of the treated preconditioned trees had been attacked unsuccessfully by *Sirex*. The crown was still green in November 1967 and although some dead patches of bark occurred below the ringbarked area, there was no evidence of development of larvae and no samples were taken from such trees.

Sample logs from naturally-attacked untreated trees were taken as controls. In Rankleburn State Forest only one naturally-attacked tree could be found in the same compartment, and this yielded only two small logs containing Sirex larvae.

The remains of the treated trees were left in the forests, so that the emerging infected adult Sirex females could introduce the Deladenus into other trees.

The sample logs were sent to the Forest Research Institute in Rotorua, where each log, with the exception of the control logs, was placed in a separate cage (4ft high) to await emergence of the insects.

The Sirex noctilio and its parasites (Ibalia spp. and Rhyssa persuasoria (L.)) emerging from the sample logs and control logs, were collected at least twice weekly and dissected to determine whether they were infected or not. Only a very small number of R. persuasoria emerged from these logs, but none was found infected.

RESULTS

The results of the dissections are given in Tables 1, 2, and 3 and summarised in Table 4. These tables show how and when the trees from which the samples were taken were treated.

*Percentages rounded off and not given where less than 10 insects emerged. Methods A, B, and C all applied 4, 15, and 27 Mar 1967.

Method D applied 25 Sep 1967.

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*Percentages rounded off and not given where less than 10 insects emerged. Methods A, B, and C all applied 7, 16, and 22 Feb 1967.

Method D applied 21 Sep 1967.

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TREATMENT OF TREES

TABLE 2-Omihi State Forest. Treatment of trees and emergence of Sirex and Ibalia from sample logs

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TABLE 4-Summary of results of trial

						Sire	<		lbal	Ibalia			
Method of treatment	Forest	No. of trees treated	No. of trees sampled	Number of samples	No. emerged	No. infected	% infection*	No. emerged	No. infected	% infection*			
A	Rai-Whangamoa	3	1	3	86	0	0	1	0	0			
	Omihi	3	2	5	18	16	89	33	1	3			
	Rankleburn	3	3	5	99	96	97	63	7	11			
	Total				$\frac{-}{203}$	112	55	97	8	8			
\mathbf{B}	Rai-Whangamoa	3	2	4	19	10	53	2	0	0			
	Omihi	3	3	7	27	2	7	24	0	0			
	Rankleburn	3	3	3	168	98	58	57	12	21			
	Total				214	110	51	83	12	14			
\mathbf{C}	Rai-Whangamoa	3	2	6	57	5	9	15	0	0			
	Omihi	2	2	5	61	11	18	30	3	10			
	Rankleburn	3	3	6	234	156	67	52	10	19			
	Total				352	172	49	97	13	13			
D	Rai-Whangamoa	4	4	4	462	235	51	76	1	1			
	Omihi	6	4	12	928	272	29	432	1	0.2			
	Rankleburn	8	5	9	300	259	86	57	3	5			
	Total				1,690	766	45	565	5	0.9			
All	Rai-Whangamoa	13	9	21	624	250	40	94	1	1			
	Omihi	14	11	29	1,034	301	29	519	5	1			
	Rankleburn	17	14	25	801	609	76	229	32	14			
jo.	Rai-Whangamoa		3	6	343	0	0	36	0	0			
Control	Omihi		3	3	334	0	0	227	0	0			
చ	Rankleburn		1	2	67	0	0	0	0	0			

^{*} Percentage rounded off.

The tables show that all four methods used in the trial gave some positive results in obtaining infection of the *Sirex* by *Deladenus*. The most satisfactory results were obtained in Rankleburn State Forest where preconditioned trees were more successfully attacked by *Sirex* than they were in the other two forests.

In several of the samples from which none or a very low number of insects emerged, it was observed that many of the *Sirex* had died in the larval stage. This was not unexpected, as observations in the past showed that *Sirex* larvae heavily infected by *Deladenus* had little chance of survival. It could not be determined, however, whether the mortality was caused by heavy infection of the nematodes or by other factors, as dead and shrivelled larvae do not readily reveal whether or not *Deladenus* had been

present when the insects were alive. It was found previously that some of the larvae in logs were heavily infected, some normally infected, and others not at all.

After the emergence of the insects had ceased, the number of exit holes was counted. This showed that very few insects had escaped from the cages and were not accounted for.

In May and June 1968, extractions for nematodes were made from several of the sample logs, especially those which yielded no nematode infection of *Sirex* and *Ibalia* or from which no *Sirex* had emerged. It soon became obvious that it was too late in the year to make these extractions, as most of the wood had dried up. The results were inconsistent; even from several sample logs from which a high number of infected *Sirex* had emerged no *Deladenus* were obtained by extraction, although other species of nematodes were present. From some of the sample logs from which no *Sirex* emerged, *Deladenus* could be extracted.

Twenty sample logs were taken above the ringbarked area or from those parts in which no nematodes were inserted. Twelve of these showed nematode infection of the emerging insects; thus indicating that the nematodes moved away from the place where they were inserted. Infection of sample logs taken 5ft away from the nearest insertion hole was recorded. In one sample log taken from a tree treated by Method A, the possibility existed that the nematodes were inserted by an infected female moving past the ringbarked area.

In several of the preconditioned trees, the holes drilled to apply Methods B and C were approximately 6in. apart, but it appears that the holes could be more widely separated and that it may be sufficient to apply a treatment only once.

An example of this is seen by comparing trees R3 and R5 treated by Method C at Rankleburn. R3 was treated only once, and eight holes were drilled. R5 was treated three times and 24 holes were drilled; in each hole three male *Sirex* were inserted. (In tree R5 the holes were approximately 6in. apart.) Despite these differences in treatment, the percentages of infection of the emerging adults were very similar.

Considering that the treatment by Method D had been carried out in late September, the results were satisfactory. Less time was available for the nematodes to infect the Sirex immature stages than with the other methods. It may be for this reason that a deviation from the normal pattern of infection was found in many specimens emerging from the sample logs taken from trees treated by this method. In a number of adult male Sirex the gonads were infected by 3-4mm-long, stubby nematodes but no large females were found in the haemocoel.

The insects referred to under the heading *Ibalia* in the tables are mainly *I. leucospoides* (Hochenwarth) but some which emerged from the sample logs from Omihi State Forest were *I. ensiger* (Norton), which were released in 1966. Only 39 of a total of 841 *Ibalia* which emerged from all the sample logs were found to be infected by nematodes. All the nematodes were dead and no infection or distortion of the reproductive organs was observed.

Infection of *Ibalia* species was found in only 14 of the sample logs, from which a total of 251 *Ibalia* emerged. Five of these logs showed 100% infection of the *Sirex*;

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the tables are mainly the sample logs from d in 1966. Only 39 of re found to be infected or distortion of the

ple logs, from which a infection of the Sirex;

four ca. 90%; two ca. 75%; one 53% and one 13.8%, while from one sample (R.10/1) from which no Sirex emerged one of the two Ibalia which emerged was infected. The total number of Sirex which emerged from these 14 sample logs was 669, of which 502 (ca. 75%) were infected.

The high figures for parasitism of *Sirex* by *Ibalia* species in some of the sample logs cannot be regarded as unusual. When *Sirex* larvae are heavily infected by *Deladenus* they may die before they reach the adult stage. The number of nematodes in *Sirex* larvae already parasitised by *Ibalia* is generally far lower than in normally infected specimens (Zondag, 1969), which gives the insect parasite a better chance of survival, although heavy infection of *Sirex* immatures may also result in mortality of the *Ibalia* larvae.

Under natural conditions in the field the infected Sirex female is the main vector in the dissemination of Deladenus siricidicola. The male does not play an important role as no transfer between infected males and uninfected females has been observed after copulation. The sex ratio of the Sirex which emerged from the logs is therefore important. The proportion of males to females was for Rai-Whangamoa State Forest 498:128 (ca. 4:1), Omihi State Forest 931:103 (ca. 9:1), and Rankleburn State Forest 632: 169 (ca. 3.7:1). Generally the sex ratio is approximately 10:1.

DISCUSSION

The tables show that each of the four methods used in the trial resulted in positive infection of Sirex.

The main handicap in the trial was the failure of several of the preconditioned trees, especially in Rai-Whangamoa and Omihi State Forests, to succumb to the attack of Sirex. In both these forests mortality associated with Sirex attack occurred in the same compartment close to the preconditioned trees. The majority of the preconditioned trees in these forests had the crown and the bark of the lower half of the trunk still green in November when sample logs were collected, and only in a short part of the tree had larval development occurred. It was observed after some of the trees had been felled that although Sirex attack on the lower half had occurred there was no larval development.

The very high pruning of the preconditioned trees in Rankleburn State Forest resulted in a more successful attack and subsequent killing of the tree, but here also the lower half of the trunk was not so severely attacked as the upper parts.

In establishing *Deladenus siricidicola* the preconditioning and treatment of trees while standing has the further disadvantage that it requires the use of ladders and safety belts. Naturally-attacked trees are preferred if they can be found at the right time of the year. During the *Sirex* flight season such trees, however, are difficult to locate, as the characteristic wilting and yellowing of the needles is seldom seen so early in the year. For this reason preconditioning of trees was resorted to for the trial.

Naturally-attacked trees are clearly more suitable for establishing *D. siricidicola*. If such trees could be found during the *Sirex* flight season all methods could be used.

The trees to be treated with Method A could be left standing, while those to be treated with Methods B, C, and D could be felled prior to treatment. However, if such felling was done too early in the year the wood might dry out too soon, resulting in death or inhibition of any further development of the *Sirex* larvae.

Method D appears the most suitable for establishing D. siricidicola artificially in the field. Trees successfully attacked by Sirex are felled and D. siricidicola inserted in those parts of the trees in which Sirex larvae are present. This method can be used during most of the year and has the further advantage that dying trees can be treated in winter when mortality is showing up.

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