

LOCUST AND GRASSHOPPER BIOCONTROL COMMITTEE

Newsletter Issue 2, February 2000

Welcome to our second Newsletter which shows how busy we have been for the past year! The main focus has again been on field trials aimed at improving the consistency and lowering the cost of the Green Guard™ product (hope you like the name). Progress has been very rapid and we have succeeded in substantially reducing the likely cost by lowering the effective field dose and reducing the volume of spray needed per hectare. We are now in a position to develop a package for submission to the NRA for registration. There are still many things to do and we have a box at the end of the Newsletter with our aims for 2000. If you would like a copy of our previous newsletter please let me know.

Richard Milner
Project Leader, CSIRO Entomology, GPO Box 1700,
Canberra 2601.
Phone: 0262464169, fax: 0262464042; e mail:
richardm@ento.csiro.au

The committee consists of:

Dr Graeme Hamilton, Director, APLC Chair

Dr Richard Milner, CSIRO Entomology

Dr Joe Scanlan, Qld. Dept. Nat. Res.

Mr John Bowler, NSW Agriculture

Mr Jim Wrenford, Tablelands Wingless
Grasshopper Committee

In addition, the following are key advisers:

Dr David Hunter and Mr Peter Spurgin, APLC.

Dr Greg Bender, SGB Pty. Ltd.

Mr Graeme Baker, Australian Museum.

CONTENTS

Highlights of past year	1
Field trials	2
Wingless grasshoppers	2
Migratory locusts	2
Australian plague locusts	3
Laboratory Studies	4
Publications and conference papers	4
Plans for 2000	4

Highlights of the Past Year

1. The name Green Guard™ has been registered by CSIRO for the *Metarhizium* grasshopper/locust product.
2. Two field trials during cool and hot conditions respectively gave very effective control of Australian plague locusts at a low dose of 1×10^{12} spores/ha (= 25g spore powder see Table on page 2) and in one trial in 500 ml of oil per hectare. At this low dose rate, the cost of the spray is comparable with chemical insecticides.
3. A new low-tech method using self-aerating bags has been adopted for mass production which has reduced costs and increased capacity.
4. Air-drying of large quantities of the *Metarhizium* material using a low cost drying room has been perfected resulting in a more environmentally stable product with good storage capabilities.
5. A large-scale field trial using 75g spores/ha against wingless grasshopper gave about 85% control of grasshoppers and provided excellent recovery of a severely damaged lucerne crop.
6. The NRA has extended our EUP to allow on-farm trials by landholders.
7. The new name for the grasshopper isolates, *M. anisopliae* var. *acridum* has been formally published in the prestigious international journal Mycological Research and has been accepted by key workers in the field.
8. Our commercial collaborator SGB Pty. Ltd. (part of the IAMA group of companies) has completed a new production facility and material produced by them has been shown to be very effective in the field.

FIELD TRIALS

Wingless grasshoppers

In November 1998, 3 plots of 35-70 ha were sprayed with a high dose of 125g spores/ml in 1 litre of oil per ha in the Peak View area east of Cooma. At the time of spraying, populations were of medium density (20 to 80/m²) and in 2/3rd instar with good pasture conditions. These are conditions under which grasshoppers would not normally be sprayed, however the trial proceeded because of the need to get data on the effect on wingless grasshoppers.

For the 7 first days after spraying the maximum temperatures ranged from 16-22°C and the minimums from 6 to 13°C. During the second week maximum temperatures were much warmer with temperatures over 25°C on 4 days but minimums were always below 15°C. The second and third weeks were also cool with maximum temperatures around 22 °C, while there were some days over 30°C during the 4 and 5th weeks.

TABLE. The dose of *Metarhizium* may be expressed as the number of spores (conidia)/ha or as the number of g of dry spore powder. This table compares these two methods.

g spore	no. spores
25	1 x 10 ¹²
50	2 x 10 ¹²
75	3 x 10 ¹²
100	4 x 10 ¹²
125	5 x 10 ¹²

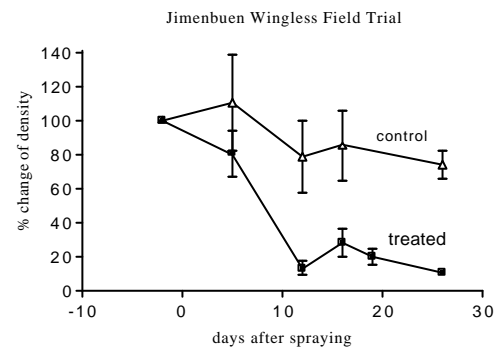
In all plots, mortality occurred after week 2 and the populations continued to decline to week 4 by which time the reductions ranged from 50-70%. Dispersal of winged species (*Austroicetes* and *Oedaleus*) and mortality of all species from nematodes caused a decline in the controls. In the treated plots, there was a clear effect of pasture height with over 80% control in short pasture and less than 50% control in long pasture. Analysis by Dr Alan Clift (Sydney University) showed that the fungus was more effective against *Austroicetes* than wingless grasshopper.

These results, while encouraging, confirm laboratory tests which show that the disease is less effective at temperatures below 20 °C, and suggest that for effective control of wingless grasshopper it will be necessary to treat when the weather is warmer and where there is very short vegetation cover.

In 1999, a single 250 ha plot at Jimenbuen (80 km south of Cooma) was sprayed from the air on December 4. The high density and dry condions meant that the grasshoppers were competing with stock justifying control and presenting an ideal opportunity for a farm-scale trial. At the centre of the area treated was a lucerne crop which over the week prior to spraying had been decimated by a population of over 100 wingless grasshoppers/m². A nearby plot was left untreated as a control. The dose use was 75g spores/l in 1 litre oil/ha.

At the time of spraying the weather was warm with a maximum of 26°C and a minimum of 12 °C. Over the first 7 days, temperatures ranged from minimums of 14°C to maximums of 30 °C and over. By day 12 after spraying the population in the lucerne had reduced by over 90% but some grasshoppers subsequently invaded from surrounding treated areas. These in turn died and so by day 26 after spraying the population was still reduced by around 85% (Figure 1). There was little reduction over this time in the untreated control plots largely because of the absence of nematode activity under these warm/dry conditions. Sampling of the vegetation indicated that the spray deposit persisted for over 10 days with a half-life of 6.5 days. However at two months after spraying the population was still high in the control plot but remained low in the lucerne as a result the crop had made a spectacular recovery.

Figure 1:



Migratory Locusts

Since the first Newsletter, a further two large-scale field trials have been completed against migratory locusts in the Central Highlands of Queensland and in all cases over 90% control was achieved by day 14 with doses of 75g spores/ha or more except for a single plot where the tree cover had intercepted much of the spray. The spray deposit persisted for over 10 days and was still able to infect 50% of locusts after 4 days.

Dr W.E. Grant (Texas A & M University) has collaborated with Drs Scanlan, Milner and Hunter to

model the effects of dose, vegetation cover and temperature on the efficacy of spray for migratory locust and a paper of this modelling has been submitted to Ecological Modelling. The model confirms the importance of secondary pickup which often contributes over 20% of the infection. The model suggests that even a low dose of 25g spores/ha can give over 90% control under sparse vegetation conditions. It further confirms that improvements in the formulation to increase the persistence and secondary pickup is probably the most likely strategy for improving efficacy and thus reducing cost.

Australian Plague Locusts

Two field trials on Australian plague locust have been completed under contrasting weather conditions. The first in the Ardlethen area near Griffith, NSW, was undertaken in cool weather during October 1999. The second was undertaken under very hot conditions at Windorah in SW Queensland.

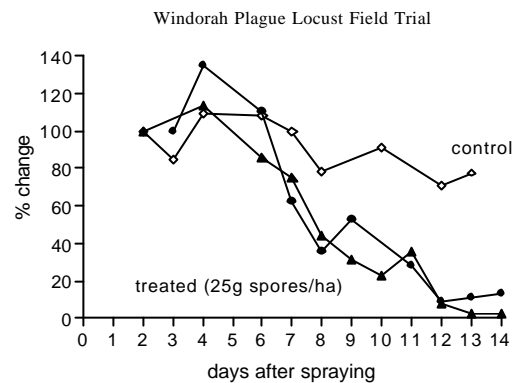
The Ardlethen trial compared low and moderate doses in two oil formulations, with and without UV protectants. A total of over 700 ha was treated in 8 blocks. The bands were followed and the populations assessed daily. At all doses the population declined sharply in the second week after treatment with control of over 95% being achieved by day 21. Two control plots did not show any consistent decline over this period. Furthermore untreated insects which invaded the treated plots or hatched within them during the first week also died, though this mortality was understandably delayed with full control being achieved by day 26. Some viable spray deposit was detected on the vegetation after 14 days, and the half-life was 4 to 5 days. The UV protectant (provided by Caltex) did not have any effect on the field efficacy and in the laboratory it did not give any detectable protection from UV.

This trial was remarkable for the spectacular control achieved with the low dose of 25g spores /ha. At this rate, *Metarhizium* would cost about the same as many chemical insecticides.

The second trial compared the low (25g spores/ha), medium (75g spores/ha), and high (125g spores/ha), doses in the same oil formulation at 1 and 0.5 litres/ha. In addition spores produced by SGB were compared with those from CSIRO. The maximum temperatures during the trial ranged from 36-42°C with very high level of UV radiation. A total of 8 blocks were treated and two blocks were also monitored as untreated controls. Despite the extreme weather conditions, even the low dose (25 g

spores/ha) plots showed a rapid decline at 7-10 days after spraying (Figure 2). Mortality was delayed at the high dose (125g spores/ha) and this was thought to be because there were problems in spraying this material which was very viscous. The CSIRO and SGB material gave similar high levels of control. Also there was no evidence to suggest that the volume of oil had an effect on final control. Thus a dose of 25g spores/ha in 500 ml oil was very effective. This has important implications for operational control as it means reduced time out for aircraft refilling. Vegetation samples indicated that while there was some persistence of the spray deposit for 10 days most of the spores had died by day 2, giving a half-life of perhaps 1 day. This may be as a result of the extreme weather conditions.

Figure 2:



LABORATORY STUDIES

One of the most critical aspects of producing an effective biopesticide based on spores of *Metarhizium* is to dry the spores, so that they remain viable, down to 5-10% moisture. If dried effectively the spores will persist much longer on the vegetation after spraying and also the material will store for a much longer time. CSIRO has undertaken tests of a range of drying techniques to find the most appropriate in terms of cost and efficacy. It has been found that air-drying on the rice for 7-10 days is both very effective and quite economical. Once dry the spores are easily removed by sieving. A new drying room has been installed at Black Mountain and the recent successful field trials have all used material dried in this way and this has resulted in improved field persistence and efficacy.

Another factor of concern was the ability of some locusts and grasshoppers to bask thereby raising their body temperatures to 40°C or more when the sun is shining. The FI-985 *Metarhizium* isolate will not grow at this temperature and if insects are maintained continuously at 40°C they will not die of the disease. In order to simulate these conditions in the laboratory, special cages have been constructed

with an incandescent lamp at one end which is linked to a timer. The insects can raise their body temperature by roosting close to the lamp while it is switched on. The results have shown that periods with the lamp "on" of 8 hrs or less have little effect on the disease. A 12 hr period or longer can increase the incubation time prior to death but this is dependent on the target species. Wingless grasshoppers were unable to bask and the lamp had no effect on mortality. For the other species tested (plague, spur-throated and migratory locusts and *Austroicetes*) there was a delay in mortality with the lamps on for 12 hr. Other tests have shown that the mycelium is not killed by treatment at 45°C or 50°C for 24 hrs suggesting that this basking is unlikely to result in the locusts curing themselves of the infection.

Tests on non-target aquatics by Dr Richard Lim at the University of Technology Sydney have shown that FI-985 at a dose 10 fold higher than a medium field dose does not affect mayflies or rainbow fish. However the viable spores did have an adverse effect on cladocerans. This is being further investigated and may be due to the very high dose used in these tests.

PUBLICATIONS AND CONFERENCE PAPERS

Driver, Felice, Milner, Richard J., and Trueman, John, W.H. (2000). A taxonomic revision of *Metarhizium* based on a phylogenetic analysis of rDNA sequence data. *Mycological Research* 104, 135-151.

Hunter, D.M., Milner, R.J., Scanlan, J.C. and Spurgin, P.A. (1999). Aerial treatment of the migratory locust, *Locusta migratoria* (L.) (Orthoptera: Acrididae) with *Metarhizium anisopliae* (Deuteromycotina: Hyphomycetes) in Australia. *Crop Protection* 18, 699-704.

Milner, R. J. (2000). Current status of *Metarhizium* as a mycoinsecticide in Australia. *Biocontrol News and Information* (in press, due June).

Milner, R.J., Baker, G.L. and Hunter, D.M. (2000). *Metarhizium* for control of wingless grasshopper (abstract). Paper given at the Australian Entomological Society 30th AGM and Scientific Conference, Canberra, Sept/Oct. 1999.

Scanlan, J.C., Grant, W.E., Hunter, D.M. and Milner, R.J. (submitted) Habitat and environmental factors influencing the control of migratory locusts (*Locusta migratoria*) with a biopesticide (*Metarhizium anisopliae*). *Ecological Modelling*

PLANS FOR 2000

1. The APLC will commence using Green Guard™ operationally and it is hoped to provide material for land-holder evaluation.
2. The registration package for NRA will be prepared.
3. Evaluation on non-targets will continue with data collection from field trial pitfall trap sampling and also further work on the apparent susceptibility of cladocerans to *Metarhizium*.
4. The possible use of Green Guard™ in combination with organic insecticides such as pyrethrum to assess the possible development of an organic spray for rapid kill to protect crops.
5. Evaluation of the use of dispersants and UV protectants to improve the handling characteristics and the persistence of the spray formulation.
6. Further work on sub-lethal effects on ovarian development, fecundity and egg hatch.
7. Optimisation of new bag method for production.