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Final report - SRDC project BSS201 - Determining the biology of rhopaea canegrub in the New South Wales sugar industry

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FINAL REPORT – SRDC PROJECT BSS201
DETERMINING THE BIOLOGY OF RHOPAEA
CANEGRUB IN THE NSW SUGAR INDUSTRY
by
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SD02011

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APPENDIX 1  Generalised phenology of *R. magnicornis* in the Tweed Valley, NSW

APPENDIX 2  Suggested protocols for initial investigations into triggers of flight by female *R. magnicornis* beetles
SUMMARY

Rhopaea canegrub (*Rhopaea magnicornis* Blackburn) is the major insect pest of the New South Wales sugar industry with about 25% of Condong farms affected. Rhopaea has also been reported as a pest in the Broadwater and Nambour canegrowing areas. At the time this project commenced little was known about the ecology and population dynamics of the pest and no commercially viable control measures existed.

Contrary to previous beliefs, some female beetles were observed to fly prior to mating. However, female beetles were also observed that emerged and did not fly, but mated on the soil surface before burrowing back into the soil where they laid their eggs. It is not yet known which behaviour is the most common. The average egg batch was found to be 21.3 eggs.

The distribution of the life stages of rhopaea canegrubs was determined using both naturally occurring populations at three sites and an introduced population at a fourth site. First and second instars are shallow feeders; they were found at an average depth of about 10 cm. The third-instar rhopaea larvae not only move deeper into the soil (average depth about 15 cm) but also become more focused around the sugarcane stool and row centre. Finally, as pupation approaches, rhopaea grubs again move up in the soil profile, where they were found to pupate at an average depth of only 6.5 cm below the soil surface. Rhopaea is a comparatively shallow-living grub in contrast to some other canegrub species.

Green cane trash blanketing was tested on six farms and “stool rolling” on five farms. Neither practice was shown to be effective but this may be due to the relatively low pest populations that prevailed during the three years of field work.

Fallowing as a control measure was tested with both naturally occurring populations and in a replicated trial where plots were seeded with 20 second instar larvae. A grass fallow was found to increase pest numbers in subsequent crops compared to replanting. In the replicated experiment, all fallow treatments reduced grub numbers by similar amounts four months after the larvae were introduced.

Field counts made after land preparation for replanting showed that cultivation can reduce pest numbers by close to 100%. However, such large decreases may also disrupt the disease cycle which is essential in suppressing pest numbers.

Disease studies showed that rhopaea is infected by a different species of *Metarhizium* fungus (*M. flavoviride*) than other canegrub species which are infected by *M. anisopliae*. The fungus affects all stages (egg-adult) of the pest so that the “window of control” is very wide. Some field observations showed over 50% of a female’s eggs could be killed by metarhizium infections. *Beauveria bassiana* was another prominent fungal disease identified in the population. This fungus has only been rarely observed in other more northern canegrub species. Both fungi have potential as biological control agents.

Other diseases found include milky disease (*Paenibacillus popilliae*), three minor fungal diseases and one incidence of a rare microsporidian disease (*Nosema sp.*).
1.0 BACKGROUND

Rhopaea canegrub (*Rhopaea magnicornis* Blackburn) is the major insect pest of the New South Wales sugar industry (Hayes, 1998), with about 25% of Condong farms affected. Rhopaea has also been reported as a pest in the Broadwater and Nambour canegrowing areas. The cost of direct losses, shortened crop cycles and additional replanting costs have been estimated as exceeding $600,000 per year.

Prior to this study it became clear that there was no successful control measure available to northern NSW canegrowers. Although SuSCon® Blue is registered for control of rhopaea canegrub, application at planting is only partly effective and the insecticide is used by few farmers. The only control measure that is commonly practised is driving a tractor along the rows after harvest to press stools back into the soil and squash any grubs present in the topsoil. This practice is known locally as ‘stool rolling’.

Unlike other canegrub pests, very little work has been done on rhopaea. Although similar studies have been conducted with canegrub pests in north Queensland, little of this work will have relevance to south Queensland and NSW because of differences in climate, biology and farming practices. There are only two published scientific papers devoted to *R. magnicornis* (Chadwick, 1970; Hayes, 1998) and only one of these (Hayes, 1998) focuses on rhopaea as a sugarcane pest.

A research project was required that would document the life cycle and biology of the insect. Other items of interest included identifying those conditions under which rhopaea exhibits a two-year life cycle instead of the normal one-year cycle. This project sought to identify the distribution of grubs through the soil over its life cycle. This will greatly assist future pest control work by identifying times when the pest is most vulnerable to control measures.

We aimed to identify important parasites and diseases of rhopaea and to identify farm management practices that may favour parasites and diseases (eg trash blanketing and fallowing).

The project sought to provide industry with basic research into a major pest in the sugarcane growing areas where it is most common. From this work, integrated pest management (IPM) control strategies will be developed. The result will be higher productivity, longer crop cycles and, because affected farmers will not have to replant as often, reduced planting costs.

2.0 OBJECTIVES

- Determine the biology of rhopaea canegrubs in the Nambour, Condong and Broadwater areas, including the distribution of rhopaea’s life stages in the soil.
- Identify the key parasites, pathogens and predators that control the population level of rhopaea canegrubs.
- In consultation with growers, identify those farming practices that favour the pest’s natural control mechanisms.
- Provide basic research that will form the basis for developing an IPM strategy
- Extend immediately applicable information to farmers.
The above objectives have been mostly met. The distribution of rhopaea canegrub in the soil is now well understood, and a range of pathogens that can affect the population dynamics of this pest has been identified.

However, during the period of this study, damage to sugarcane by rhopaea canegrub was scarce. This absence of large field populations of the pest meant that no clear treatment effects were evident in field trials conducted to determine the impact of two cultural practices (green cane trash blanketing, stool rolling). More research is needed under conditions of higher pest pressure to determine the precise effects of these practices.

Fallowing appears to be capable of affecting the population dynamics of this canegrub pest, but more research is required to fully understand the processes involved and the effects.

Based on the research work completed in this project, it is now possible to commence development of an IPM strategy for rhopaea canegrub in the northern NSW sugar industry. Areas where further research is required have also been identified.

Farmers have been kept aware of the research project’s progress through talks at annual field days and by informal contacts. The primary investigator, Mr Austin McLennan, presented a seminar in June 2002 on the overall findings of this SRDC-funded project. Production of a BSES fact sheet on the pest is also planned.

3.0 METHODS

3.1 Measuring the distribution of rhopaea canegrubs in the soil

I recorded the distribution of the life stages of rhopaea canegrub by sampling from naturally occurring field populations at three sites on several occasions over 12 months. In a second study, I seeded specific locations in a single field with mated female beetles. These locations were sampled about every two months for a period of 10 months and the distribution of life stages was measured.

Sites with naturally occurring rhopaea populations

Three sites were initially chosen for studying the distribution of rhopaea in the soil. Details of each of these sites are given in Table 1.

<table>
<thead>
<tr>
<th>Site label</th>
<th>Farm name</th>
<th>District</th>
<th>Year planted</th>
<th>Ratoon (’99)</th>
<th>Cultivar</th>
<th>No. of times sampled</th>
</tr>
</thead>
<tbody>
<tr>
<td>Site1 (RH1)</td>
<td>Rockview Farm</td>
<td>Tumbulgum</td>
<td>1996</td>
<td>3</td>
<td>CP44-101</td>
<td>4</td>
</tr>
<tr>
<td>Site2 (WS1)</td>
<td>W &amp; N Stainlay</td>
<td>Murwillumbah</td>
<td>1996</td>
<td>3</td>
<td>Q124</td>
<td>4</td>
</tr>
<tr>
<td>Site3 (TT1)</td>
<td>TJ &amp; MJ Twohill</td>
<td>Kynumooneen</td>
<td>1996</td>
<td>2</td>
<td>CP44-101</td>
<td>8</td>
</tr>
</tbody>
</table>
All sites were sampled fewer times than planned because there were low populations at each. Sites 1 and 2 were mainly used to obtain distribution data on rhopaea’s early stages. I persisted with site 3 for longer (because of its larger population) to obtain distribution data on the later stages of rhopaea canegrub.

At each of these sites, the sample consisted of a trench focused around a sugarcane stool. Each trench included a 30-40 cm section of cane row that was excavated about 70-80 cm into the inter-row space either side of the sugarcane stool (sugarcane rows area spaced at 1.5 m intervals). Each trench was excavated to the depth of the clay subsoil underlying the black/‘peat’ topsoil. This was usually between 30-40 cm below the soil surface.

I recorded three measurements for each rhopaea individual found. These were its depth below the soil surface at that point, its lateral position either side of the centre of the sugarcane row/stool, and on which side of the row I found it.

**Site with ‘artificially boosted’ rhopaea population**

Because natural populations of rhopaea cane grub were low in 1999 and 2000, I sought to maximise the number of rhopaea individuals encountered per excavation by increasing the naturally occurring density of rhopaea larvae at specific known locations in a single field. I did this by placing field-captured mated females at known locations, permitting them to burrow into the soil at these locations and subsequently oviposit. By artificially boosting oviposition at known sites, I could later excavate these locations with confidence of obtaining more individual observations on the distribution of rhopaea individuals than if I had relied on natural oviposition alone.

In the data obtained from this site, each excavation or ‘sample’ refers to a trench that focused around the specific location at which a mated female beetle had been placed during the 1999 flight and mating season. These locations were marked by fibreglass rods, highlighted with flagging tape and pushed into the soil.

Each trench included a section of cane row that was excavated 50-80 cm into the inter-row space either side of the sugarcane. For each trench, I excavated to the depth of the clay subsoil underlying the black/‘peat’ topsoil (about 30-40 cm). The distance that the trench extended along the row either side of the female beetle release point varied as the season progressed. As larvae emerged and moved along the length of the row, I also extended the length of these trenches in an attempt to ‘capture’ all the progeny of the released female as they moved away from the initial oviposition site. By September, when pupation had commenced and larvae had the most opportunity to disperse, some excavations involved the removal of up to nearly 5 m of cane row. As a general rule, I kept excavating along the row away from the release point until I excavated a 20-30 cm section of cane row without encountering a single rhopaea individual. I assumed at this point that I had encountered all the possible progeny of the female I had initially released, though this assumption remains untested.

I recorded the same three measurements for each rhopaea individual found, depth, lateral distribution, and which side of the row it was found it on (east or west). I also recorded the distance along the length of the cane row (north or south) that an individual was found from the oviposition location.
3.2 Estimates of female fecundity

Mating female beetles were captured in the field after dusk in November-December 2000. Females were weighed the following morning and some were placed into 750 mL round plastic containers filled with moistened peat moss (Yates®). These females were kept in the laboratory and observed daily to determine the start of oviposition and their realised fecundity under controlled laboratory conditions.

Other female beetles were allocated to a field experiment where individuals were placed in specific marked locations in a sugarcane field where these females laid their egg batches. Extra details on this experiment have already been outlined in section 3.1 and in the technical summary part of this final report. We excavated 10 females and their eggs in December and again in January, giving some indication of realised fecundity under field conditions.

The relationship between a female’s mass at mating and her realised fecundity were investigated using linear regression.

3.3 Green cane trash blanket trials 1998-1999

Research in the north Queensland sugarcane industry has demonstrated lower numbers of greyback canegrub under a green cane trash blanket (GCTB) (Robertson and Walker, 1996). To test whether R. magnicornis populations might also be suppressed under a GCTB, I required harvested sites that had rows with a GCTB adjacent to rows without a GCTB (ie no trash or minimum till). Several growers in the Condong mill area were approached who had sites matching these criteria.

In the second half of the 1998 harvesting season, we negotiated seven sites to be set up on six farms. Each site consisted of a section of cane that had been cut green with the resultant trash blanket retained, adjacent to another section of harvested cane where no GCTB was retained (ie the cane was either burnt before harvesting and/or the trash was burnt post-harvest).

Table 2 shows the background details for each of the seven experimental sites at the end of the 1998 harvesting season.

<table>
<thead>
<tr>
<th>Farmer</th>
<th>District</th>
<th>Cultivar</th>
<th>Ratoon #</th>
</tr>
</thead>
<tbody>
<tr>
<td>Partridge G.</td>
<td>Evron</td>
<td>Louisiana</td>
<td>3rd ratoon</td>
</tr>
<tr>
<td>O’Keefe P.</td>
<td>Tumbulgum</td>
<td>Q124</td>
<td>2nd ratoon</td>
</tr>
<tr>
<td>Stainlay W.</td>
<td>Murwillumbah</td>
<td>Encore</td>
<td>2nd ratoon</td>
</tr>
<tr>
<td>Stainlay W.</td>
<td>Murwillumbah</td>
<td>Encore</td>
<td>2nd ratoon</td>
</tr>
<tr>
<td>Quirk A.</td>
<td>Tumbulgum</td>
<td>Q124</td>
<td>2nd ratoon</td>
</tr>
<tr>
<td>Quirk R.</td>
<td>Tumbulgum</td>
<td>Q124</td>
<td>1st ratoon</td>
</tr>
<tr>
<td>Anderson R.</td>
<td>Tygalgah</td>
<td>Louisiana</td>
<td>4th ratoon</td>
</tr>
</tbody>
</table>
All samples for estimating larval abundance consisted of a section of soil centred around a sugarcane stool. Each sample was about 30 cm long by 40 cm wide (ie 20 cm either side of the row centre) by 40 cm deep. Between 10-20 samples were taken per treatment strip (ie burnt trash or GCTB).

### 3.4 Stool rolling trials 1998-1999

Five sites were established to assess the effect of ‘stool rolling’ on rhopaea canegrub larvae in the second half of the 1998 harvesting season. Stool rolling is a practice that involves driving a tractor’s wheels directly over the stools of the sugarcane plant after harvest. The practice is not a routine operation, but is commonly used by farmers on the light peat soils when grubs or some other factor (eg lodging) causes the stools to be dislodged during harvest. Rolling is used to press these stools back into the soil, thereby promoting crop recovery, but farmers have suggested that rolling may kill canegrubs by squashing. Growers based this assumption on their observation that if they rolled a ‘grubby’ paddock one year, frequently no grub problem was evident the following year.

Each of the study sites consisted of a recently harvested block of sugarcane where a portion of the field was rolled and an adjacent portion (or portions) was left unrolled. Rhopaea numbers were generally assessed soon after the rolling event to detect any immediate mortality.

All individual samples for estimating larval abundance consisted of a section of soil centred around a sugarcane stool. Each sample was about 30 cm long by 40 cm wide (ie 20 cm either side of the row centre) by 40 cm deep. Between 10-20 samples were taken per treatment strip (ie rolled or unrolled).

### 3.5 Fallow trials 1999 and 2001

In rhopaea canegrub, where previous observations suggested at least the majority of females oviposit close to their emergence site (Hayes, 1998), it was expected that the possibility of reinvasion by females from neighbouring infested fields would be low. Provided that long-fallowing is able to destroy the great majority of grubs in a single field, fallowing may keep grub numbers low for several years with the (assumed) very low rate of immigration. Fallowing, therefore, was considered to have a potentially great impact by reducing populations of rhopaea canegrub prior to replanting with sugarcane. This assumed that the population of rhopaea larvae remaining (if any) after a fallow period is lower than would remain after the more conventional ploughout/replant operation.

Two experimental approaches were used to investigate the effect of fallowing on rhopaea’s population dynamics.

**Experiment 1**

The aim of the first study was to assess whether the envisaged effect of fallowing on rhopaea abundance could be detected two to three years after planting.

Four sites in the Tweed Valley were selected for sampling in 1999. Sites were selected on the basis that they contained at least two adjacent blocks of sugarcane planted in the same year, one in a ploughout/replant operation, the other into ground that had been fallowed.
All sites were located on the ‘peat’ soil type that is most susceptible to attack by rhopaea canegrub.

At all sites, sampling locations were selected by a computer-generated set of random co-ordinates consisting of a row number and the number of paces along that row from the headland, after allowing for a 20 pace buffer at the end of each row. A single sample consisted of a section of soil around a sugarcane stool, 40 cm deep by 30 cm long (along row length) by 40 cm wide.

Site 1 consisted of two adjacent blocks of cane, both planted to cultivar Q124 in 1995; one fallow-plant, the other ploughout/replant. This site was randomly sampled once as second-ratoon cane, with 15 samples taken from each block. Samples were taken from an area in each block 12 rows wide by 100 paces long (Alan Brown1).

Site 2 consisted of two adjacent blocks of cane, both planted to Q124 in 1998, one fallow-plant the other ploughout/replant. This site was randomly sampled as plant cane, with 10 samples taken from each block. Samples were taken from an area 16 rows wide by 100 paces long (R Quirk1).

Site 3 consisted of two adjacent blocks of cane both planted to CP44-101 in 1997; one fallow plant, the other ploughout/replant. The site was randomly sampled as second-ratoon cane, with 12 samples taken from each block. Samples were taken from an area 13 rows wide by 50 paces long (R Quirk2).

Site 4 was a replicated trial, comprising six blocks of sugarcane in total, three of which were ploughed out in 1995, fallowed and planted in 1996. The farmer reported that some grass growth had occurred during the fallow period. The remaining three blocks were ploughed out in 1996 and replanted the same year. All six blocks were planted to cultivar CP44-101 and stood over as plant cane. All six blocks were sampled twice in 1999 (April/May and November) when the crop was in its second ratoon. Samples were taken from an area 18 rows wide by 200 paces long.

Experiment 2

A major field study was undertaken in the first half of 2001 to investigate the effects of different types of fallow management on rhopaea persistence or survival in the soil.

In the absence of natural grub infestations, small plots (1 m by 1.5 m) under different fallow regimes were seeded with 20 field-collected second instar larvae. The different fallow management practices compared in this trial were bare, grass, legume and minimum-till fallows. Each mini-plot was bounded by a trench into which we laid builders’ black plastic. This plastic acted as a barrier to prevent the introduced larvae from escaping.

Healthy larvae were carefully introduced to the mini-plots in March. A minimum of four plots per treatment type were excavated in each of April, June and July. Excavated larvae were counted and weighed. The trial concluded in July because the farmer needed to prepare the field for planting.
3.6 Spatial distribution of rhopaea canegrubs in two fields 2000-01

Two blocks of sugarcane in the Condong area were selected in September 2000 for a detailed investigation into the fine-scale distribution of rhopaea canegrub. I selected both sites on the basis that they had a history of canegrub damage and that they would be available for easy sampling in both September 2000 and September 2001. Easy sampling required a harvest date in both years prior to sampling.

Details of two fields chosen for studies on the within-block distribution of rhopaea canegrub

The two sites were on different farms; one on Knightleigh Plantations, Dulguigan (Martin), and the other on Questfield Pty Ltd, Eviron (Grippo).

Each site possessed an element of environmental heterogeneity that I realised may have an effect on the underlying abundance and distribution of rhopaea canegrub larvae within that site. This heterogeneous factor was different for each site. In the case of Martin, there was a slight slope from one end of the field to the other. During the extremely wet weather and local flooding of 1999, the soil towards the lower end of the block had been inundated more frequently than the soil at the higher end. For the second site, Grippo, the main source of heterogeneity consisted of a sudden change in soil texture within the block. While the majority of the block consisted of ‘peat’ soil, a clay ridge at the other end of the block meant a dramatic difference in soil texture (and therefore grub habitat) between opposite ends of this particular canefield. It is known that abundance of some canegrub species can vary with changes in soil texture (Cherry and Allsopp, 1991).

Sampling Details

All individual samples for estimating larval abundance consisted of a section of soil based around a sugarcane stool. Each sample was 30 cm by 40 cm by 40 cm deep. The number of rhopaea individuals were counted and divided into the following categories: second-instar larvae; third-instar larvae (one year); third-instar larvae (possibly two years old); pre-pupae and pupae. Pupae were sexed whenever possible.

At both sites, in both 2000 and 2001, I sampled on regular grid patterns to give a good indication of rhopaea’s distribution within each block. The precise details are as follows.

Martin:

In 2000, I sampled using three different grid sizes. In 2001, I sampled using the largest grid only. The largest grid involved sampling along six rows, each 20 m apart, with sampling points 20 m apart from each other down the length of the row (taken from 0-200 m).

The second grid size used in 2000 involved samples taken at 10 m intervals (ie rows were 10 m apart, sample points along rows were 10 m apart), and these were taken in part of the area encompassed by the large grid.

Two 10 m by 10 m sections within this medium grid were chosen for intensive systematic sampling on a very fine grid. One of these 10 m x 10 m squares was chosen to represent an area of possible high rhopaea density (based on the abundance data for the four corner
samples) while the other was chosen to represent an area with a probable low density of rhopaea. These 10 m by 10 m areas were sampled on a very fine grid pattern. Each grid consisted of seven rows of sugarcane (approx. 1.5 m apart), with seven samples taken in each adjacent section of row for a distance of only 10 m (ie each sample was approximately 1.4 m apart along the length of the row).

From 20-25 September 2000, we took samples at all the points in the field specified by the systematic sampling method outlined above. The field had only been recently harvested. The original plan when resampling this field in 2001 was to sample again in mid-September after harvest for sampling ease. However, this was not possible until after mid-October when the block was eventually harvested. In 2001, I only sampled using the largest 20 m grid spacings, in the same rows as the previous year.

**Grippo:**

As distinct from the first site (Martin), Grippo was only sampled in both 2000 and 2001 using the largest 20 m grid spacings. This block was both shorter and narrower than the other site, so less individual points were sampled to build up a map of rhopaea’s distribution within this commercial sugarcane block.

Four rows, each 20 m apart were sampled at regular 20 m intervals for a distance of 200 m in 2000 and for 240 m along the row the following year. I sampled this field on 2 October 2000 and on 3-4 July 2001. I originally hoped to sample this field 12 months after the original sampling in October 2000, but had to bring the second sampling period forward to July 2001 because the farmer wanted to ploughout this block for replanting with a new crop in August/September.

### 3.7 Pathogens of *R. magnicornis*

During the course of the other experimental work in 1999 and 2000, over 1500 field-collected larvae or cadavers were sent to Dr Richard Milner (CSIRO Entomology) for pathogen screening.

### 4.0 RESULTS

#### 4.1 Distribution of rhopaea canegrubs in the soil

The distribution of rhopaea canegrubs in the soil has now been established. This information will assist in deciding on the correct placement of insecticides next time there is an outbreak of rhopaea canegrub.

My studies found that young first-instar larvae migrate towards the soil surface after hatching. Second instars are also shallow feeders; they were found at an average depth of about 10 cm. However, the damaging third-instar rhopaea larvae not only move deeper into the soil (average depth about 14 cm) but also become more focused around the sugarcane stool and row centre. Finally, as pupation approaches, rhopaea grubs again
move up in the soil profile, where they were found to pupate at an average depth of only 6.5 cm below the soil surface. This research shows that rhopaea is a comparatively shallow-living grub in contrast to other canegrub species (Logan, 1999).

**Site with artificially boosted populations**

Table 3 presents the average depth and distribution data from this trial.

**Table 3: Changing depth and distribution of rhopaea canegrub throughout 2000.**

<table>
<thead>
<tr>
<th>Stage:</th>
<th>Nov/Dec</th>
<th>March</th>
<th>July/August</th>
<th>October</th>
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<tr>
<td></td>
<td>Eggs</td>
<td>1st instar</td>
<td>2nd instar</td>
<td>3rd instar</td>
</tr>
<tr>
<td>DEPTH (cm)</td>
<td>mean</td>
<td>14.5</td>
<td>12.4</td>
<td>8.4</td>
</tr>
<tr>
<td></td>
<td>std dev</td>
<td>3.21</td>
<td>5.39</td>
<td>3.97</td>
</tr>
<tr>
<td></td>
<td>n</td>
<td>129</td>
<td>91</td>
<td>112</td>
</tr>
<tr>
<td>DISTANCE from ROW CENTRE (cm)</td>
<td>mean</td>
<td>8.7</td>
<td>12.4</td>
<td>12.4</td>
</tr>
<tr>
<td></td>
<td>std dev</td>
<td>5.06</td>
<td>6.81</td>
<td>10.77</td>
</tr>
<tr>
<td></td>
<td>n</td>
<td>125</td>
<td>93</td>
<td>120</td>
</tr>
</tbody>
</table>

Only rhopaea individuals exhibiting the more typical one-year life cycle were included in the above analysis.

The overall picture presented in Table 3 shows that eggs are laid at an average depth of about 14 cm below the soil surface. The distance they are found from the centre of the sugarcane row is obviously influenced by the behaviour of the ovipositing female and no clear trend was observed. Upon emerging from the eggs, it appears that young first instar larvae move closer towards the soil surface. Since data on newly emerged larvae (still in the original location of their eggs) were used in compiling Table 3, their movement towards the soil surface is underestimated by the mean depth shown.

Second instar larvae are also very shallow feeders. The change to the very active and most damaging third larval stage shows a marked change in larval distribution. Not only do the larvae exhibit a general movement deeper into the soil, they also become much more focused in their lateral distribution around the centre of the sugarcane row. There is another sharp contrast in rhopaea’s distribution when pupation begins. Larvae generally move upwards to pupate closer to the soil surface, as well as dispersing laterally.

The following figures (Figures 1-6) present the same data on which the above means are based, except in the form of a frequency distribution.
Figure 1: Depth of eggs and first instar larvae from D Cowderoy site.

Figure 2: Lateral distribution of eggs and first instar larvae (D Cowderoy).
Figure 3: Depth of 2\textsuperscript{nd} instar larvae, D Cowderoy trial, 2000.

Figure 4: Lateral distribution of 2\textsuperscript{nd} instar larvae, D Cowderoy trial, 2000.
Figure 5: Depth of 3rd instar larvae, D Cowderoy trial, 2000.

Figure 6: Lateral distribution of 3rd instar larvae, D Cowderoy trial, 2000.

Figure 7 shows the frequency at which 3rd instar larvae were detected at certain distances along the row from the site where a female beetle had been placed to oviposit. These data show that most rhopaea larvae do not apparently move more than 1-1.5 m along the cane row from the site where they hatch.
Third instar larvae were generally found to migrate towards the soil surface and towards the inter-row space to pupate. Average pupal depth was very shallow (6.5 cm; n=65) and they ranged from 2.5 to 13 cm below the soil surface. The average distance that pupae travelled into the inter-row was 22.2 cm (n=71) but the range was between 5 and 50 cm (only three pupae were found greater than 40 cm away from the row centre). Out of 97 pupae examined, the sex ratio was male biased (1 female pupa to 1.2 male pupae).

**Figure 7:** Movement of 3rd instars along the row away from oviposition site, D Cowderoy trial, 2000.

### 4.2 Estimates of female fecundity

Figure 8 presents data that shows a positive relationship between the mass of the female beetle at mating and realised fecundity under field conditions. Females were found to lay between 12-32 eggs, with an average egg batch made up of 21.3 eggs (n=38).

**Figure 8:** Fecundity of female rhopaea beetles in the field and the laboratory.
4.3 Green cane trash blanket trials 1998-1999

There was no evidence to confirm that green cane trash blanketing suppressed numbers of rhopaea canegrub in the 1998-1999 growing season. However, this lack of a conclusive result may have been primarily due to a low rhopaea population at all seven trial sites. Wet and waterlogged conditions for most of that year may also have negated the potential effects of a GCTB on rhopaea mortality.

Tables 4 and 5 show typical results from two of the seven trash blanket trials conducted where grub numbers were low and no clear effect was evident.

Table 4: Mean number of rhopaea/sample under GCTB or no-trash conditions, 1999 (farm: Stainlay).

<table>
<thead>
<tr>
<th></th>
<th>June 1999</th>
<th>December 1999</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Trash</td>
<td>2.4</td>
<td>0.6</td>
</tr>
<tr>
<td>s.e.</td>
<td>(0.73)</td>
<td>(0.80)</td>
</tr>
<tr>
<td>Trash</td>
<td>2.1</td>
<td>0.3</td>
</tr>
<tr>
<td>s.e.</td>
<td>(0.19)</td>
<td>(0.12)</td>
</tr>
</tbody>
</table>

Table 5: Mean number of rhopaea/sample under GCTB or no-trash conditions, 1999 (farm: Partridge).

<table>
<thead>
<tr>
<th></th>
<th>May 1999</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Trash</td>
<td>0.67</td>
</tr>
<tr>
<td>s.e.</td>
<td>(0.27)</td>
</tr>
<tr>
<td>Trash</td>
<td>1.27</td>
</tr>
<tr>
<td>s.e.</td>
<td>(0.71)</td>
</tr>
</tbody>
</table>

One site (A Quirk) did suggest a very mild suppressing effect of a GCTB on rhopaea canegrub but the number of grubs in the burnt treatment were too low (less than 0.25 grub/stool) to draw any firm conclusions.

One of the seven sites (O’Keefe, Tumbulgum) was abandoned when preliminary digs in December showed the population of rhopaea larvae in this cane field was already extremely low.

4.4 Stool rolling trials 1998-1999

None of the trials showed conclusively that stool rolling kills a significant proportion of rhopaea canegrubs. There were no consistent significant differences between larval density in rolled and unrolled sections of cane crops. Tables 6 and 7 represent typical results.
Table 6: Mean number of rhopaea/sample under rolled and unrolled cane (farm: R Bartlett).

<table>
<thead>
<tr>
<th></th>
<th>Rolled</th>
<th>Unrolled</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean /sample</td>
<td>3.07</td>
<td>1.91</td>
</tr>
<tr>
<td>s.e.</td>
<td>0.91</td>
<td>0.6</td>
</tr>
<tr>
<td>n</td>
<td>12</td>
<td>12</td>
</tr>
</tbody>
</table>

Table 7: Mean number of rhopaea/sample under rolled and unrolled cane (farm: R Hawken).

<table>
<thead>
<tr>
<th></th>
<th>Rolled</th>
<th>Unrolled</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean /sample</td>
<td>1.25</td>
<td>1.1</td>
</tr>
<tr>
<td>s.e.</td>
<td>0.31</td>
<td>0.35</td>
</tr>
<tr>
<td>n</td>
<td>10</td>
<td>10</td>
</tr>
</tbody>
</table>

4.5 Fallow trials 1999 and 2001

4.5.1 Effect of fallowing observations, 1999

Sites 1-3

*Rhopaea magnicornis* numbers were very low at these three sites in 1999, with no difference evident between grub populations in the fallow-planted or ploughout/replant blocks. As such, no significant differences in rhopaea abundance between the ploughout/replant and fallow/plant could be detected. Table 8 shows the results from one of these sites.

Table 8: Mean number of rhopaea/sample under fallow planted cane versus ploughout/replant cane (farm: A Brown).

<table>
<thead>
<tr>
<th></th>
<th>Rolled</th>
<th>Unrolled</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean /sample</td>
<td>0.33</td>
<td>0.47</td>
</tr>
<tr>
<td>s.e.</td>
<td>0.62</td>
<td>0.74</td>
</tr>
<tr>
<td>n</td>
<td>15</td>
<td>15</td>
</tr>
</tbody>
</table>

Site 4

Despite low populations at the other three ‘fallow’ sites, rhopaea was sufficiently abundant to detect statistically significant differences in grub numbers between the two treatments. Results for each of the blocks are shown in Figure 10 and summarised in Table 9.
Figure 9: Grubs/sample in April 1999, three years after planting the crop into either a fallowed block with grass (Fallow 1,2 and 3) or into a ploughout/replant block (Replant 1,2 and 3). Three replicates/treatment.

Table 9: Comparing average numbers of larvae/sample three years after fallow planting or ploughout/replant. Treatment averages are pooled data from all three replicates (n=45 / treatment).

<table>
<thead>
<tr>
<th>Sampling date</th>
<th>Grass fallow in 1995-96</th>
<th>Ploughout/Replant in 1996</th>
<th>% 2 Year grubs</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apr-99</td>
<td>6.03</td>
<td>2.2</td>
<td>35%</td>
</tr>
<tr>
<td>Nov-99</td>
<td>3.67</td>
<td>0.47</td>
<td>38%</td>
</tr>
</tbody>
</table>

As is evident from Table 9 and Figure 9, there were significantly more rhopaea larvae in the blocks that had been fallow-planted than in the ploughout/replant blocks. This was the opposite of what had been expected. The difference evident in April’s sampling also persisted through to sampling again in November.
Persistence of rhopaea under different fallow regimes, 2001

Figure 10: The mean number of larvae recovered from mini-plots subject to different fallow regimes.

All plots had twenty 2\textsuperscript{nd} instar larvae added in March 2001, except for the control plots which remained unploughed and where the cane regrowth was killed with a glyphosate application at the same time as the ‘spray-out’ fallow treatment.

Rhopaea larvae initially persisted the best under the minimum-till fallow situation where the cane stool was killed with herbicide but not removed by cultivation. However, this effect had disappeared by July when persistence under the different fallow regimes was equivalent.

All rhopaea larvae under all the fallow conditions were significantly lighter and therefore less well nourished than grubs collected and weighed from the same field that the experimental larvae were originally collected from.

Some grubs were able to survive for several months even under the bare fallow treatments, apparently getting some nutrition from decaying crop and stalk residues. Despite being less well nourished, some larvae removed from the various fallow plots were even able to complete their development to adult beetles. Neither the vigour nor fecundity of these survivors was assessed but larval weights suggest that ‘starved’ beetles would be less fecund.

Effect of cultivation on rhopaea survival

Data from the cultivated versus uncultivated fallow plots in this experiment also provided data on the effect of cultivation by rotary hoe on canegrub survival.
While excavating the various treatment plots, large third instar larvae were sometimes encountered that obviously did not belong to the cohort of 20 second instar larvae introduced into each plot. These larger and often yellowish larvae were ‘two-year’ larvae in the second year of their extended life cycle.

These two-year larvae were only ever found in the ‘unploughed’ fallow treatments (ie spray-out and control plots). None were ever found in plots belonging to any of the other three fallow treatments (ie bare, legume and grass fallow). Figure 11 shows the average number of two-year larvae for both the spray-out and control plots located at each monitoring occasion.

![Persistence of ‘two-year lifecycle’ larvae in ‘unploughed’ fallow treatments](image)

**Figure 11: Average number of two-year *rhopaea* larvae in each of the unploughed fallow treatments.**

At least one two-year larva was found in 43% of the 26 spray-out or control plots excavated during this experiment. In contrast, none of the 55 plots that had been cultivated prior to setting up this experiment yielded a single two-year larva. This effect is statistically significant.

Since two-year larvae have the same distribution in the soil as larvae undergoing a typical one-year life cycle, these results clearly show that the standard ploughout practices used to prepare a canefield for replanting or falling can virtually eliminate the *rhopaea* canegrub population present in a field.

### 4.6 Spatial distribution of *rhopaea* canegrubs in two fields 2000-01

The final form of the data from each sampling exercise was the abundance data from an individual sample linked with the spatial coordinates (ie row number versus distance along row) of that sample. The fact that all the sample points were taken according to a grid pattern enabled me to produce a map of *rhopaea* canegrub abundance and distribution within the sections of the fields I sampled.
At Site 1 (Martin), I found a significant correlation between rhopaea abundance and the distance of the sample point from the lowest headland. The highest abundance of rhopaea larvae was found in the highest parts of the paddock (Figure 12). In the previous year, when extremely wet weather had prevailed, the lowest parts of the field were the most subject to inundation. In 2000, this trend was evident across the area covered by both the 20 m and 10 m sampling grids.

Sampling at the finest scale (1.7 m grid) showed that samples near each other in the same row are more likely to have similar numbers of larvae than samples taken nearby stools in the adjacent row.

At Site 2 (Grippo), sampling revealed abrupt changes in rhopaea density that correlated exactly with an abrupt change in soil texture. In 2000, the highest populations were found in the lighter-textured peat soil, whereas the population fell to almost 0 larvae/sample in the heavy clay ridge at the other end of the paddock (Figure 13). Interestingly, the situation was reversed in the following year, with an almost negligible larval population in the light soil (where the ratoon crop was very poor as a result of grub damage the previous season), but with significantly higher numbers around the stools on the clay ridge (undamaged).

4.7 Pathogens of *R. magnicornis*

The disease data from the *Rhophaea magnicornis* canegrubs collected at Condong and reared by Dr Richard Milner during 1999 and 2000 have been collated and summarised.

<table>
<thead>
<tr>
<th>Year collected</th>
<th>Total examined (n)</th>
<th>% of rhopaea killed by various mortality agents</th>
<th>% of rhopaea survived to pupa</th>
</tr>
</thead>
<tbody>
<tr>
<td>1999</td>
<td>(721)</td>
<td>Metarhizium 15.67</td>
<td>Beauveria 11.10</td>
</tr>
<tr>
<td>2000</td>
<td>(569)</td>
<td>Metarhizium 14.76</td>
<td>Beauveria 10.72</td>
</tr>
</tbody>
</table>

This work showed that rhopaea is infected by a different species of *Metarhizium* fungus (*M. flavoviride*) from other canegrub species, which are infected by *M. anisopliae* (R. Milner, pers. comm.). The fungus affects all stages (egg-adult) of the pest so that the “window of control” is very wide. Some field observations showed that over 50% of a female’s eggs could be killed by metarhizium infections. *Beauveria bassiana* was another prominent fungal disease identified in the population. This fungus has only been rarely observed in other more northern canegrub species but was only slightly less prevalent in the pest population than *M. flavoviride*. Both fungi have potential as biological control agents.

Other diseases found in the population include milky disease, *Paenibacillus popilliae*, three minor fungal diseases and a minor disease caused by Microsporidia (*Nosema* sp.). Unlike similar studies in north Queensland (Robertson *et al.*, 1998) protozoan diseases don’t appear to be an important influence on the population dynamics of *R. magnicornis* in the Tweed Valley. This suggests that green cane trash blanketing is not likely to be an effective control measure in NSW as it has been in Queensland.
Figure 12: Density map of rhopaea canegrub in a single field based on samples spaced at regular 20 m intervals. The field has a barely perceptible slope away from the lowest headland. Over 60% of all the larvae collected in early September 2000 were larvae undergoing the 1st year of a two-year life-cycle.
Figure 13: Density map of rhopaea canegrub in a single field based on samples spaced at regular 10 m intervals. The field gradually rises as the distance along the field (y-axis) increases. Between 120 and 130 m there is a dramatic change in soil texture from a light ‘peat’ to a heavy clay ridge.
5.0 DISCUSSION

5.1 Distribution of rhopaea canegrub in the soil

The distribution of rhopaea canegrubs in the soil under sugarcane has been studied for the first time. The confirmation of its relatively shallow living habit has implications for both its control and for sampling.

**Sampling implications**

Most sampling of scarab larvae in sugarcane production systems is based on an approximate cube of soil excavated around and including a sugarcane stool. The size of this sample is generally reported as being 30 cm wide x 30 cm long x 30 cm deep. In such instances, the area included in each sample extends only 15 cm into the inter-row space either side of the centre of the sugarcane row. In most instances these samples have been taken during the time that third instars are present in the soil.

The results from my observations of rhopaea canegrub indicate that populations of third instar larvae are best estimated by samples with a width of 40 cm (ie extending 20 cm away from the row centre into the inter-row space). This can be seen from Figures 5 and 6, where in all instances at least 90% of the total third instars encountered would have been encountered by using a trench of these dimensions.

However, my results clearly show that wider samples are important in determining the population of early instar larvae and the later pupal stages.

While eggs are generally oviposited within 20 cm of the row centre, first instar larvae are frequently found further out in the inter-row. Figure 2 shows that 90% of first instars are likely to be encountered if the trench boundary extends to at least 50 cm either side of the row centre (ie 1 m wide trenches). While this does increase the sampling effort required to estimate early larval populations, this increase need not necessarily be as great as it first seems. The main reason is that mobile first instar larvae (as opposed to those newly hatched larvae still in the location where they were oviposited) tend to be found at fairly shallow depths in the soil profile as they move further out into the inter-row space (see Figure 1). Therefore, the width of row excavated to estimate the population of first instar larvae will be shallow at the outer extremities.

A similar situation exists also for sampling when pupae are present. The indications from sampling at the ‘artificial’ site are that, just prior to pupation, third instar larvae move away from their feeding site close to the stool. This final dispersal activity places many pupae outside the standard 30 cm wide sample. Again, though, pupae are typically shallow and so, even though a wider trench may be advocated for sampling when pupae are present, this trench would not need to be deep at the extremities in the inter-row space.

Second instar larvae are more aggregated around the stool than first instar larvae and pupae but less aggregated around the stool (row centre) than third instar larvae. Third instar larvae are the stage that is most aggregated around the stool. This is not surprising because third instar larvae are the most aggressively feeding stage in scarab larvae, and the cane stool represents a large concentration of root (ie food) material.
Table 11 contains my recommendations for the optimum sample sizes for obtaining accurate estimates of rhopaea populations in canefields for all stages, based on the observed distribution of rhopaea larvae in this study.

**Table 11: Suggested sampling method**

<table>
<thead>
<tr>
<th>Month</th>
<th>Dominant stages</th>
<th>Distance to excavate either side of row centre (for &gt;90% accuracy)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dec/Jan</td>
<td>eggs/L1</td>
<td>50 cm</td>
</tr>
<tr>
<td>Feb/March</td>
<td>L2</td>
<td>30 cm</td>
</tr>
<tr>
<td>April/July</td>
<td>L3</td>
<td>20 cm</td>
</tr>
<tr>
<td>Aug/Oct</td>
<td>L3/pupae</td>
<td>30 cm</td>
</tr>
<tr>
<td>Nov</td>
<td>Inaccurate sampling</td>
<td>n/a</td>
</tr>
</tbody>
</table>

5.2 GCTB and stool rolling

The fact that I did not detect a clear effect of GCTBs on rhopaea’s survival does not mean that such an effect could never occur under NSW sugarcane. However, under the conditions of the trials, both bare soils and soil trash blankets were severely waterlogged for most of the year. GCTBs are probably better assessed for their effect on rhopaea mortality under drier growing conditions, since the relative difference in soil moisture under a trash blanket would be greater than at those times than when the soil profile is essentially waterlogged.

Generally, low larval populations in both trash blanketing and stool rolling sites made it difficult to detect any real effect of these practices on rhopaea canegrub. However, even if they do contribute some mortality, it appears that their effect is relatively minor.

5.3 Discussion of fallowing results

1999 observations

The low rhopaea populations in sites 1-3 were probably related to the generally low rhopaea numbers found throughout the Tweed Valley in 1999. These low numbers appeared to come from the combined effect of wet weather and diseases on rhopaea survival. Thus, seasonal factors had a greater effect on rhopaea abundance at these sites than any possible influence of fallowing.

However, the fallow planted blocks in site 4 were some of the few blocks sampled in 1999 where rhopaea canegrub was not scarce, and it was at this site where a clear difference was found between the fallow planted and ploughout/replant blocks. The result from site 4 is somewhat difficult to explain because the expectation was that if fallow planting had any effect, it would be to reduce the canegrub population rather than increase it.

Site 4 was selected for study in late 1998, two years after the six blocks at this site were replanted with sugarcane. Therefore, the fallow period prior to replanting occurred two to three years before the commencement of this study. In 1998, no questions were asked about how the fallow was managed in 1995-96. A bare fallow was assumed. However, after the unexpected result, the farmer explained that during the fallow period, those blocks had a lot of grass cover.
Fallowing is supposed to work for cane grub control at least partly because of the lack of food for any remaining grubs during the fallow period. However, the presence of abundant grass cover during the fallow period at site 4 indicates that there would have been a food supply for any larvae left in the fallow blocks after the initial ploughout operation.

Therefore, it was reasonable to conclude that the higher rhopaea densities under fallow-planted sugarcane at site 4 related to the presence of significant grass growth during the fallow period. That is why I designed an experiment to assess the ability of rhopaea larvae to persist in the soil during a long fallow period under different fallow regimes.

**Larval persistence during a long fallow period under different fallow regimes**

The data in Figure 10 do not confirm the theory that grass in the fallow blocks at R Brown’s was the major contributor to higher numbers in the fallow planted cane in 1999 relative to the adjacent ploughout/replant blocks. However, more trial work would be required to exclude grass fallows as a risk factor for rhopaea cane grub damage.

The minimum till fallow was as effective in limiting grub persistence as the other fallow regimes, despite the fact that there was substantially more crop residue present in this treatment and available as food for the added canegrubs. This suggests that rhopaea needs living cane roots for food to maximise its survival. Another possibility is that a lack of soil disturbance in this treatment did not disrupt the action of canegrub pathogens as much as the cultivated fallow treatments.

**5.4 Spatial distribution studies**

Through these studies I was able to identify two broad features of individual cane fields that may influence the distribution and abundance of rhopaea within a single canefield.

**Soil waterlogging**

At Site 1 (Martin), I found a significant correlation between rhopaea abundance and the distance of the sample point from the lowest headland. The highest abundance of rhopaea larvae was found in the highest parts of the paddock. In the previous year, when extremely wet weather had prevailed, the lowest parts of the field were the most subject to inundation. My results indicate that rhopaea mortality was most likely to be highest in soils where waterlogging was most persistent. This trend was most pronounced in the 2000 sampling, but was also evident the following year. The apparent effect of waterlogging on rhopaea abundance I detected at this site ties in well with the overall lack of canegrub abundance I detected during the 1999 sampling and in the following two seasons.

**Soil texture**

At Site 2 (Grippo), sampling revealed abrupt changes in rhopaea density that correlated exactly with an abrupt change in soil texture. In 2000, the highest populations were found in the lighter-textured peat soil, whereas the population fell to almost 0 larvae/sample in the heavy clay ridge at the other end of the paddock. Interestingly, the situation was
reversed in the following year, with an almost negligible larval population in the light soil (where the ratoon crop was very poor as a result grub damage the previous season), but with significantly higher numbers around the stools on the clay ridge (undamaged).

The result in 2000 was unsurprising because growers had always insisted that grubs were not a problem in ‘heavy’ country. Even the 2001 results were not that surprising because the cane appeared undamaged and healthy, despite carrying an average of 2.15 grubs/stool. What was surprising, though, was the massive decline in grub numbers in what had been the most suitable habitat the previous year, ie the light peat soil.

There are three main competing explanations for this observed decline from 2000 to 2001. The first suggestion is that the damaged crop with its gappiness, reduced stool mass and root vigour provided less food for an initially high 2001 larval population. These were effectively starved out, resulting in a population crash. It is also possible that a disease epizootic (eg *Metarhizium*) massively affected rhopaea survival in 2001. Both these scenarios rely upon a major mortality event. An alternative hypothesis is that the majority of female beetles emerging from the damaged cane section in late 2000 did not mate on the ground but instead flew away before mating and ovipositing elsewhere. It is even possible that the increased numbers of rhopaea in the heavier soil were due, not to natural increase, but to an influx of female beetles from nearby damaged stools. Perhaps the taller, undamaged cane in this heavier soil was attractive to female beetles seeking mating/oviposition sites.

6.0 ASSESSMENT OF LIKELY OUTCOMES OF THIS RESEARCH

This project has concluded just as grub numbers in the Condong area are resurging. On some farms damage not seen since the 1980s has become evident with up to 40 grubs per stool being found in some blocks.

The project has identified fecundity and survival rates of the pest and detailed the distribution of the pest through the soil over time. This and other information gained through the project will be valuable when sampling and/or applying control techniques. Fallows should be maintained in a grass-free state to reduce pest populations. Green cane trash blanketing did not appear to be effective in reducing numbers. With a proposed move to whole crop harvesting for cogeneration and the problems associated with cool wet weather in NSW, trash blanketing would not be an attractive proposition in any case. Stool rolling was not shown to be effective and is likely to decline, which will be better for soil and crop health.

Work during the project has confirmed the effectiveness of ploughout/replant as a control measure. The project has also identified when and where to place knockdown insecticides as an alternative to replanting. Local trials have shown promising results with Confidor® (imidacloprid). This research has shown that application of this chemical in November to December should provide effective control.

In the longer term, the project has identified a number of potential bio-insecticides based on *Metarhizium* and *Beauveria* fungi.
This project was also successful in seeding small experimental plots with larvae using the oviposition of field-collected mated female beetles. We do not know whether this technique has previously been applied to canegrub research. In another experiment, field-collected rhopaea larvae were successfully introduced and confined inside experimental mini-plots using plastic barriers. Similar techniques have occasionally been attempted with other canegrub species but not always successfully. Both these experimental techniques are discussed in the following section.

7.0 TECHNICAL SUMMARY

Two methods were developed and tested for their applicability to field studies of *R. magnicornis*. These are emphasised since they may also be applicable to the study of other canegrub species and would definitely be appropriate for further research on rhopaea canegrub.

7.1 Artificially infesting known sections of a canefield using field-collected mated female beetles

Low naturally occurring infestations of rhopaea canegrub in 1999 and 2000 meant that it was difficult to obtain data on the distribution of rhopaea’s life stages. To maximise the number of rhopaea individuals encountered per excavation, I attempted to increase the naturally occurring density of rhopaea larvae at specific known locations in a single field.

I did this by placing field-captured mated females at known locations, permitting them to burrow into the soil at these locations and subsequently oviposit. By artificially boosting oviposition at known sites, I was able to excavate these locations later, confident of obtaining more individual observations on the distribution of rhopaea individuals than if I had relied on natural oviposition alone.

This technique was successful and females did oviposit under the stool where they were originally placed.

The main drawback of this method is that the precise number of eggs/larvae initially placed into an excavation cannot be known since this relies on knowing the exact number of eggs laid by the female beetle placed in that location. However, the relationship between a female’s mass at mating and her realised fecundity under field conditions was investigated using linear regression. In this way, it was possible to estimate realised fecundity under field conditions based on female mass and the oviposition data obtained early in the trial.

This technique is probably limited to use with canegrub species that have similar mating, dispersal and oviposition habits to rhopaea. Rhopaea females only lay one egg batch and do not disperse again after oviposition.

Also, because mating or gravid females are not attracted to light traps or feeding trees (as in greyback canegrub), collecting a lot of mated/mating females for experiments can be quite difficult. It essentially involves walking up and down many rows of cane with a torch after dusk, trying to spot females as they are either mating on the ground or hanging from the leaves of ratoon cane. Ground mating females must be captured during the flight period (about 20 minutes maximum, after dusk) before they burrow into the soil. Leaf-
mating females stay attached to both their mate and the cane plant for at least several hours after the flight/mating period. Observed mating behaviour was clearly consistent with the use of a female pheromone to attract male beetles (Allsopp, 1993).

7.2 The use of physical barriers to confine larvae within small (1 m x 1.5 m) experimental plots

The use of physical barriers to confine larvae within 1 m x 1.5 m experimental plots appears to have been successful, with no evidence that large numbers of larvae (if any) were able to escape.

In the cultivated plots to which 20 larvae were added in March (ie bare, legume and grass fallows), larval recovery in April was generally high. Even in the bare-fallow plots where I may have expected grubs to be more mobile in search of food, initial larval recovery was quite high, ranging from 6-15 larvae in April (mean = 9.8 rhopaea/plot, n=5). I should also note that despite all due care, some of the larvae placed into these experimental plots may have been injured during handling, resulting in mortality. Secondly, by April, there might also have been considerable mortality associated with the moulting from second instar to third instar larvae.

A further piece of evidence suggesting that rhopaea larvae did not escape through the barriers used in this experiment is that the majority of larvae recovered were not found in close proximity to the barriers. None were found in the soil immediately outside the barriers though excavations in this zone were limited. In light of all these factors, the apparent mortality rates evident after the initial setting up of this experiment are not so high as to warrant suspicion of significant grub escapes from the experimental plots.

Overall, the recovery data suggest that any observed differences between the number of larvae introduced and subsequently recovered per plot can be generally attributed to mortality rather than escape.

The barriers in this study were made from builders’ black plastic. At least one other study (Ward, 1998) used metal steel plates pushed into the soil. I did not have the financial or workshop resources to have these barriers made up but the plastic did not seem inferior in terms of confining the grubs to the mini-plots, and was certainly cheaper.

The shallow-living habits of R. magnicornis larvae meant that the trenches to accommodate the plastic barriers did not have to be very deep to prevent larvae from escaping beneath the fence. Therefore we did not need to worry about putting the plastic ‘fences’ down below 40 cm. In practice, the heavy clay subsoil began about 30-40 cm beneath the lighter peat soils, and it is known that rhopaea canegrubs do not penetrate into this soil layer. Therefore, by extending the barriers 5-10 cm below the beginning of this clay layer we ensured that no grubs would escape by burrowing underneath the plastic.

The trenches in one direction were excavated using a small ‘self-propelled’ trenching machine (“ditch witch”) hired from a local landscaping business, which was not suitable. The trenches in the other direction (completing the square) were dug with a mini-excavator. Subsequent workers would do better to consider excavating either all the trenches with a mini-excavator and operator, or by hiring (with operator) a heavier duty trenching tool such as is used for laying agricultural pipes or telecommunications cables.
8.0 RECOMMENDATIONS

8.1 Pest management

_GCTB and stool rolling_

Due to the low rhopaea populations in my trial sites during 1999, there was no clear evidence that either green cane trash blanketing or stool rolling suppresses the abundance of rhopaea canegrub. Stool rolling appears to be necessary for stool recovery in situations where the stool has been loosened by a combination of either grub, harvester or wind (lodging) damage.

_Fallowing_

Fallowing itself cannot be relied upon to give better pest management in the subsequent crops. Increased persistence of rhopaea larvae under certain fallow regimes could be a factor, with results suggesting that grass growth during the fallow period is a risk factor. In any case, the relationship between long fallowing and subsequent rhopaea abundance is not straightforward. The two most likely explanations for the result observed at Robert Brown’s where fallow planted cane had significantly higher canegrub numbers than ploughout/replant blocks are:

- that fallow planted crops may be more attractive to flying ovipositing females than replant cane (ie increased immigration into fallow planted blocks);
- that long fallowing alters some aspect about a field, making it a more suitable habitat for rhopaea canegrub (with higher survival/lower mortality of canegrubs). Diseases could be implicated in this. A reasonable hypothesis could be that long fallowing reduces the abundance of pathogen hosts, in turn reducing the pathogen load in the fallowed area, creating a more favourable environment for canegrub survival and population increase.

Further testing of these hypotheses is required.

_Harvesting of lightly damaged blocks_

There is no indication that canegrub numbers necessarily keep increasing each year until they are checked by ploughout. Indeed, it is likely that numbers will decline after a damage incident (migration effect or disease). Therefore lightly damaged crops should be harvested with care if there is an intention to try and ratoon that crop. This will result in less stools being damaged and removed, and therefore a more productive ratoon crop.

_Insecticide options_

Growers may consider applying suSCon® Blue to early cut young ratoon crops, especially if they are adjacent to blocks with large populations of canegrubs (and therefore close to a potential influx of ovipositing female beetles). Plant crops are not likely to function as trap crops for mobile rhopaea females in the NSW industry where autumn planting is not practised. In NSW, cane is planted in September so is only two months old by the time of the main beetle flights in November. It is certainly not 0.5 m tall by this time, which is the height at which cane can function as a trap crop for female greyback beetles in north Queensland (Ward, 1998). suSCon® Blue is not an easy product to apply
into ratoon crops so there is an opportunity for other knockdown or systemic products to be applied to fields identified as ‘at risk’.

8.2 Further research

Diseases obviously play an important role in the population dynamics of rhopaea canegrub under sugarcane. Some specific trials that measure the effect of long fallowing versus ploughout/replant on pathogen abundance would be useful. Minimum-till fallowing should also be assessed for its ability to foster insect pathogens.

Research into the flight behaviour of female rhopaea beetles should be a priority. The amount and impact of migration by female rhopaea beetles was greatly underestimated by the previous study into *Rhopaea magnicornis* (Hayes, 1998) and I made only qualitative observations.

Appendix 2 contains some suggestions for experimental work that would test some preliminary hypotheses about what may trigger flight in female beetles. If we can understand the factors that make rhopaea populations move around then farmers may be able to identify ‘at risk’ fields and take preventative actions (eg trap cropping, insecticide application into ratoon crops).

9.0 PUBLICATIONS

No journal articles have been published to date.

Mr Austin McLennan is still preparing his Master’s degree thesis for submission to the University of Queensland. When completed, SRDC will receive a copy of this thesis. Copies will also be distributed to his academic supervisors, Dr Peter Allsopp (BSES) and Professor Myron Zalucki (UQ), and to Mr Peter McGuire (BSES).

10.0 ACKNOWLEDGMENTS

Thank you to all the growers who allowed me to sample for rhopaea canegrub on their farms and who co-operated in the implementation of field experiments. Growers I would particularly like to thank include Graham Martin, David Cowderoy, Robert Hawken, Bill Stainlay, Joe Grippo and Graham Partridge. Thanks also to those casual workers who assisted in data collection. Dr Wayne Rochester, UQ, was invaluable in the analysis of the spatial data. I would also like to thank Dr Peter Allsopp for his comments on the draft of this report and Prof. Myron Zalucki for discussions. Finally, I must thank Mr Peter McGuire for his assistance throughout the project, and especially for his efforts in the preparation of this report.
11.0 REFERENCES


Robertson LN, Dall DJ, Lai-Fook J, Kettle, CG and Bakker P (1998). Key factors in control of greyback canegrub populations. SRDC Final Report SD98014, BSES.


APPENDIX 1

Generalised phenology of *R. magnicornis* in the Tweed Valley, NSW.

<table>
<thead>
<tr>
<th>Month</th>
<th>One-year larvae</th>
<th>Two-year larvae (first year)</th>
<th>Two-year larvae (second year)</th>
</tr>
</thead>
<tbody>
<tr>
<td>June</td>
<td>L3 (feeding)</td>
<td>L3 (difficult-impossible to distinguish from one-year L3s).</td>
<td></td>
</tr>
<tr>
<td>July</td>
<td>L3</td>
<td>L3 (two-year form impossible to distinguish).</td>
<td></td>
</tr>
<tr>
<td>August</td>
<td>L3</td>
<td>L3 (two-year form impossible to distinguish).</td>
<td></td>
</tr>
<tr>
<td>September</td>
<td>Pupation commences.</td>
<td>Pupation commences.</td>
<td></td>
</tr>
<tr>
<td>October</td>
<td>Adult flight and mating commences.</td>
<td>Adult flight and mating commences.</td>
<td></td>
</tr>
<tr>
<td>November</td>
<td>Mating flights/oviposition. First eggs can be found.</td>
<td>Mating flights/oviposition. First eggs can be found.</td>
<td></td>
</tr>
<tr>
<td>December</td>
<td>Some mating flights continue. Oviposition continues. Eggs dominate and some first instar larvae can be found.</td>
<td>As for one-year larvae.</td>
<td></td>
</tr>
<tr>
<td>January</td>
<td>First instar larvae common. Some eggs can still be found. Early second instars also may be detected.</td>
<td>As for one-year larvae.</td>
<td></td>
</tr>
<tr>
<td>February</td>
<td>Most larvae are second instars. Occasional early third instar</td>
<td>As for one-year larvae.</td>
<td></td>
</tr>
<tr>
<td>March</td>
<td>Most “one-year” larvae have by now moulted into third instar larvae.</td>
<td>Most remaining second instar larvae are now ‘committed’ to the two-year development timeframe.</td>
<td></td>
</tr>
<tr>
<td>April</td>
<td>All third instar larvae are feeding and will continue to do so until they commence pupation in September.</td>
<td>All second instar larvae will remain as such until they moult into third instar larvae around September.</td>
<td></td>
</tr>
<tr>
<td>May</td>
<td>Feeding as L3</td>
<td>Feeding as L2</td>
<td></td>
</tr>
<tr>
<td>June</td>
<td>L3</td>
<td>L2</td>
<td></td>
</tr>
<tr>
<td>July</td>
<td>L3</td>
<td>L2</td>
<td></td>
</tr>
<tr>
<td>August</td>
<td>L3</td>
<td>L2</td>
<td>Some may commence moult into third instar larvae.</td>
</tr>
<tr>
<td>September</td>
<td>Pupation commences.</td>
<td>By the end of September all surviving two-year larvae will have moulted into L3s.</td>
<td></td>
</tr>
<tr>
<td>Month</td>
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<td>L3 (able to distinguish from one-year L3s at this point, ie bigger and more yellow in colour).</td>
<td></td>
</tr>
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<td>L3</td>
<td></td>
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</tbody>
</table>
APPENDIX 2

Suggested protocols for initial investigations into triggers of flight by female *R. magnicornis* beetles

Aim:

The aim of these proposed experiments is to gain some preliminary insights into the proportion of female beetles that do fly, and what might trigger emigration of females from infested fields.

Hypothesis:

Fields with higher rhopaea densities may be more likely to produce females that, when emerging from the soil, are more likely to take to the wing, mating and laying eggs in a site some distance from where they emerged.

The basis of this suspicion is that I noticed high numbers of beetles mating on the leaves of a crop that did not apparently have a high larval density. These high numbers of mating pairs seemed equivalent to numbers in a nearby damaged crop. I suspected that they may have come from the neighbouring field that did have high larval populations, where damage to the cane was evident.

It is reasonable to suggest that female beetles from high-density damaging populations are less well nourished than grubs from low density areas (due to competition for food resources). This less-nourished physiological state may act as a trigger for female flight. It makes sense that limited food supply (resulting in less mass) could act as a trigger for female flight.

The experiments here are to identify a possible trigger for flight initiation. There is no attempt to determine mating or oviposition site selection.

In all the following proposals, the number of replicates/sites etc. should only be taken as a guide.

Experiment 1:

**Aim:** To determine whether grubs/female beetles from damaged fields are lighter (less nourished) than those from undamaged fields.

- Identify three fields with damaging populations and several more with lower populations.
- Collect about 30 larvae from each field if possible, weigh and discard. Sample to estimate the density of canegrubs at that site.
- **Data analysis:** Compare mean weights from each of the populations.
- Include a linear regression of (population) density of field versus weight of larvae. The weight of individual larvae could also be plotted against the density of the sample from which it was collected to see if there is a negative correlation (where density equals the number of rhopaea larvae in a sample).
Experiment 2:

Aim: To determine if heavier beetles are less likely to fly.

- Prior to the flight season collect non-emerged female beetles, perhaps from a high density and a low density site.
- Weigh beetles, mark each one with a liquid paper dot or similar.
- Put into plastic containers of some kind, place into field during flight season. Record the air temperature and other weather conditions.
- Collect and return containers to the laboratory that night. Check for escapees. Record data onto form (ie escapees, mating beetles or mated in container).
- Set out beetles again the following night, repeat process.
- Continue process for five nights.
- **Data analysis:** Three groups - escaped, mated in container or did not escape/unmated. Compare the mean weights of these three classes of beetles. Is there any significant difference? Report the simple percentage of each class.

Experiment 3:

Aim: To determine if beetles that fly to mating sites on leaves are lighter than ground-mating beetles.

- Collect 20 females mating on ground over two nights.
- Collect 20 females mating on leaves over same two nights.
- **Data analysis:** Weigh/measure female beetles. Is there any significant difference between the two groups (ie leaf-mating versus ground mating)?