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1 Acknowledgments

The project team would like to acknowledge the inputs of numerous smallholder coffee farmers in the highland provinces of Papua New Guinea; these inputs ranged from information about their farm operations, to views about the green scale problem and advice.

We would also like to thank in a similar vein others in the coffee industry, research and extension staff of R&GSD, CIC, and nursery growers, who have frequently given up their time to help with some of the project activities. Additionally, we would like to thank Mr Katayo Sagata formerly of the Wildlife Conservation Society-PNG.

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2 Executive summary

The overall aim of this research project was to improve smallholder returns from coffee production through participatory improvements to integrated control of coffee green scale (CGS). The major CGS problem in PNG seems to be limited altitudinally (to approx. 1500masl; metres above mean sea level) and the main species involved is *Coccus celatus*, a species that originates from East Africa. Another species of CGS is present, *Coccus viridis*, also from East Africa but this species appears less common. The scales are mostly attended by ants and surveys have shown that the major species involved are also alien invasive species.

The socio-economic surveys covered smallholders in the three major coffee growing provinces. In general it was found that food production is the main priority for smallholders. For Eastern and Western Highland Provinces most farmers who produce coffee have been doing this for ten or more years; i.e. there are few young farmers entering into the system; but 65% of coffee production is done by women. Income from coffee is mostly used to pay for school fees. The low priority placed on coffee probably contributes to the fact that smallholders do not manage coffee very well and yield production is low. The awareness and understanding of ants and CGS are also very limited among smallholders and a major barrier to uptake of proactive management. But some farmers have ‘experimented’ with control measures and these include shade management and the use of pumpkins to attract ladybird species. A very important issue is that farmers are in general unaware of the production potential of their coffee crop and also the losses that may be incurred. Losses that can be assigned to CGS attack are not known and need urgent study.

Experimental studies indicated that a number of factors seem to be generating the population outbreaks of CGS. High shade is an important limiting factor for CGS. Where shade is not used / has been removed then infestations are higher. Also, and importantly, local natural enemies, in the absence of ants, are able to suppress CGS infestations to insignificant levels. The continued movement of CGS through human mediated and natural pathways appears to have been a significant factor contributing to the spread of scale.

The project greatly increased CIC’s capacity to undertake classical biological control. CIC now have much improved rearing facilities and also protocols for classical biological control. One agent was introduced from Australia for CGS: the host specific parasitoid, *Diversinervus stramineus*. Classical biological control options can be considered but a decision has to be made by relevant authorities in PNG about the relative merits of introducing agents when good local natural enemies exist.

Based on the results of the project studies, there are a number of key components that need to be included and validated in a management plan for CGS: viable existing local controls; banding to deter ants, but the cost of this for farmers in different income brackets need to be investigated; and the development of awareness schemes to alert farming communities about CGS. This may include talks and posters etc. Project impacts include: awareness about CGS and ants have been raised in the project area; knowledge and skills of key staff in CIC in ecological assessment and biological control have been developed and research staff at all levels (including Extension Officers) have been trained in a range of methodologies for generating socio-economic data. Key recommendations from the project include: a general study on coffee yield loss due to CGS to be undertaken as a matter of priority; CIC to produce a dossier on the CGS problem and the benefits and risks of releasing *D. stramineus*; the management plan to be tested in a few coffee farming villages to measure suitability, cost, degree of uptake and impact on CGS; another important aspect is that the management plan needs to be institutionalised within CIC’s strategy on coffee development.

3 Background

3.1 Key Issues

Coffee is the largest earner of foreign exchange within the Papua New Guinea (PNG) agricultural sector (> 43% of PNG agricultural exports and 10% of total exports), and is primarily produced by smallholders (85–90% production), with few management inputs. Despite the low inputs, smallholders produce reasonable quality coffee beans due to available family labour, timely harvesting and links to processors who provide assistance with finance and inputs. The highlands region depends almost entirely on coffee (250,000–300,000 smallholder families, in 2002). Although many farmers lack organisation, assets, inputs and education, a new generation of better educated farmers will become more significant over the next decade and this will increase the need/demand for strategies to improve productivity. With the general high regard for PNG coffee and reasonable demand for top qualities, there is scope for smallholder farmers to improve incomes through improvements in productivity with relatively low increases in inputs.

Because smallholders can afford few inputs, one of the major factors limiting productivity is pests (the most serious disease threat is currently controlled adequately by resistant cultivars), with coffee green scales (CGS) the most serious (causing up to 50% yield reduction at the CIC Aiyura field station). In consultation with smallholders, the PNG CIC has confirmed that there is an urgent need to develop and introduce integrated CGS management recommendations suitable for smallholders as well as estates. The overall aim of this research project was to improve smallholder returns from coffee production through participatory improvements to integrated control of CGS. This project therefore addressed the following research and development objectives:

1. To document baseline information on the distribution, impacts, biology and control of CGS in the coffee growing regions of PNG;
2. To evaluate biological and other control methods of CGS, taking into account grower information;
3. To develop regional and national strategies for wider evaluation and implementation of CGS control.

The key issues within these objectives included: the development of a better understanding of what the pest species are and why they are such a problem; establishment of the best short term strategy based on currently available methods; development of a long term sustainable strategy based as much as possible on use of natural and cultural control methods; fostering the engagement and commitment of smallholders into this strategy; harmonisation of this strategy with any overall strategic plans for PNG coffee and any certification requirements that export coffee may be subject to; and skills training for PNG research and extension.

3.2 Project Justification

The socio-economic and ecological constraints to coffee production in PNG suggest that measures to control CGS must be tailored within these constraints, with external support provided by additional biocontrol agents (following one introduced in 1986). PNG coffee is mostly grown without the use of chemical inputs, which has provided a unique footing for the industry in the 'organic' boutique market. Smallholder coffee growers will be the primary beneficiaries of this project, with benefits flowing on to plantation and block holders, processors, exporters and consumers through improvements to the supply and quality of PNG coffee. Australia has a small (organic) boutique coffee industry which supplies a specialist local market but the primary benefit for Australia from this project is through the assistance to the economic development of PNG. Additionally, although CGS not been recorded in Australia, its presence in PNG presents a risk, and control options developed in PNG may be transferable should the necessity arise.

In 2004, approximately 68,177 tonnes of coffee were produced from 96,584 ha, with average smallholder plantings of 20 ha. This production involved 397,000 families and earned K300 million (AUD 120 million). The 20 ha block sector and plantations account for the remaining 15% of production in 2004. Current average losses due to CGS are currently estimated circa10% (and up to 50%), which represents at least K30-40 million in lost income for PNG (CIC estimates that farmers get 50–60% of FOB price, i.e. circa K2.50/kg.)

PNG has a comparative advantage in coffee production. The climate and many of the highland soils suit the crop and production requirements and market prospects suit smallholder needs and aspirations – this is a suitable cash crop even for remote communities (Fleming, 2002) because of the relatively high value/kg, storability and because it can be transported easily. While coffee research and development (R&D) has had a long history in PNG and a number of successful innovations continue to provide benefit, such as the use of rust resistant cultivars, a key community need now is to focus on innovations that are useful to and adoptable by smallholders, for whom affordability of inputs, availability of labour and access to finance are often limited. Recognising these constraints, a number of the coffee processor organisations work with growers by providing access to inputs and finance, while some farmer co-operatives are also proving successful. Working with these sectors of the industry has been a key strategy for development and implementation of improved controls that complement (and do not interfere with) the impacts of any biocontrol agents that have proved to be suitable.

The research and development challenges have included:

- *Assessment of the nature of the problem*, including the biological and environmental factors influencing scale infestations.
- *Assessment and understanding of socio-economic constraints*, fostering participatory involvement and uptake of integrated pest management (IPM) by smallholders
- *Development of additional IPM approaches*, including the possible strategic use of pesticides and ant control strategies that accommodate smallholder needs and optimise the chance of success of biocontrol approaches.

The priority for work on CGS was agreed at the PNG National Coffee Workshop in 2002 and confirmed at the 2004 ACIAR-PNG consultations. The idea for the project was submitted to ACIAR by CIC in 2004 and agreed as a priority for CABI funding in the 2004 ACIAR-IARC funding round.

The proposed work fitted within the overarching strategy for PNG in the 2005–2006 ACIAR Annual Operational Plan for applied technical research aimed at the enhancement of incomes for smallholders, with an emphasis on plantation crops. The proposed work also met ACIAR's priority for project specific funding for IARCs, of strengthening links between Australian research institutions, national research systems (coffee smallholder R&D) and IARCS (key focus/comparative advantage in coffee R&D) and helping to focus the ACIAR-CABI support on an ACIAR bilateral priority (PNG/smallholder benefit).

The project concept was developed during 2004–05 through interaction between ACIAR, CIC, CABI and the University of New South Wales (UNSW).

The project capitalised on the synergies with ACIAR projects on marketing (ASEM/2004/042) and postharvest quality management (PHT/2004/017) in undertaking surveys and in assessing socio-economic aspects.

4 Objectives

The overall aim of this project has been to improve smallholder returns from coffee production through participatory improvements to integrated control of CGS. This was addressed through the development of the following objectives:

Objective 1: To document baseline information on the distribution, impacts, biology and control of CGS in the coffee growing zones of PNG.

Activity 1: Survey the incidence and impacts of CGS and their associated insects.

Activity 2: Survey of current control methods by growers, including the rationale for such measures.

Objective 2: To evaluate biological and other control methods of CGS, taking into account grower information.

Objective 3: To develop regional and national strategies for wider evaluation and implementation of CGS control.

The following sets the context and approach planned for the project.

Because CGS has spread within a human mediated system, the biological status included assessment of the severity and extent of economic damage caused by CGS and how its impact is related to key variables such as altitude, associated vegetation and soil; also, the relative abundance of associated species, e.g. ants, predators and parasitoids which affect colonisation levels.

A key to understanding the nature of an enduring problem was to ask relevant stakeholders for their inputs and to elucidate basic information about availability of labour, time, money and other major variables that might affect the choice of control measures to test. These are usually best carried out through the use of socio-economic survey instruments, and less formal ethnographic techniques (section 5).

The project did not attempt to import new species of parasitoids from Africa or elsewhere because it was deemed cost- and quarantine-ineffective (and would require a separate project). The project facilitated actions of another project (EU funded) that is attempting to introduce the exotic parasitoids *Diversinervus stramineus* and *Coccophagus* spp. from the Department of Primary Industries (DPI), in Queensland.

5 Methodology

All field and experimental work was conducted in the Eastern Highlands Province (EHP) of PNG. Methodological strategies were developed to address and operationalise the research objectives.

Socio-economic details were generated in stages to address parts of research by CIC and UNSW. Ecological and biological control components used a combination of farm surveys and CIC station experiments conducted by CIC and CABI. The research station at CIC Aiyura (EHP) was used as the central base for research, and the quarantine facility for the development of rearing biological control agents.

5.1 Objective 1

5.1.1 Distribution Studies

Preliminary ecological survey

In November 2006 a preliminary survey was conducted in 47 smallholder farms, selected to cover a range of altitudes and shade levels where Arabica coffee is typically grown in the EHP. It was deemed sensible to focus mainly in the EHP as this area is representative of other coffee growing areas; it was also logistically easier.

CIC staff were trained in survey methods and used a diagonal transect method for assessing CGS presence at each farm. In this method the surveyor walked diagonally across a coffee garden and recorded the presence / absence of CGS, ants, ladybirds and the entomopathogen *Lecanicillium lecanii* on individual trees. The proportion of trees affected by CGS, etc., on the diagonal was then calculated.

Data analysis

Step one was to determine simple relationships between the variables surveyed, e.g. between CGS and ant abundance, using Spearman rank correlations (rho) with a two tailed test of significance. Other environmental factors such as altitude and shade were then analysed using the same method.

Step two used the Generalized Linear Model (GENLIN) function in SPSS (version 16) to conduct a regression analysis. As is common with count data it was highly skewed and the analysis conducted was specified as a negative binomial distribution (Pedigo & Buntin, 1994; SPSS, 2008). Altitude and shade were entered as factors and all non-significant factors were sequentially removed to simplify the model.

Major ecological survey

In order to clarify the findings and confirm the accuracy of the diagonal transect method in the preliminary survey, a second survey was conducted in 2007. Here 128 farms were surveyed using two methods; a diagonal transect survey (as before) and a whole block survey. This latter survey was a more robust approach and allowed for the confirmation of the preliminary survey results.

A diagonal transect walk across the farm was made and every tree encountered was assessed for presence / absence of CGS, ants and predators. The whole block survey included every tree. Data analysis was conducted as for the previous preliminary survey. Table 5.1.1 shows the measurements taken and for a full description of the methods used in both preliminary and major surveys see appendixes 11.1 and 11.2.

Table 5.1.1: Variables surveyed with both diagonal transect method and whole coffee block method.	
Coffee Green Scale Presence.	Pest of coffee.
Ant Presence.	Formicidae – associated with CGS.
Ladybird Presence.	Coccinellidae – natural enemy of CGS.
Lecanicillium lecanii Presence.	Entomopathogenic fungus – natural enemy of CGS.
Altitude.	Several altitudinal zones surveyed.
Shade Level.	Four levels of shade; 0 shade; <30%; 30-75%; >75%.
Farm Size.	Three groups, based on farm size; <500 trees; 500-1000 trees; >1000 trees.
Coffee Age.	Age of coffee trees; young/immature; mature; mix of both.
Garden Site.	Aspect and topographical position; east facing slope; west facing slope; valley; hill; valley and hill.

Ant diversity survey

In order to identify possible problematic ant species associated with CGS a survey was commissioned by the project team and conducted by Mr Katayo Sagata (formerly of the World Conservation Society, PNG) (see full report: Sagata, 2008). Four coffee farms were chosen situated along the Okuk Highway, within the EHP. These farms ranged over an altitude of 1410–1714masl (metres above mean sea level) and were sampled between January and March 2008.

To measure ant diversity and abundance and their spatial distribution within coffee farms, five 50m long transects were established at 5m intervals from the adjacent road at each farm. More detail is given in appendix 11.3, but a brief summary of the measurements is given below:

- Surface dwelling ants and arboreal ants: sampled using pitfall traps (for surface dwelling ants) and baited traps (for arboreal ants). All species were identified and reference collections retained.
- Effect of canopy cover and shade tree size on ant diversity and abundance and CGS abundance.
- Ant species attending CGS.

Effect of roads on number of coffee trees infested by CGS.

5.1.2 Socio economic studies methodology (coffee farming, farm budgets, yield loss, CGS knowledge, current controls and assessment of support services to farmers)

After extensive consultation between CABI, UNSW and CIC, it was decided that the socio-economic data would be best generated using a mixed-methods strategy, and training workshops were carried out with CIC staff. Data was then generated using the implementation of large-scale survey instruments, focus groups, case studies and research observations (including the use of digital photography[1]) to document baseline information about coffee production, grower awareness and techniques used to control CGS, pathways of CGS contamination and spread, and the capacity for growers/farmers (and constraints) to adopt 'best practices' to control CGS, once established. Reports were written after each stage of field work (see reports by Inu and Bofeng in bibliography)

The first large-scale survey (completed in collaboration with ASEM/2004/017) provided baseline information from over 500 growers, on coffee production and perceptions of

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[1] *A digital camcorder was purchased and used during fieldwork but was stolen from Kainantu before data was downloaded.*

insect infestations in the Eastern Highlands, Western Highlands and Simbu Provinces, in 2006 (Inu, 2006a). Smaller surveys of EHP (Inu, 2006b, 2007a) were administered to provide finer-grained details about farmer knowledge about CGS, its spread and impacts on yield. More details were provided about farm budget capacities and current controls of CGS and the role, if any, of chemical inputs.

Following the more targeted surveys, focus groups surveys, in-depth case studies (EHP), and spot surveys (WHP) were conducted to more accurately determine actual coffee crop losses to CGS (Inu, 2007b,c,d,e). In-depth case studies were carried out in 2008 in Bena Bena and Lufa villages in Henganofi (EHP) (Inu, 2008a,b). These locations were selected for a range of reasons, which included accessibility and safety for the research team. The case studies were conducted in two sites with 11 farmers in each site (22 farmers) with blocks of coffee gardens with up to 500 trees. Data was collected on: revenue generating activities, expenditure activities, labour rates and cherry harvesting.

In 2009, educational posters about CGS and ways to manage it were produced and distributed, and feedback was sought from farmers about their efficacy (Bofeng, 2009). Farmers reported their appreciation for the details -- those with access to posters expressed that they had become better informed about CGS. Farmers reported that they would like to see more extension work, and training. Farmers expressed their need to have more input from CIC: more access to CIC staff would mean that farmers could consult on issues such as how to maintained quality, pest and disease problems, latest research findings, information on markets, and any other relevant issues that affect coffee production. This, they remarked, will help to motivate them to work on their coffee gardens and produce more coffee (Bofeng, 2009).

5.1.3 Studies on potential factors contributing to CGS outbreaks *(*and thus factors that can be exploited in development of controls)

The main ecological factors that might influence CGS population outbreaks were analysed at the project inception workshop in October 2006 and then in the preliminary and major ecological surveys. These factors, which are discussed below, include:

- Human factors
- Local natural enemies and ants
- Shade levels over coffee
- Nutrients

Human factors

A ‘scoping study’ was conducted in November/December 2007 on the sites that were identified as high risk zones, with high levels of infestation in new coffee gardens, during the March/April (2007) survey (Inu, 2007e).

A main aim of the scoping study was to identify pathways of CGS spread, which included identifying the source of coffee seedlings, and the level of grower knowledge about CGS (for identification purposes during sourcing).

The scoping study was conducted using semi-structured interview techniques with household members (including extended family members who were present at the time of interview). Such households typically included a family of 5–8 members (12–15 including extended family members) and the interviews were conducted in Tok Pisin.

The latest approach to extension was to work with growers who had organised themselves into groups of at least 20 households. The interviews were conducted with members of these groups.

It must be noted that to get a good response rate to any such ethnographic research (be it surveys, focus groups, interviews, etc.) required a lot of trust building. The research team found that matters of livelihood were considered sensitive and private. The research team particularly noted that the success in gathering information reflects the good relationships established already between the growers and the Extension Division of CIC. The farmer assessments of the Extension support, during that four-week interview period carried out in the Eastern Highlands, was positive.

The results were collated to generate both quantitative data, and qualitative descriptions of pathways.

Ant exclusion experiments 1 to 4

Ants are thought to provide both sanitation and protective services to CGS (Bach, 1991; Stadler & Dixon, 2008). The exclusion of ants may reduce scale levels by either: (i) halting the removal of excess honeydew by ants thus increasing the mortality of crawlers – the sanitation hypothesis; (ii) allowing natural enemies access to scales and thus increasing predation – the protection hypothesis. For logistical reasons, i.e. staffing and time required in the field, four staggered experiments were set up to test the overall hypothesis that ants enhance CGS infestations.

Experiment 1:

To assess the impact of ants on CGS infestation levels a total of 20 young coffee trees, circa two years old, and 20 mature coffee trees were randomly selected within a non shaded coffee block. These trees were then split into two treatment groups, control and ant exclusion. The control allowed ants access to the trees, and the barrier method (Tanglefoot) was used in the ant exclusion treatment. The barrier method was essentially the same as in previous works in other coffee growing countries, e.g. Hawaii, (Reimer, Cope & Yasuda, 1993). Any differences in scale numbers would therefore be due to either the presence or absence of ants. Regular recordings were conducted from 14th December 2007 to 31st March 2008. Recordings included assessing levels of live adult scale, parasitised scale, *L. lecanii*-infested scale, abundance of ladybirds (coccinellids), number of green cherry and number of internodes.

All the data was subjected to statistical testing using the repeated measures function in Genstat 11 within the REML or ANOVA package. A full description can be found in appendix 11.4.

Experiment 2:

This was essentially the same as experiment 1 but run from 22nd April 2008 to 28th August 2008. A full description can be found in appendix 11.5.

Experiment 3:

This was essentially the same as experiments 1 and 2 but to assess two methods of ant control on CGS infestation levels. A total of 30 trees were selected within a non shaded coffee block. These trees were then split into two treatment groups and a control. The control allowed ants access to the trees, and the barrier method (Tanglefoot) was used as one of the ant exclusion treatments. Tanglefoot may restrict some natural enemies access as it acts as a barrier around the basal part of the tree and can also act as a trap. The second treatment consisted of the ant bait Amdro which does not restrict natural enemies access to the tree. This, however, is an insecticide and would therefore not be appropriate as a potential control method in organic certified coffee. It was just used experimentally. Regular recordings were conducted from 9th October 2008 up to 22nd December 2008. A full description can be found in appendix 11.6.

Experiment 4:

The previous experiments showed that the removal of ants significantly reduces the abundance of CGS on coffee trees compared to trees where ants have access to CGS. The findings strongly suggest that ants play a crucial role in CGS infestations as found by Bach (1991). Thus an experiment was set up to investigate the impact of specific natural enemies on CGS abundance, as it was still unclear what roles specific natural enemies have on these infestation levels, e.g. is any reduction due to ladybirds or parasites, or a combination? The main two questions addressed in this experiment were:

1. What is the effectiveness of parasitoid natural enemies in the presence / absence of ants?
2. What is the effectiveness of the larger coccinellid (ladybird) natural enemy predators in the presence / absence of ants?

The experiment was set up at CIC Aiyura (Block 248) to both assess the two methods of ant control on CGS on coffee and determine the impact of the respective natural enemies on CGS abundance. In total 120 coffee trees were selected and split into ten blocks of 12 trees and treatments were randomly assigned within each block (see appendix 11.7; Figure 11.7.1).

As in previous experiments the control allowed ants access to the trees, and the barrier method (Tanglefoot) was used to exclude ants. In contrast to previous experiments, 2mm mesh sleeves were used in order to exclude coccinellids on branches on half the trees. This would allow for changes in CGS abundance to be directly related to either parasitism or fungal infection. Additionally, half the treatments also had artificial shade applied (70% shade), a shade method often used in nurseries.

The experiment was set up in October 2008 in order to coincide with increasing CGS abundance, and initial recordings of CGS, natural enemy, and ant abundance were taken. However, subsequent recordings by CIC did not start until 1st May 2009, and ended 22nd September 2009. This is after peak abundance and when CGS populations appear to naturally decline. Recordings were taken as in previous experiments.

CGS infestation of coffee and the role of ants

Anecdotal field observations have indicated that ants transport the early stages of CGS. The mutualistic relationship between ants and CGS may encompass the movement by ants of CGS to different parts of the plants, or new plants. As yet there have been limited studies into the role that ants play in facilitating the process of CGS infestations via transportation. The question is: can ants facilitate CGS infestations of coffee seedlings or can they merely enhance them?

Plants were chosen from the CIC nursery representing clean plants, infested plants without ants, and infested plants with ants. These were placed within a sealed mesh cage and clean plants were paired with CGS plants either with ants or without ants, i.e. two plants per cage. A wooden bridge was then placed between the top green shoots of each plant potentially allowing CGS and ants access to the clean plant.

During monitoring the presence of CGS on the clean plants was recorded and any CGS found on clean plants were counted and then removed. This removal allowed the rate of infestation from the infested plants to be determined.

In total three species of invasive ant were used separately, these were: *Technomyrmex albipes* (white footed ant), *Pheidole megacephala* (African big-headed ant) and *Anoplolepis gracilipes* (yellow crazy ant). Monitoring of CGS and ants took place at regular intervals. A full description can be found in appendix 11.8.

Impact of artificial shade on CGS abundance

In March 2008 a manipulative shade study was planned, and started in late 2008, at CIC Aiyura. The aim was to assess the influence of artificial shade levels on the development of CGS on seedlings, mimicking the variety of artificial shade found at nurseries. Simple wooden frames were constructed to allow netting of different shade levels to be applied over coffee seedlings whilst allowing natural enemies access. Three levels of shade were used, 0% shade, 50% shade and 70% shade.

CGS infested plants were acquired from the CIC nursery and randomly allocated the three shade levels. Cages were laid out in a randomised design near the CIC Aiyura laboratory to avoid interference from local people. Each cage was placed at least 1m apart from the neighbouring cage in order to limit shading of one treatment by another. Plant height (cm) and CGS abundance measurements were collected approximately every two weeks for 204 days. A full description can be found in appendix 11.9.

Soil nutrients and CGS abundance

In October 2007 it was planned to use an existing field-based fertiliser trial to examine the influence of soil fertility on CGS infestations. However, as the previous history of the treatments used in experimental fields was uncertain, a small-scale study in a greenhouse was undertaken. Primarily the aim was to investigate the effects of soil nutrients on CGS on coffee seedlings. Trials of differing fertiliser regimes on coffee seedlings and the responses of CGS to the soil nutrients were examined.

Within the greenhouse coffee seedlings were assembled into replicated treatment blocks. Three levels of nutrients were tested using sulphate of ammonia (SOA), where the standard field rate used is 80 kg/ha (N Simbiken, pers. comm.). Each block contained a control (no additional nutrients) and two treatments with SOA at 40 kg/ha and 80 kg/ha.

Infestations were developed on plants untreated with insecticides or fertilisers grown and then inoculated with field reared CGS. CGS populations were assessed prior to treatments being imposed, to establish baseline data. After treatment imposition, CGS infestations were assessed on a regular basis from June 2008 to July 2009. A full description can be found in appendix 11.10.

5.2 Objective 2

5.2.1 Studies on classical biological control

The aim in the project was to import available exotic encyrtid parasitoids (Hymenoptera) from Australia (used for green scale control on citrus) and switch them from citrus to coffee in the quarantine facility. Emphasis was put on the parasitoid, *Diversinervus stramineus.* This species originates from Kenya and was introduced by DPI, Queensland to Australia in 1996 to control green scale in citrus. The laboratory studies done in Australia on the biology of *D. stramineus* and the specificity test on 16 non-target species of green scales found that the parasitoid is host specific to *C. viridis*, although other CGS were not tested in these early studies.

The main areas addressed in the project were: improving rearing facilities at R&GSD Aiyura and Bena Bena; addressing staffing needs and training for biological control; importation of exotic parasitoids; rearing and assessment of impact of these on CGS; and planning for release and trial in coffee farms.

5.2.2 Studies on the development of a control package

The combination of socio-economic surveys and ecological studies should provide evidence of what factors are important in the development of a control package. A package would need to take account of socio-economic constraints and agronomic practices as well as the ecology of the CGS problem. Therefore there was a need to draw on the outputs of these studies. The key points were:

- Determine the level of understanding and awareness by farmers of the CGS problem.
- Determine any existing measures (from the socio-economic studies) and what should be built on (especially those supported by on-station research).
- Determine the presence of any local natural enemies that have a significant impact on CGS.

- Determine the role of environmental / management factors, as well as the presence of problematic invasive ants, in the CGS problem.
- Determine whether control of invasive ants (e.g. by banding) is acceptable and affordable to farmers.
- Reassess classical biological control technology: how effective might it be, and how much of a problem would these agents be in competing with local natural enemies.
- Account for local constraints when developing a control package / management plan.

These aspects were to be assessed in a few villages in EHP to enable a ‘best practice’ management package to be developed and validated.

5.3 Objective 3

This objective covered two aspects: the development of a ‘best practice’ management plan that is based on the results of the validation studies on the control package (section 5.2.2); and the development of a draft strategy for the implementation of the control package across the main coffee producing provinces.

5.3.1 Draft management plan for current best practice

This was undertaken by staff of CIC. As of January 2010 a technical advisory circular on CGS control methods and how to intervene on CGS pathways to reduce the spread is currently being developed. Plans are also underway to produce a DVD on CGS for distribution in coffee producing villages.

5.3.2 Draft strategy document for short, medium and long term management

In July 2009 a meeting was held in PNG between CIC, Dr Murphy and Dr Shaw. The focus of this meeting was to develop a framework for a strategy document for the short- to long-term management of CGS. The outline of this can be seen in section 7.3.2.

6 Achievements against activities and outputs/milestones

Objective 1: To document baseline information on the distribution, impacts, biology and control of CGS in the coffee growing zones of PNG

No.	Outputs/ milestones	What has been achieved?	What has not been achieved?	Are there additional outputs that could have been achieved?
1.1	Survey the incidence & impacts of CGS and their associated insects. Distribution maps, graphs & database of CGS related to key variables.	Both preliminary and major ecological surveys were completed by CIC within the EHP providing data for relating CGS infestations with key variables. This provided data for distribution maps. CIC were trained in sampling methods, the routine collection of core data was enabled and the distribution of CGS clarified.		It would have been good to repeat this survey on an annual basis to monitor fluctuations in CGS populations and relate this to changes in management. However, this would have been quite difficult logistically.
	Classification and abundance of key associated species.	Ant species diversity, abundance, distribution and associations with CGS survey completed. A list of ant species and their associations with CGS is now available; exotic invasive ant species have been identified as a major problem. The role of ants and natural enemies refined. More trees were infested near roads: human mediated spread(?)		
	Quantification of effect of ants and other agents on CGS abundance and coffee yields	To some extent preliminary and major ecological surveys address this. Removal of ants significantly reduces CGS infestations - 4 studies completed which allows for the quantification of the effect of ants on CGS abundance.	In the preliminary and major ecological surveys it was not possible due to logistics and time constraints to identify the actual species of ants present at the individual farms surveyed. However, other studies (ant diversity study) have shown CGS to be strongly associated with invasive exotic ants.	By identifying the actual ant species at specific farms a targeted approach to ant control and further assessments of the practicalities of ant control could have been assessed. Additionally, yield loss could have been addressed.

			The 4th study on ant exclusion was a large experiment whereby more information on the role of specific natural enemies was to be measured. It was timed for peak occurrence of CGS but was unfortunately delayed and did not occur until the CGS populations were in natural seasonal decline. This study was only recently completed.	This would have gleaned further information on the impacts of specific natural enemies i.e. by excluding coccinellids and including parasitoids, within combinations of ant control.
			Unfortunately, assessing coffee yields with inclusion / exclusion of ants was not achieved. The reason for this is that the studies did not cover the entire growing season.	It would have been better to set up trials from the outset of the project and follow them all the way through to the end. Thus providing information on yield loss. Unfortunately it proved very difficult to retain experimental sites for long periods of time.
		Shade and soil nutrient studies also completed. The data has only recently been made available.		Apart from the ecological surveys and ant diversity study, it was not possible to invoke a more detailed survey of shade i.e. introduced shade species vs. indigenous shade. Habitat quality / disturbance may play a role in CGS outbreaks.

1.2	Survey of current control methods by growers, including the rationale for such measures Current control practices evaluated in relation to production costs Farmers' constraints & perceptions itemised and evaluated Support services to farmers evaluated with future recommendations	Using a range of ethnographic data-generating techniques: A thorough assessment of current control practices, and evaluation in relation to cost (2006-9); Assessment of constraints (economic and social), and Assessment of support services evaluated. Farmers could not quantify the impact of CGS because of a lack of knowledge about its existence, presence and potential impacts on coffee plants. Since implementation of the first socio-economic survey instruments, work with extension has provided farmers with a greater understanding of the problem. The following socio-economic surveys completed: 2006: Preliminary CGS 2006: Major Coffee Post Harvest (ASEM2004/017) which provided base-line details for CGS, Eastern Province, Western Province and Simbu 2007: Major CGS with ASEM/2004/017 survey 2007: Scoping for CGS case study 2008: Spot survey for CGS 2008: Case Studies 2009: 'feedback' from farmers' survey completed.	An adequate understanding of yield loss due to CGS. Overall yield losses can be estimated but loss due to CGS remains unclear.	Farmers lacked knowledge of CGS which makes it difficult to quantify the full economic costs of CGS.

Objective 2: To evaluate biological and other control methods of CGS, taking into account grower information

No.	Outputs/ milestones	What has been achieved?	What has not been achieved?	Are there additional outputs that could have been achieved?
2.1	Evaluation of release and establishment of available biocontrol agents and a range of existing and novel control practices	The quarantine facility in Aiyura has been dramatically improved. Parasitoid, *Diversinervus stramineus*, effectively raised (on Citrus, Gardenia and coffee) and successful with *Coccus viridis*	Parasitoids have not been effectively switched to *C. celatus*, being the more common highland species. Release of *D. stramineus* not recommended because natural predators already effective on *C. viridis*.	Switching a parasitoid effectively to eradicate *C. celatus*.
2.2	Training for technicians and other professionals on rearing, release and evaluation methodologies	CIC staff, including recent recruits, have received significant training in all areas of the project, in both scientific and social scientific methodologies		With more efficient use of time (and communications) training in the production of DVD training documentaries would have been useful.

Objective 3: To develop regional and national strategies for wider evaluation and implementation of CGS control

No.	Outputs/ milestones	What has been achieved?	What has not been achieved?	Are there additional outputs that could have been achieved?
3.1	Draft strategy document for short, medium and long term CGS management	In process (CIC Aiyura)	Draft management strategy and roll-out is underway but not yet complete	
3.2	Draft management plan for current best practice consistent with socio-economic conditions and sustainable certification schemes	In process (CIC Aiyura)	Sustainable certification schemes – probably not achievable within current project	More effective communications with CIC Aiyura would have meant more efficient progress on outputs such as certification

7 Key results and discussion

7.1 Objective 1

7.1.1 Distribution studies

Preliminary ecological survey

The diagonal transect method, relating the trees assessed for CGS presence / absence on a diagonal walk, to the total proportion of the trees on a farm infested with CGS was significantly positively correlated. This indicated that the diagonal transect method was reasonably reliable. There were also significant positive correlations between number of trees infested with CGS and both ants and the entomopathogen, *Lecanicillium lecanii*.

The effects of environmental factors (e.g. altitude and shade level) were then analysed using the same method. Here a strong negative correlation with altitude and shade levels were found, i.e. the higher the altitude the lower the shade level reflecting the distribution of forestation with altitude. There were no other significant correlations found but further analysis was conducted using the environmental variables incorporated into a general linear model. See appendix 11.1 for full details.

From Table 7.1.1 it can be seen that altitude is a significant driving factor on the number of CGS infested trees. However, this analysis does not take into account other variables measured e.g. ants which have a close relationship with CGS.

Table 7.1.1: Results of the Generalized Linear Mixed Model on CGS and environmental variables.

	Wald Chi-Square	Df	Significance
Altitude	14.113	5	.015
Shade Level	*11.833*	*7*	*.106*
Total Trees (Covariable)	*1.600*	*1*	*.206*
No. Trees Assessed (Covariable)	11.694	1	.001
Cells in italics are the terms deleted from the main effects model. Non italicised = final terms in the model.			

An additional analysis using the ants as a covariable in addition to tree abundance was conducted (Table 7.1.2). Here, it can be clearly seen that ant presence, tested in the main effects model, was a significant driving factor and using ants as a covariable makes shade and altitude non-significant.

Table 7.1.2: Results of the Generalized Linear Mixed Model on CGS and environmental variables with the presence of ants as a covariable.

	Wald Chi-Square	Df	Significance
Altitude	8.230	5	.144
Shade Level	7.580	7	.371
Total Trees (Covariable)	*.034*	*1*	*.853*
No. Trees Assessed (Covariable)	*.263*	*1*	*.608*
No. with Ants (Covariable)	15.921	1	.000
Cells in italics are the terms deleted from the main effects model. Non italicised = final terms in the model.			

Further ecological studies were required in order to investigate these findings. A full major ecological survey was conducted in November 2007 surveying well over 128 farms versus the 47 in this preliminary survey. This was in order to enhance the power of the surveys.

Major ecological survey

The first step in this major survey was to compare both the diagonal transect (D) and whole coffee block survey methods (W). This diagonal survey method was reliable in comparison to the whole block survey method as all measures were significantly correlated. The next step used the same method to look for simple relationships between the variables surveyed, e.g. relationships between CGS and ant abundance. Strong positive correlations between CGS and: ants, *Lecanicillium* sp. and ladybirds were found.

Significant positive correlations were found with farm size and tree abundance which would be expected because large farms have more trees. However, there were no relationships with the proportion of trees infested with CGS and: altitude, farm size and age. Shade was the only significant correlation and was negative indicating that with increasing levels of shade, the proportion of CGS infested trees declines. See appendix 11.2 for full details.

A more detailed analysis using the Generalized Linear Model (GENLIN) function in SPSS (version 16) was then used. Number of trees with CGS was used as the response variable and altitude, age, garden site, farm size and shade were used as factors. Number of trees was entered as a covariable in the model in order to take account of the differing number of trees surveyed at each farm. All non-significant factors were sequentially removed to simplify the model. Table 7.1.3 shows the final results.

Table 7.1.3: Results of the Generalized Linear Mixed Model for Dcgs / Wcgs presence / absence and environmental variables. With number of trees surveyed as a covariable.

	Tests of Model Effects Dcgs Type III			Tests of Model Effects Wcgs Type III		
	Wald Chi-Square	df	Sig.	Wald Chi-Square	df	Sig.
Altitude	34.860	5	.000	70.260	5	.000
Farm Size	12.899	2	.002	20.571	2	.000
Shade	9.470	3	.024	13.531	3	.004
Age of Trees	*5.691*	*3*	*0.128*	16.225	2	.001
No. of Trees (Covariable)	2.402	1	0.121	3.610	1	0.057

In both data sets, altitude, farm size and shade consistently show significant effects. Most striking is the significantly higher infestations of CGS at 1500masl; above and below this, infestations are significantly lower (Figure 7.1.2). Farm size was also significant with large farms (>1000 trees) having greater infestations of CGS. However, this analysis did not take into account other variables measured e.g. ants which have a close relationship with CGS. An additional analysis using the ants as an additional covariable was conducted.

Table 7.1.4: Results of the Generalized Linear Mixed Model for Dcgs / Wcgs presence / absence and environmental variables. With number of trees surveyed and presence / absence of ants as a covariable.

	Tests of Model Effects Dcgs Type III			Tests of Model Effects Wcgs Type III		
	Wald Chi-Square	df	Sig.	Wald Chi-Square	df	Sig.
Altitude	*3.891*	*5*	*0.565*	*5.628*	*5*	*0.344*
Farm Size	*2.791*	*2*	*0.248*	7.515	2	.023
Shade	*0.693*	*3*	*0.875*	8.313	3	.040
Age of Trees	*0.865*	*3*	*0.834*	42.066	3	.000
No. of Trees (Covariable)	*1.769*	*1*	*0.183*	*0.113*	*1*	*0.737*
Ants (Covariable)	58.174	1	.000	47.232	1	.000
Cells in italics are the terms deleted from the main effects model. Non italicised = final terms in the model.						

Table 7.1.4 shows no significant differences in the factors recorded in the diagonal transect. However, there are three significant factors associated with CGS in the whole block coffee counts (using tree and ant abundance as covariables). It appears as though farm size, shade level and age of trees are the underlying factors. CGS infestations are highest where there are large farms (>1000 trees), where shade levels are very low and where farms consist of young and immature trees. Additionally, the ant covariable showed significant effects in both survey methods. The implications are discussed below.

Ants were then used as the response variable in order to investigate the influence of CGS on ants. Table 7.1.5 shows the results and here altitude and shade show significant differences in ant abundances in both survey methods (Figure 7.1.2 - Figure 7.1.3).

Table 7.1.5: Results of the Generalized Linear Mixed Model for ant presence / absence and environmental variables. With number of trees surveyed and presence / absence of CGS as a covariable.

	Tests of Model Effects Dcgs Type III			Tests of Model Effects Wcgs Type III		
	Wald Chi-Square	df	Sig.	Wald Chi-Square	Df	Sig.
Altitude	11.405	5	.044	17.740	5	.003
Farm Size	*4.156*	*2*	*.125*	3.965	2	0.38
Shade	17.305	3	.001	11.296	3	.010
Age of Trees	*2.485*	*3*	*0.478*	*0.326*	*3*	*0.955*
No. of Trees (Covariable)	*1.737*	*1*	*0.188*	*0.221*	*1*	*0.638*
Dcgs / Wcgs (Covariable)	41.997	1	.000	21.330	1	.000

Initially, factors influencing CGS appeared to be altitude, farm size and shade level. As ants are thought to provide protection of CGS from natural enemies it was deemed sensible to include their presence in the model as a covariable. When the presence of ants is included as a covariable, and thus removing their influences on CGS, most of the environmental variables in the diagonal transect have no significant effects on CGS presence. Only in the whole coffee block survey were shade, farm size and age of trees found to be significant.

However, conducting the same analysis on ants (i.e. ants as the response variable) reveals that altitude and shade are the significant factors for the presence of ants with CGS as a covariable. Thus the complex intertwined relationship between ants and CGS, and the infestations in the EHP of PNG appear to be influenced by altitude and shade levels. Where ants have favourable conditions CGS will thrive, i.e. where shade is low and at an altitude of 1500masl (Figure 7.1.1 – Figure 7.1.4). Also, the significant effect of shade on ants could be as a result of disturbance, i.e. farms with more shade could be seen as a less disturbed habitat and thus more resilient to invasive ants. This could be seen as human mediated spread.

Figure 7.1.1: Relationship between CGS and ant infestations – preliminary and major ecological surveys.

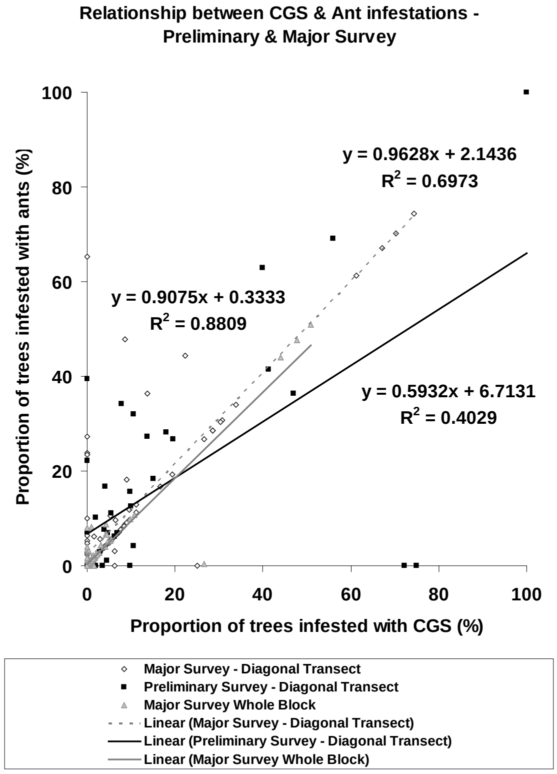


Figure 7.1.2: Relationship between CGS and ant infestations at differing altitudes – major ecological survey. All values are means +/- 1 s.e.

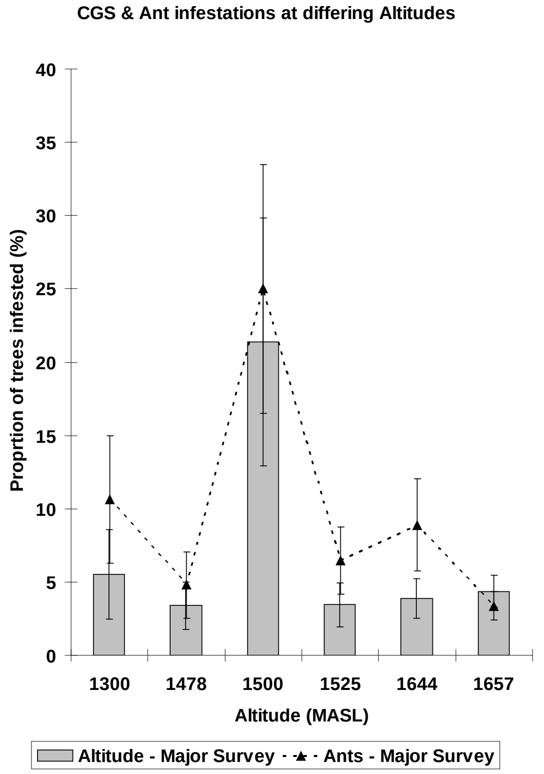


Figure 7.1.3: Relationship between CGS and ant infestations at differing levels of shade – major ecological survey. All values are means +/- 1 s.e.

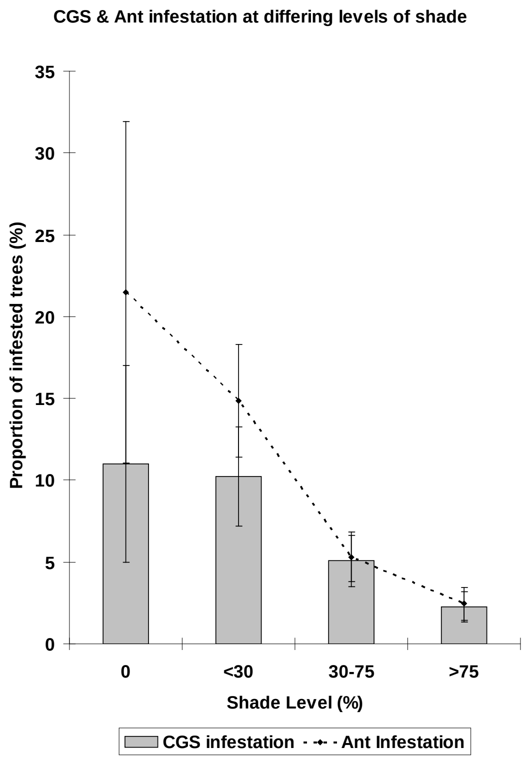
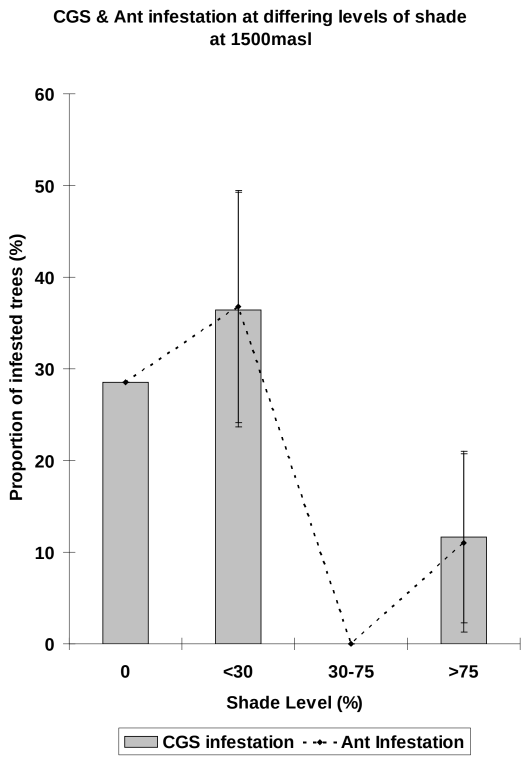


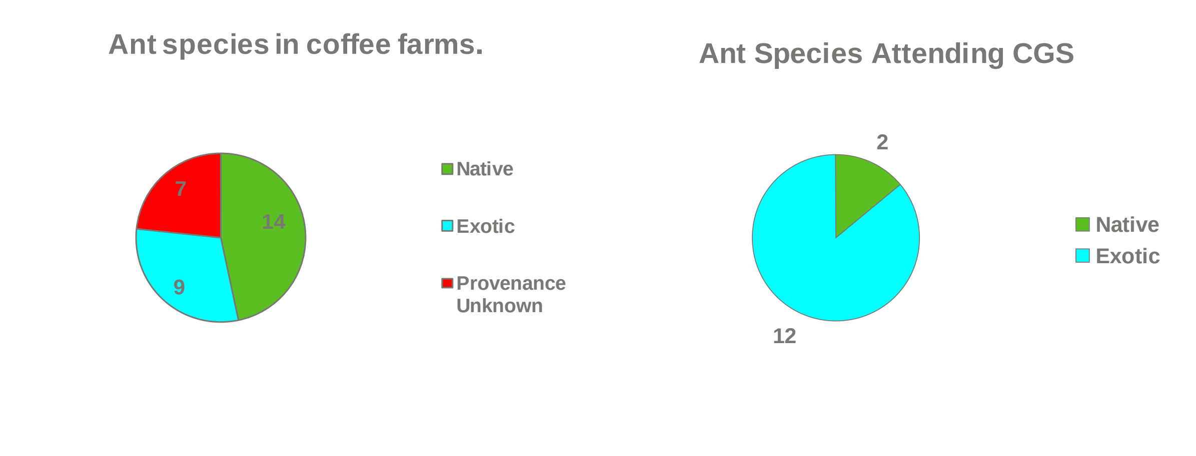
Figure 7.1.4: Relationship between CGS and ant infestations at differing levels of shade at 1500masl – major ecological survey. All values are means +/- 1 s.e.



Ant diversity survey

Species diversity on four coffee farms in EHP was examined together with the effects of ant–CGS association on coffee trees. Ant species diversity varied among the four farms with estimated number of species not exceeding 25 per farm. Combining all the farms resulted in 30 ant species being found. Of these 14 were native, nine were exotic and seven were ant species whose origin is unknown. Of the total number of ant species 14 species attended CGS and of these only two were native (*Rhoptromyrmex* sp1 and *Rhoptromyrmex melleus*). By contrast six exotics and six species (origin unknown) attended CGS. See Figure 7.1.5 below.

Figure 7.1.5: Number of native and exotic ant species found in Eastern Highlands Province of PNG and number of ant species attending CGS.



Although canopy cover and size of shade trees did not have an effect on ant diversity in all the coffee farms, other variables such as litter layer, pruning, use of chemicals and land-use history are perhaps responsible for the difference among the farms and low diversity (Sagata, 2008). An increase in intensity of management can decrease ant species richness in coffee farms (Philpott, Perfecto & Vandermeer, 2006). Also, the presence of invasive ants such as *Technomyrmex albipes* (white footed ant), *Pheidole megacephala* (African big-headed ant) and *Anoplolepis gracilipes* (yellow crazy ant) can potentially displace less competitive ants because invasive ants are known to displace native ant fauna and invertebrates (Ward, 1987; Holway et al., 2002; Abbott & Green, 2007).

The high abundances of *A. gracilipes* and *P. megacephala* observed were perhaps from single colonies because these ants are known to consist of spatially separated super-colonies consisting of thousands of workers (Holway, 1999; Holway et al., 2002). Where they invade, *A. gracilipes* and *P. megacephala* use their numerical abundance and aggressive behaviour resulting in the displacement of native and less aggressive ants (Hoffmann, Andersen & Hill, 1999; Abbott, 2006). The three spatially most common and abundant exotic species were *A. gracilipes*, *P. megacephala* and *T. albipes*.

The survey suggested that these exotic and abundant ants are tracking resources on a spatial structure (e.g. honeydew from CGS). This is not surprising because the tracking of resources is a common phenomenon in invasive ants (Abbott & Green, 2007). Domination of both space and resources using numerical strength can interfere with natural enemies of CGS such as ladybird beetles (Coccinellidae) or biological controls. For example, during the survey *A. gracilipes* and *P. megacephala* were observed “scaring away” spiders, and attacking coccinellids and native ants that came to parts of the coffee tree infested with CGS.

7.1.2 Studies on coffee farming, budgets, impact (yield loss, etc.) and knowledge of CGS

The initial socio-economic surveys (Inu, 2006a,b, 2007a) indicated that coffee farmers rely on participatory extension for information about CGS. The lack of record-keeping by farmers on production levels and incomes, and the lack of farmer knowledge about CGS, especially amongst smallholders, meant that it was impossible to determine yield loss due to CGS at this stage.

The impact of CGS on yield was also difficult to determine in the 2007 survey as the same issues of lack of record-keeping prevailed (Inu, 2007a). The case studies and spot surveys, in 2008 (Inu, 2008a,b), produced data which suggests that the impact can be high (Extension Officers estimate up to 50%). In-depth case studies revealed that household income is varied (estimated at between K159.00 to K979 per annum) and that the methods used to control CGS were usually low or no-cost because of the need for cash for other household commitments. The main expenditures for coffee growers are minimal as little time is spent managing coffee properly. Although school fees must be paid, compensation, bride and death costs are the main expenses (with death costs estimated at 45% of household expenditure).

Case study results (Inu, 2008b) show that although awareness of CGS has been raised since the 2007 surveys, there was still a negligible level of CGS control implemented by farmers, and no change in yield details (it is clear that records will not be kept). By 2008, CGS was still not regarded as a priority by farmers as long as coffee trees were producing cherry. Income details were presented anecdotally, rather than through record-keeping. These details indicated that in Bena Bena, 91% of household revenue came from activities other than coffee. However in Lufa, 45% of income was estimated to come from coffee sales. Other sources of income included: sale of chickens and livestock, and vegetable production. The researchers decided that participants were unwilling to record details about coffee yield or sales, preferring to refer to other income streams. Growers did state that parchment was the main coffee product sold (2007 survey).

Studies on potential factors contributing to CGS outbreaks

The 2007 survey (Inu, 2007a) indicated that for growers who noticed CGS, it was in the advanced stages of infestation, when present on young branches or shoots. Although 63% of growers identified CGS as a problem (but not a priority to control), approximately half of these (32%) associated the presence of ants – rather than the less obvious CGS – as the problem. The remedies used therefore focussed on the eradication of ants, including hand removal (59% of growers used this method) while 6% tried other methods (see below). The remainder of growers did not try to eradicate the problem, with approximately half stating that they could not afford remedies, such as chemicals, and the other half believing that their lack of knowledge rendered them helpless. On the spread of CGS, more than half the growers blamed “poor management” (externally).

By 2008, progress on CGS awareness had been slow (Inu, 2008a,b). The case studies confirmed that even with knowledge about CGS – most farmers were able to describe CGS, which demonstrated some progress in CGS education/monitoring – farmers were still not fully aware of its potential to dramatically reduce crop yields, and therefore income (note: research station observations show that CGS can reduce yields but what is unknown is the extent of the impact especially on farms). Most farmers did indicate a willingness to take up CGS control techniques if they were low cost and used minimum human-hour input so as not to conflict with time needed for family vegetable gardens and actual coffee production activities. Educational posters were distributed in late 2009, which did prove to be a valuable tool in distinguishing CGS from ants. The pressing issue for all growers was coffee theft (Extension Officers are acutely aware of this) (Bofeng 2009).

The socio-economic surveys revealed that although farmers were first briefed about CGS by CRI (Coffee Research Institute) in 1994, knowledge and educational material remain limited. Extension Officers provide information during the Participatory Rural Appraisal Program (PRAP), and will use this avenue to provide updated details on 'best practice', and implementation of the National Strategy (see section 7.3.1).

A 2007 scoping study was conducted in high risk zones where the main route of CGS spread had been previously identified as via nurseries. Smallholder coffee trees were planted from seedlings from the Department of Primary Industries (DPI) in the 1970s and 1980s. By the 1990s most seedlings came from CRI (Aiyura) and other DPI established nurseries. On the continued spread of CGS, human mediated and natural pathways appear to have been involved. Nurseries have grown and supplied seedlings infested with CGS. Spread also occurs within and between farms where farmers propagate their own seedlings from infested plants. This was corroborated in the 2008 case studies.

The 2007 survey identified that CGS was widespread (near 100% infestation rate of farms in EHP), with 60% of growers identifying an insect problem.

Ant exclusion experiments 1 to 4

Experiment 1

There is plenty of evidence for CGS reducing yield but in this experiment no differences were found, and here the experiment centred primarily on removing ants and thus scale. This experiment was conducted for four months and is likely to have been too short to detect differences in yield. Whilst there were no significant reductions in CGS levels in terms of the day × treatment interaction, as a result of the ant exclusion method, it is clear that there were significant differences in the proportion of healthy scale populations over time for mature coffee trees. The data does suggest that increased non-healthy scale levels, e.g. parasitised by species such as *Metaphycus baruensis* or entomopathogenic fungi such as *Lecanicillium* sp*.* might be the cause, again this is not clear-cut. See appendix 11.4 for full details.

Experiment 2

There were significant reductions in CGS levels (Figure 7.1.6) and in the proportion of healthy scale populations over time as a result of the ant exclusion method (Figure 7.1.7) compared to the control (Figure 7.1.8). This suggests that the there are natural enemies present which, if ants are controlled, can control CGS. It is not fully clear what is responsible for this control but the data does suggest that increased parasitism by *Metaphycus baruensis* or entomopathogenic fungi such as *Lecanicillium* sp. might be the cause. It is also possible that the highly mobile coccinellids may also be responsible but these, to some extent, may be restricted by the Tanglefoot barrier method. See appendix 11.5 for full details.

Figure 7.1.6: Mean CGS infestation level per tree over time (log n+1). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

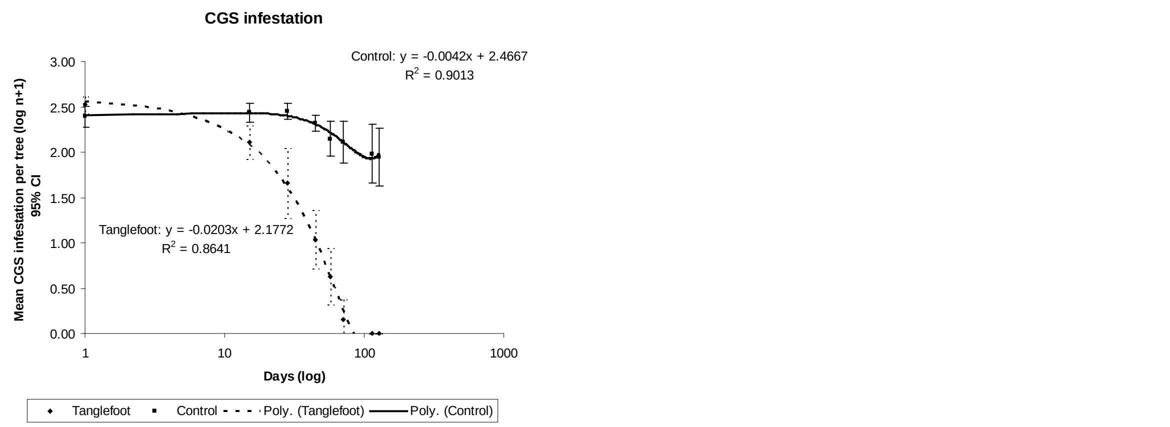


Figure 7.1.7: Mean proportion of non-healthy CGS where ants are absent. Non-healthy scale is defined as scale that is parasitised or infected with *Lecanicillium* sp. over time. All values are means with 95% confidence interval error bars.

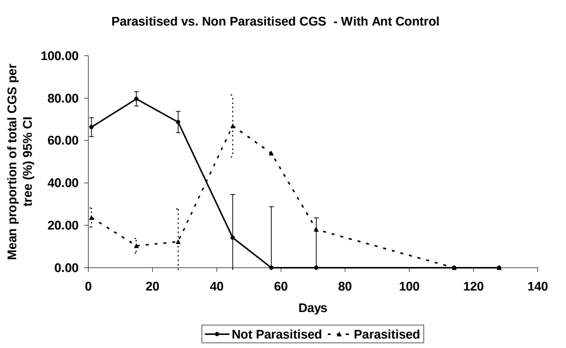
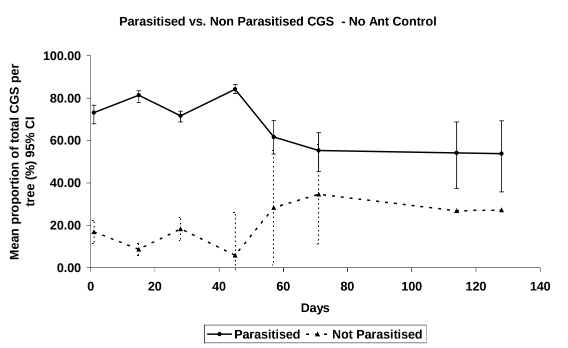


Figure 7.1.8: Mean proportion of healthy CGS where ants are present. Healthy scale is defined as scale that is neither parasitised nor infected with *Lecanicillium* sp. over time. All values are means with 95% confidence interval error bars.



Experiment 3:

Again there were significant reductions in CGS levels and in the proportion of healthy scale populations over time as a result of the two ant exclusion methods. This suggests that the there are again natural enemies present which, if ants are controlled, can control CGS. See appendix 11.6 for full details.

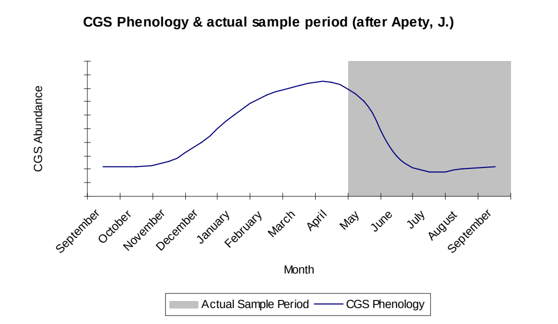
Experiment 4:

In this experiment it appears as though there are no differences between CGS abundance on the ant excluded and ant present trees. This goes against previous findings where ant exclusion successfully reduces CGS abundance or at the very least increases the proportion of non-healthy CGS. In order to understand these results possible causes were investigated which included: (i) Had Amdro reduced ants within the experimental field to such an extent that CGS populations declined on both ant excluded and ant present treatments and (ii) Had there been a natural decline in CGS abundance, greater than that of the effect of excluding ants.

In previous studies the exclusion of ants increased the proportion of non-healthy over that of healthy CGS, either parasitised or fungus infected. This was again plotted for this experiment, and if Amdro had reduced ants to such an extent in the whole field then this pattern would be expected throughout the treatments, including the control. This pattern did not exist so possibility (i) was excluded.

Data on CGS phenology, supplied by CIC, suggests the peak period of CGS abundance is April. A natural decline in CGS abundance could be the cause and, as stated in the methods section, the original planned timing of the experiment was to coincide with increasing CGS abundance and include the peak period. However, the experiment was delayed by some six months, starting in April and ending in September (Figure 7.1.9). It is very probable that this was the cause of the inconsistent findings. The results from coccinellid exclusion and shade elements also showed the same patterns. See appendix 11.7 for full details.

Figure 7.1.9: CGS phenology throughout the year. Data supplied by CIC, recreated after Apety, J.



CGS infestation of coffee and the role of ants

All three invasive ant species tested had no significant impact on the rate of spread from infested to clean plants suggesting that CGS disperses by crawling (at the first instar stage) or is wind blown. Transportation of CGS as noted in the field may be occurring but at such low levels that it does not enhance infestation rates. As we know from the ant exclusion experiments, ants do reduce the mortality of CGS and are therefore more likely to be indirectly, rather than directly, enhancing the spread of CGS. This enhancement is likely to be via protecting them from natural enemies. This implies that the presence of the invasive exotic ant species benefits CGS by protecting them from natural enemies but they do not appear to actively transport CGS. As a consequence the spread of these species may enhance CGS survival and thus indirectly encourage the spread of CGS. See appendix 11.8 for details.

Impact of artificial shade on CGS abundance

Plant growth over time was not significantly affected by increased shade levels. However, there were initial differences in height between the shade treatments (Figure 7.1.10). Figure 7.1.11 shows CGS abundance in the differing shade levels and it can be seen that CGS levels are higher where artificial shade is present, i.e. increasing levels of shade increase CGS abundance. However, the artificial shade used was a fine mesh and would have acted as a barrier to natural enemies and thus enhanced CGS infestations. See appendix 11.9 for details.

Figure 7.1.10: Coffee seedling growth under different levels of artificial shade: control = no shade, 50% shade, 70% shade. Growth was measured by using plant height (cm). Data was log transformed and displayed with mean values with estimated standard errors.

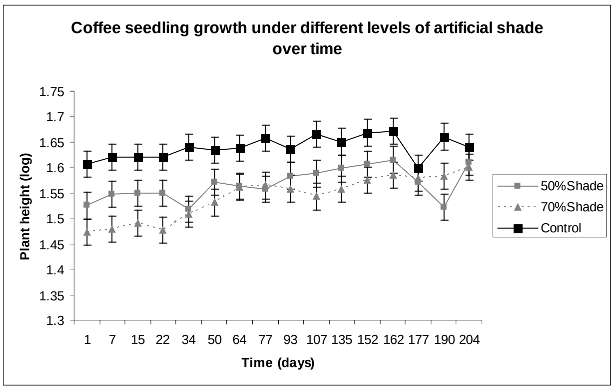
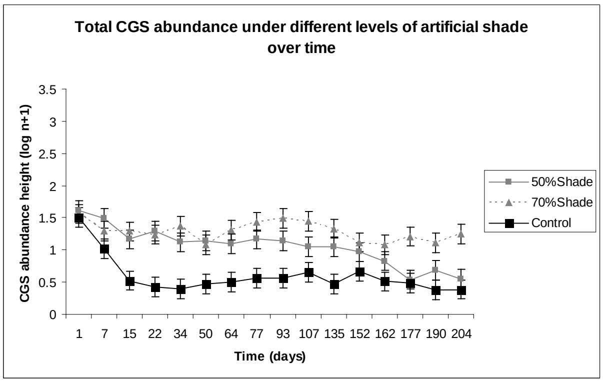


Figure 7.1.11: CGS abundance under different levels of artificial shade: control = no shade, 50% shade, 70% shade. Total CGS abundance was measured by counting all scales present. Data was log n+1 transformed and displayed with mean values with estimated standard errors.



Soil nutrients and CGS abundance

In the greenhouse experiment, plant growth, as measured by plant height, was not significantly increased by the addition of SOA (sulphate of ammonia) over time. There were no significant differences in height between the SOA treatments. In this experiment it does not appear that soil nutrients are important in CGS development (see Figure 11.10.2; appendix 11.10). However, the plants did not appear to grow over the period of 427 days which is a questionable finding. It would be expected for plants to grow over such a length of time. These results should be treated with caution. For details see appendix 11.10.

7.2 Objective 2

7.2.1 Studies on classical biological control

In November 2006 CIC imported 200 *Diversinervus stramineus* from DPI, Queensland and attempted to culture these at the facilities at Aiyura but the culture failed. It was recommended by CABI in early 2007 that attention needed to be given to developing the facilities in PNG for rearing and to ensure that a sustainable culture of CGS on citrus and coffee was available before any more importations were attempted.

Plans were made improve facilities and other aspects related to the operation and to develop a ‘citrus system’ to be ready in September 2007. But this was not achieved. Thus the situation was again reviewed in October 2007 and a series of possible causes were identified, e.g. lack of human resources, and problems with the facilities such as temperature control, problems with light, lack of cages and contamination problems. During 2008 two consultancies were made to address these issues. A brief account of what was done is given below.

Improving rearing facilities at the CIC Bena Bena and Aiyura

Citrus plants are grown at Bena Bena and coffee plants at Aiyura. CIC attempted growing CGS on citrus in poly-tunnels at Bena Bena in order to maintain a citrus scale population. Temperature fluctuations have been reduced by replacing the ceiling with black mesh allowing rain in and heat out. However, this facility was not entirely secure with a real potential for allowing predators in. Additional security features have been implemented such as the use of purpose built cages, construction of benches and a moat. These facilities are important for this project and will add to the capacity of CIC.

The quarantine facility at Aiyura comprises of three rooms; high quarantine, medium quarantine and a large rearing area. As this facility would be the receiving unit for biocontrol agents a thorough review of its status was carried out. Temperature, lighting and security were all identified as potential problems.

Significant progress had been made but persistent problems of inadequate light and overheating were evident. A significant amount of strip lighting was also installed, suspended from the ceilings by chains, and is thus adjustable. The result is an air-conditioned room that can accommodate large aluminium cages with adequate light and temperature control. Also, structural and procedural improvements have addressed this, e.g. all the rooms and doors have been sealed and standard operating procedures imposed.

Addressing staffing needs and training for biological control

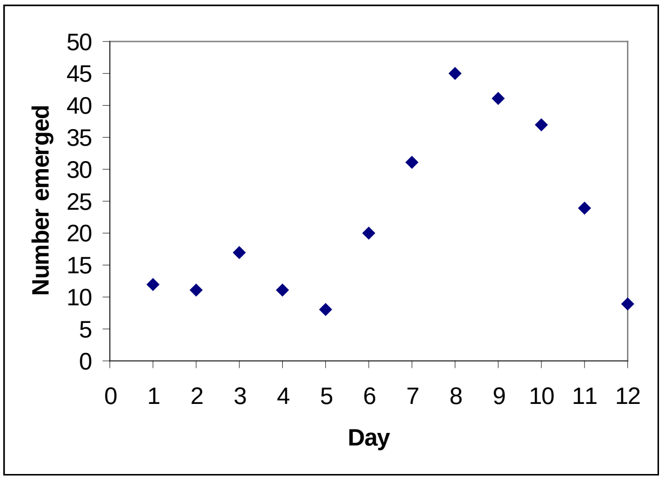
Staffing issues became a concern during meetings in 2007. It was felt that there was a need for more staff in order to meet the objectives of the project. Job descriptions for posts within CIC were discussed and general guidance was given in order to provide 'terms of reference'. Since then, four high quality staff have been put in place on the project which addresses this issue.

The importation of exotic parasitoids

The permit to import *D. stramineus* from Australia was approved by the Department of Environment and Conservation and the National Agriculture Quarantine & Inspection Authority (NAQIA) in 2008. After a visit to the Queensland Department of Lands rearing facility in Nambour, a collection of emerged adult *D. stramineus* and parasitised scales on leaves of citrus and gardenia was made. This material comprised a shipment of 400 parasitised scales which was hand-carried by R. Shaw and A. Brook to Aiyura via Port Moresby (where it cleared quarantine) and Goroka.

Upon arrival 45 adult *D. stramineus* were still alive along with a further 12 which had emerged en-route. The emergence over the following days is summarised below (Figure 7.2.1)

Figure 7.2.1: Plot of adult *D. stramineus* emergence over time.



The rearing and assessment of impact of D. stramineus on CGS

Diversinervus stramineus was cultured on several potted green scale (*C. viridis*) host plants: Arabica and Robusta coffee, citrus, gardenia, and ixora in the quarantine facility. The daily room temperature was between 23°C to 25°C. Table 7.2.1 shows the number of *C. viridis* parasitised by *D. stramineus* and the number of parasitoid emerged from various host plants.

Table 7.2.1: Culturing of *D. stramineus* on *C. viridis* on various host plants.

Generation No.	Host plant	No. of *C. viridis* parasitised	No. of *D. stramineus* emerged
1	Arabica	645	235
	Citrus	12	5
	Gardenia	709	221
	Robusta	367	115
	Ixora	70	9
Total		1,803	585

By the end of November 2008, more than 1000 *D. stramineus* had been produced. However, despite the use of insect screens and separate plant stocks at Bena Bena, the scale on plants sourced from there was heavily parasitised. The team were trained in techniques to produce clean plants; the contaminants (*Coccophagus* sp. and other local parasites) have affected the number of parasitoids reared in the quarantine facility and reduced the number down to around 100. Measures were undertaken to clean up the contaminated cultures after recommendations were made by a visiting scientist from CABI. By the end of February 2009, more than 200 parasitoids had been produced. Currently, the parasitoids are being monitored daily and new host plants with cleaned cultures are being used to try building up their numbers again. The host plants *Coffea rabica* and *C. cenaphora* are being used.

Since April 2009 a culture of *D. stramineus* has been maintained on *Coccus viridis* on coffee. A number of trials have been set up to try and switch the parasitoid onto *C. celatus* but all of these have failed, so in June of 2009 a decision was made to halt this work and focus on the utilising the potential of *D. stramineus* as a control agent for *C. viridis*.

The planning for release and trial in coffee farms

Following the rearing and impact work, plans were discussed and agreed for the trial release of *D. stamineus* in PNG. The first stage involves the drafting of a dossier on the parasitoid that includes information about the benefits and risks of releasing the parasitoid. This needs to be sent out for consultation with key stakeholders, coordinated by NAQIA.

As of 2010 the dossier was still being prepared by CIC and will then be submitted to PNG Department of Environment and Conservation for approval.

7.2.2 Development and validation studies of a management package

The management package included raising awareness of CGS with the use of talks, posters and information videos. It aimed at promoting local controls by enhancing natural enemies and shade, and at the same time trialling the use of ant barriers (e.g. Tanglefoot) on smallholder farms. Along with this was the need to determine which smallholder income levels could afford such a control method. Additionally, extension workers were included in the package.

In order to validate the management package, ‘before and after’ surveys were planned in a small number of villages in the EHP for August 2009. In September 2009 CIC conducted the surveys in four villages addressing these elements but as yet we have limited information. All that can be said is that based on the information collected, a poster on CGS was produced, in both English and Tok Pisin, and distributed to the farmers in order to address:

- Farmers’ limited knowledge on what CGS is,
- Natural enemies and CGS control methods,
- Decision making and what method to be used when CGS becomes a problem.

A follow up survey was conducted in October 2009 to assess the impact of the poster, but again we have no further information. It appears as though the control methods were not trialled, or at least have not been reported. All of this has major implications for the draft management and strategy document (section 7.3.1 and 7.3.2).

7.3 Objective 3

7.3.1 Draft management plan for current best practice

The draft management plan depends on the validation studies proposed in section 7.2.2. As this has not been completed all that can be reported is that a technical advisory circular on control methods and how to reduce the spread of CGS is currently being developed. A request for information on where CGS is in other provinces of PNG has also been sent to CIC officers. Additionally the production of a DVD on CGS was being discussed, but may not be produced until mid 2010.

7.3.2 Draft strategy document for short, medium and long term management

What was planned

The draft management strategy framework was set out at a meeting in PNG in July 2009. Here the importance of validating any management package was highlighted (section 7.2.2). From this the strategy could then be integrated into CIC's strategic plan for coffee development, i.e. Papua New Guinea Coffee Industry Corporation Strategic Plan 2008 – 2018 (see CIC, 2008).

The current state of the strategy document

As discussed in the previous sections (7.2.2 onwards) the validation has not been reported but nevertheless CIC have produced a draft strategy. This proposed strategy document has the central aim of improving farm productivity of smallholders through sustainable management of CGS. The roll out programme has three main outputs, shown in Table 7.3.1.

Table 7.3.1: Main outputs from proposed CIC draft management strategy. Simbiken (2009). "Roll out of Coffee Green Scale Management Strategy". CIC Aiyura, PNG.

Output 1:	Develop CGS awareness material Conduct informal farmer training Conduct training of "farmer trainers"
Output 2:	Develop awareness of planting material and supply of clean seedlings Establish central nursery programme for distribution of healthy seedlings
Output 3:	Develop CGS training modules for formal and informal education for coffee farmers

Management schemes for a rollout programme

CGS management strategy requires productivity improvement of smallholder farms through farmer capacity building and the creation of an enabling environment. There are three main categories of smallholders (Simbiken, 2009), these are: low, moderate and high input husbandry. Thus the management schemes will be tailored to each of these smallholder categories.

The roll out of the management practices would require working with the three categories of smallholder farmers because the level of engagement is influenced by the ethnographic factors influencing their decision-making abilities. The CIC extension model of participatory training of group farmers will be used to create a learning scheme for management of CGS. It will require the development of training modules on CGS management and then on-farm training on selected farms. The training is aimed at improving farmers' knowledge and overall farm productivity.

Six-months programme

A smallholder rehabilitation programme has been established at two selected smallholder sites in the EHP. The farmers were grouped into three categories as discussed at a meeting in Aiyura in July 2009. A farm evaluation was conducted and then a rehabilitation plan developed with the participating farmer groups. CGS control measures were implemented in the rehabilitation exercise and community stewardship promoted through provision of field monitoring guides.

A central nursery has also been developed to aid in the distribution of clean seedlings to farmers.

Two-year programme

Similar rehabilitation and nursery programmes are extended to two other farmer groups in Simbu and Western Highlands Provinces.

Long-term programme

A rehabilitation programme will be built into the coffee growers support services programme and will provide on-going support to smallholder farmers in other provinces.

8 Impacts

8.1 Scientific impacts – now and in 5 years

No scientific impacts were achieved during the project but there are strong implications of the project finding that local natural enemies have a significant impact on CGS in the absence of ants. Thus agricultural organisations such as CIC should strongly consider putting more emphasis on conservation biological control rather than introducing agents for classical biological control. More likely to be achieved in five years.

8.2 Capacity impacts – now and in 5 years

Knowledge and skills of key staff in CIC in ecological assessment and biological control have been developed. Research staff at all levels (including Extension Officers) have been trained in a range of methodologies for generating socio-economic data.

Quarantine and laboratories for biological control work were developed considerably including the introduction of an operating procedure.

8.3 Community impacts – now and in 5 years

Awareness about CGS and its relation to ants has been raised in the project areas. This has been corroborated in a follow-up socio-economic survey in August 2009 (37 farmers).

8.3.1 Economic and social impacts

Prior to this project farmers had very limited knowledge, if any, about CGS. Since the project's commencement farmers have become aware of CGS and generally have a better understanding of the potential economic impacts. The impact of the roll out of information on 'best practice' using posters (in Tok Pisin) will need to be tracked in the future; recent documentation suggests that growers appreciate this input. The production of a DVD of 'best practice' would potentially reach more growers (which was emphasised in discussions with CIC).

Some of the farms with higher incidences of CGS replenished their trees from nursery stocks. These stocks have since been identified as a source of CGS and the nurseries have been informed of this. Smaller farms usually restocked from saplings reared from their existing trees, which may or may not be infested with CGS. The dissemination of 'best practice' and uptake of this knowledge needs to be tracked into the future.

The impact of the implementation of large-scale socio-economic survey instruments, and more intensive focus groups and case studies, is that farming communities are now much more aware of CGS, and also the distinction between CGS and the ants that attend CGS. Information gathered on financial capacity and priorities has reinforced the understandings that chemical inputs (insecticides) are not only unaffordable but undesirable for the current organic status of coffee production in the PNG highlands. Thus constrained, CGS research has continued to pursue alternatives to chemical controls. The dissemination of the results of cost-effective 'best practice' will undoubtedly have social and economic impacts through improved coffee quality, and potential for higher sales if CGS is effectively controlled. The issue of coffee theft, which is a major constraint to coffee production in PNG, is now widely recognised. The impact of the PNG government's recent ban on the sale of unprocessed cherry to roadside buyers will impact upon the overall desire for smallholders to grow coffee, or record sale information. In April 2009, CIC was reported in the media as urging coffee growers to urgently harvest and sell parchment (because of the cherry ban) to take advantage of high coffee prices (K3.80 per kilo) and to avoid coffee theft (http://www.thenational.com.pg/042009/biz8.php). The overall social impacts of CGS control uptake and its implications are particularly important to women who contribute more than 65% of coffee production. The extent to which coffee continues to be an important cash crop needs to be tracked into the future. The reliance on other cash-generating activities is indicative of the need to by-pass the impacts of coffee theft (an issue which goes beyond the scope of this project).

8.3.2 Environmental impacts

The presence of exotic invasive ant species has been identified as a major environmental impact particularly where biodiversity is concerned. These species compound the CGS problem and subsequently impact on the farming EHP community. According to the IUCN Invasive Species Specialist Group (ISSG), the yellow crazy ant and African big-headed ant have been nominated as among 100 of the world's most invasive species. They have been identified as major causes in the loss of local / indigenous biodiversity e.g. the African big-headed ant *Pheidole megacephala* (Andersen, Woinarski & Hoffmann, 2004; Hoffmann & Parr, 2008).

This project has highlighted the problem with invasive ants and by encouraging change in farmers behaviour, i.e. management, controlling the spread of these could have environmental benefits in the future.

8.3.3 Communication and dissemination activities

A national strategy of disseminating 'best practice' details has commenced with the roll out of information posters (in Tok Pisin), through extension. The production of an information DVD, using existing footage of farmers / growers in action, will provide another, more wide-ranging, dissemination strategy. This DVD will be made available for screening at local church halls throughout the coffee-growing highlands. Coffee producers who tend to attend church (particularly women) will be the targeted audience for DVD screenings.

9 Conclusions and recommendations

The major coffee green scale (CGS) problem in PNG seems to be limited altitudinally (to approx 1500masl) and the main species involved is *Coccus celatus*, a species that originates from East Africa. This is the case for all the major coffee growing provinces. Another species of CGS is present, *Coccus viridis*, also from East Africa but this species appears less common. The scales are mostly attended by ants and surveys have shown that the major species involved are also alien invasives, the major species being *Anoplolepis gracilipes*, *Pheidole megacephala* and *Technomyrmex albipes*.

The socio-economic surveys covered the three major coffee growing provinces. Smallholders generally grow a variety of crops although results showed that in general that food production is their main priority. Land is family owned and divided through an inheritance system but a proportion of the income from crops is given to family elders. For Eastern and Western Highland Provinces most farmers who produce coffee have been doing this for ten or more years; i.e. there are few young farmers entering into the system; but most coffee production is done by women (65%). When income from coffee is secured, it is mostly used to pay for school fees. The low priority placed on coffee is likely to contribute to the fact that, by and large, smallholders do not manage coffee very well and yield production is low on the smaller farms. The awareness and understanding of ants and CGS is also very limited among smallholders and a major barrier to uptake of proactive management. But some farmers have ‘experimented’ with control measures and these include shade management and the use of pumpkins to attract ladybird (Coccinellidae) species. A very important issue is that farmers are in general unaware of the production potential of their coffee crop and also the losses that may be being incurred. Losses that can be assigned to CGS attack are not known and needs urgent further study. Attempts were made to make estimates in this project but CIC have lacked the capacity to conduct studies on this to date. Lack of knowledge of this is a likely to be a major disincentive

Recommendation 1: A general study on coffee yield loss due to CGS is undertaken as a matter of priority.

A number of factors seem to be generating the population outbreaks of CGS. High shade is an important limiting factor of CGS infestations. Where shade is not used or has been removed (i.e. disturbed habitat) then infestations are higher. Also, and importantly, local natural enemies, in the absence of ants, are able to suppress CGS infestations down to insignificant levels. If the infestations are not entirely controlled the proportion of parasitised, non-healthy, CGS is increased. The natural enemies involved include parasitoids (mostly of the family Encyrtidae) and ladybird species. The continued movement of CGS through human mediated and natural pathways appears to have been a significant factor contributing to the spread of the problem. Nurseries have grown and supplied seedlings infested with CGS. Spread also occurs within and between farms and occurs where farmers propagate their own seedlings from infested plants; this was corroborated in the 2008 case studies. In the case of ants, human disturbance is a likely route of the spread, e.g. through highways and farm management practices. But ants do not appear to directly increase the spread of CGS; they appear to enhance CGS survival by inhibiting indigenous natural enemies which in turn increases the likelihood of survival and spread.

The project greatly increased CIC’s capacity to undertake classical biological control. CIC now have much improved rearing facilities and also protocols for the introduction, rearing and screening of biological control agents. One agent was introduced from Australia for CGS: the host specific encyrtid parasitoid, *Diversinervus stramineus*. A culture of this is now held by CIC at Aiyura but it has been found that it will only attack *C. viridis*. A major point that arose from the project studies was the observation that local natural enemies are able to have a significant impact on CGS in the absence of ants. Thus a question is raised about whether an exotic biological control agent would be any more efficient. The standard procedure to address this type of question falls under international protocols for conducing biological control. This procedure includes a production of a dossier on the CGS problem and the benefits and risks of introducing particular agents. This dossier is then used by relevant authorities to decide whether or not a release should be made.

***Recommendation 2: CIC to produce a dossier on the CGS problem and the benefits and risks of releasing* Diversinervus stramineus.**

Based on the results of the project studies, there are a number of key components that need to be included and validated in a management plan for CGS.

Recommendation 3: management plan components include:

Viable existing local controls, such as shade management, spraying soap or chilli onto CGS colonies and the conservation of local natural enemies; the last includes the use of pumpkins to attract ladybird species onto farms.

The testing of using ‘banding’ (e.g. through use of Tanglefoot) on coffee tree boles to prevent invasive ant species attending CGS colonies and driving away local natural enemies. The affordability of this for farmers in different income brackets would need to be investigated.

Development of awareness schemes to alert farming communities about the problem of CGS. This may include talks and posters, etc.

Classical biological control options, and also as a component of an integrated management plan, can be considered but a decision has to made by relevant authorities in PNG about the relative merits of introducing agents when good local natural enemies already exist. One factor might be whether an exotic agent can avoid attack by ants.

Recommendation 4: The management plan needs to be tested in a few coffee farming villages to measure suitability, cost, degree of uptake and impact on CGS.

This would best be done in EHP. Plans were agreed in August 2008 for this to be done but results are not available. Once this plan has been validated, it should be disseminated and tested in other coffee growing provinces.

It is clear that most Extension Officers working in CIC have little experience or knowledge of CGS and suitable management techniques. This is a major gap as these people provide advice and technical backstopping to farmers.

Recommendation 5: Extension Officers must be included in the management plan validation exercise.

Recommendation 6: Another important aspect is that the management plan needs to be institutionalised within CIC’s strategy on coffee development.

This needs to include geographical areas to be targeted and timelines for the role out of the strategy.

But all of this needs to be seen in the context of larger socio-economic issues: that the inheritance system might be a disincentive to invest in coffee; and that at the more macro level, there is also a lack of infrastructure and commercialisation initiatives necessary for the development of coffee.

10 References

10.1 References cited in report

Abbott, K.L. & Green, P.T. (2007) Collapse of an ant-scale mutualism in a rainforest on Christmas Island. Oikos, 116, 1238-1246.

Abbott, K.L. (2006) Spatial dynamics of supercolonies of the invasive yellow crazy ant, *Anoplolepis gracilipes*, on Christmas Island, Indian Ocean. Diversity and Distributions, 12(1), 101-10.

Andersen, A.N., Woinarski, J.C.Z. & Hoffmann, B.D. (2004) Biogeography of the ant fauna of the Tiwi Islands, in northern Australia's monsoonal tropics. Australian Journal of Zoology, 52(1), 97-110.

Bach, C.E. (1991) Direct and Indirect Interactions between Ants (*Pheidole megacephala*), Scales (*Coccus viridis*) and Plants (*Pluchea indica*). Oecologia, 87(2), 233-39.

Bofeng, I. (2009) Trip Report For Visit To Henganofi, Lufa, and Daulo Districts In Eastern Highlands Province, October 2009 (Asem/2004/0047), Research and Grower Services Division, Coffee Industry Corporation Ltd, Po Box 470, Ukarumpa, Eastern Highlands Province

Bolton, B. (1994) Identification guide to the ant genera of the world. Harvard University Press, Cambridge, Mass.

Buckley, R.C. (1987) Interactions involving plants, Homoptera, and ants. Annual Review of Ecology and Systematics, 18, 111-35.

Buckley, R. & Gullan, P. (1991) More aggressive ant species (Hymenoptera: Formicidae) provide better protection for soft scales and mealybugs (Homoptera: Coccidae, Pseudococcidae). Biotropica, 23(3), 282-86.

CIC (2008) Papua New Guinea Coffee Industry Corporation Strategic Plan 2008 – 2018. Coffee Industry Corporation Ltd. Goroka, Eastern Highlands Province, Papua New Guinea.

Colwell, R.K. & Coddington, J.A. (1994) Estimating terrestrial biodiversity through extrapolation. Philosophical Transactions of the Royal Society of London B Biological Sciences, 345, 101-118.

Crawley, M.J. (2005) Statistics: an introduction using R. John Wiley & Sons Ltd, Chichester.

Dytham, C. (1999). Choosing and using statistics: a biologist's guide., 1st edn. Blackwell Science Ltd., Oxford.

Elton, C.S. (1958) The ecology of invasions by animals and plants. Methuen, London.

Fleming, E. (2002) Analysing the opportunities and threats facing the coffee industry in Papua New Guinea toward the year 2020. Proceedings and recommendations of the national Coffee Workshop "Raising Production and Quality of PNG Coffee in the New Millennium. Papua New Guinea Coffee Corporation, Goroka, 22-24 April 2002, pp. 40-51*.*

Hanks, L.M. & Sadof, C.S. (1990) The effect of ants on nymphal survivorship of *Coccus viridis* (Homoptera, Coccidae). Biotropica, 22(2), 210-13.

Hoffmann, B.D., Andersen, A.N. & Hill, G.J.E. (1999) Impact of an introduced ant on native rain forest invertebrates: *Pheidole megacephala* in monsoonal Australia. Oecologia, 120(4), 595-604.

Hoffmann, B.D. & Parr, C.L. (2008) An invasion revisited: the African big-headed ant (Pheidole megacephala) in northern Australia. Biological Invasions, 10(7), 1171-81.

Holway, D.A. (1999) Competitive mechanisms underlying the displacement of native ants by the invasive Argentine ant. Ecology, 80(1), 238-51.

Holway, D.A., Lach, L., Suarez, A.V., Tsutsui, N.D. & Case, T.J. (2002) The causes and consequences of ant invasions. Annual Review of Ecology and Systematics, 33, 181-233.

Inu, S.M. (2006a) Assessment of Smallholder Post Harvest Technique, ASEM2004/017, Coffee Industry Corporation Ltd.

Inu, S.M. (2006b) Preliminary Scoping Survey of Coffee Green Scale, November 2006, Coffee Industry Corporation Ltd.

Inu, S.M. (2007a) Major Socio-Economic Survey Report, ASEM/2004/042 March/April 2007, Coffee Industry Corporation Ltd.

Inu, S.M. (2007b) Socio-Economic Survey Report, ASEM/2004/042 & PHT/2004/017, May 2007, Coffee Industry Corporation Ltd.

Inu, S.M. (2007c) Assessment of Smallholder Post Harvest Technique Project Report, November 2007, PNG PHT/2004/017, Coffee Industry Corporation Ltd.

Inu, S.M. (2007d) Socio-Economic Survey of Coffee Green Scale, Preliminary Report, PNG ASEM/2004/047, Coffee Industry Corporation Ltd.

Inu, S.M. (2007e) Scoping Study Report, ASEM /2004/042, November 2007

Inu, S.M. (2008a) Spot Survey Report ASEM/2004/042, January/February 2008

I Inu, S.M. (2008b) Socio-Economic Case Study Report in Eastern Highlands Province, Draft Report November 2008, ASEM/2004/047, Coffee Industry Corporation Ltd.

Kock, A.E.D. & Giliomee, J.H. (1989) A survey of the Argentine ant, *Iridomyrmex humilis* (Mayr), (Hymenoptera: Formicidae) in South African fynbos. Journal of the Entomological Society of Southern Africa, 52(1), 157-64.

Longino, J.T., Coddington, J. & Colwell, R.K. (2002) The ant fauna of a tropical rain forest: estimating species richness three different ways. Ecology, 83(3), 689-702.

Lowe, S., Brown, M. & Boudjelas, S. (2000) 100 of the world's worst invasive alien species. Aliens, 12, S1-S12.

Mack, R.N., Simberloff, D., Lonsdale, W.M., Evans, H., Clout, M. & Bazzaz, F.A. (2000) Biotic invasions: causes, epidemiology, global consequences, and control. Ecological Applications, 10(3), 689-710.

O'Dowd, D.J., Green, P.T. & Lake, P.S. (2003) Invasional 'meltdown' on an oceanic island. Ecology Letters, 6(9), 812-817.

Pedigo, L.P. & Buntin, G.D., eds. (1994) Handbook of sampling methods for arthropods in agriculture. CRC Press, Inc., Florida.

Philpott, S.M., Perfecto, I. & Vandermeer, J. (2006) Effects of management intensity and season on arboreal ant diversity and abundance in coffee agroecosystems. Biodiversity and Conservation, 15(1), 139-55.

Quinn, G.P. & Keough, M.J. (2002) Experimental design and data analysis for biologists. Cambridge University Press, Cambridge.

Reimer, N.J., Cope, M.L. & Yasuda, G. (1993) Interference of *Pheidole-Megacephala* (Hymenoptera, Formicidae) with biological control of *Coccus viridis* (Homoptera, Coccidae) in coffee. Environmental Entomology, 22(2), 483-88.

Sagata, K. (2008) Ant Species Diversity, Abundance and Distribution and their Association with Coffee Green Scales and Effect of this Association on Coffee in Eastern Highlands Province. Wildlife Conservation Society-PNG, Goroka, EHP, PNG.

Simbiken, N. (2009) "Roll out of Coffee Green Scale Management Strategy". CIC Aiyura, PNG.

SPSS (2008) SPSS for Windows. SPSS Inc., Chicago.

Stadler, B. & Dixon, T. (2008) Mutualism: Ants and their Insect Partners. Cambridge University Press.

Tilman, D. (1999) The ecological consequences of changes in biodiversity: A search for general principles. Ecology, 80(5), 1455-74.

Ward, D.F. & Harris, R.J. (2005) Invasibility of native habitats by Argentine ants, *Linepithema humile*, in New Zealand. New Zealand Journal of Ecology, 29(2), 215-19.

Ward, P.S. (1987) Distribution of the introduced Argentine ant (*Iridomyrmex humilis*) in natural habitats of the lower Sacramento valley and its effects on the indigenous ant fauna. Hilgardia, 55(2), 16 pp.

White, G.C. & Bennetts, R.E. (1996) Analysis of frequency count data using the negative binomial distribution. Ecology, 77(8), 2549-57.

10.2 List of publications produced by project

Publications

Shaw, W.S. Serendipitous Coffee Tourism in Papua New Guinea. In L. Jolliffe, Coffee Culture, Destinations and Tourism (Channel View Publications, forthcoming).

Shaw, W. S., Inu, S. & Smart, G. (2008) Ethnographic Geographies of Coffee Production in Papua New Guinea, INSULA: International Journal of Island Affairs (Dossier: Island Sustainable Development), 17(1), 13-22.

Theses

Smart, G.L. (2008) ‘Validity of Biocontrol of Coffee Green Scales in Papua New Guinea’, unpublished Honours thesis, University of New South Wales, Sydney, Australia.

Presentations

2007 Ethnographic Geographies of Coffee production in Papua New Guinea, Inaugural Commission on Islands – International Geographical Union conference, Taipei, Taiwan.

2008 Geographies of In/difference: whose geographies matter (ethnographic geographies of Coffee Green Scale in PNG), Royal Geographical Society and Institute of British Geographers conference, London, UK.

2009 The researcher as adventure-tourist: gadding about in the coffee-growing highlands (of Papua New Guinea), Institute of Australian Geographers, Cairns, Australia.Conclusions and recommendations

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11 Appendixes

11.1 Appendix A: Preliminary (pilot) ecological survey

Introduction

The first objective of this project is to document baseline information on the distribution, impacts and current control methods of CGS in the coffee growing zones of PNG. In November 2006 a preliminary survey of 47 farms were selected to cover a range of altitudes where Arabica coffee is typically grown in Eastern Highlands. This preliminary ecological survey assesses the distribution of CGS in the Eastern Highlands Province and gathered additional information on CGS insect pest ecology.

Methods

Smallholder farms were selected where Arabica coffee is commonly grown and to cover altitudes from low to high in EHP. A diagonal transect method was used for assessing CGS presence. In this method the surveyor walked diagonally across a coffee garden and counted CGS, ants, ladybirds and *Lecanicillium lecanii* on individual trees. Data was recorded as presence/absence. The measurements taken are shown in Table 11.1.1.

Table 11.1.1: Variables surveyed with diagonal transect method.	
Variable Surveyed:	**Notes:**
Coffee Tree Count.	
Coffee Green Scale Presence.	Pest of coffee.
Ant Presence.	Formicidae – Associated with CGS.
Ladybird Presence.	Coccinellidae – Natural enemy of CGS.
Lecanicillium lecanii Presence.	Entomopathogenic fungus – Natural enemy of CGS.
Altitude.	Several altitudinal zones surveyed.
Shade Level.	Four levels of shade; 0 shade; <30%; 30-75%; >75%.
Environmental / Farm variables surveyed with both methods.	
Variable Surveyed:	**Notes:**
Altitude.	Six altitudinal zones surveyed.
Shade Level.	Four levels of shade; 1=0 shade; 2=<30%; 3=30-75%; 4=>75%.

Simple analyses

The next step was to look for simple relationships between the variables surveyed, e.g. relationships between CGS and ant abundance. There appear to be strong correlations between the number of trees assessed during the diagonal transect survey and the total number of trees at a farm (Table 11.1.2), thus indicating that the method is reasonably reliable. Additionally, there are strong positive correlations between trees infested with CGS and both ants and *Lecanicillium*.

Environmental factors were then analysed using the same method. Table 11.1.3 clearly shows a strong negative correlation with altitude and shade level, i.e. the higher the altitude the lower the shade level. There were no other significant correlations found but further analysis was conducted using the Generalized Linear Model procedure in SPSS.

Table 11.1.2: Spearman Rank (rho) correlations between the respective variables measured.

		Total No. of Trees	No. Trees Assessed	No. Trees with CGS	No. with Ants	No. with *Lecanicillium*
No. Trees Assessed	Correlation Coefficient	.354*				
	Sig. (2-tailed)	0.01				
No. Trees with CGS	Correlation Coefficient	0.07	0.14			
	Sig. (2-tailed)	0.64	0.34			
No. with Ants	Correlation Coefficient	0.06	0.14	.606**		
	Sig. (2-tailed)	0.70	0.36	0.00		
No. with *Lecanicillium*	Correlation Coefficient	0.25	0.26	.444**	.553**	
	Sig. (2-tailed)	0.09	0.08	0.00	0.00	
No. with Ladybirds	Correlation Coefficient	0.02	0.16	0.29	.486**	.545**
	Sig. (2-tailed)	0.90	0.29	0.05	0.00	0.00

** Correlation is significant at the 0.01 level (2-tailed). * Correlation is significant at the 0.05 level (2-tailed).

Table 11.1.3: Spearman Rank (rho) correlations between the environmental variables and CGS, ants and natural enemies.

		Altitude	Shade Level	% CGS infested	% Ant Infested	% Vert Infested	% Ladybirds
Altitude	Correlation Coefficient	1.000	-.466**	.117	.008	.040	.083
	Sig. (2-tailed)	.	.001	.434	.959	.787	.580
Shade Level	Correlation Coefficient			-.275	-.051	-.032	.021
	Sig. (2-tailed)			.061	.735	.831	.886

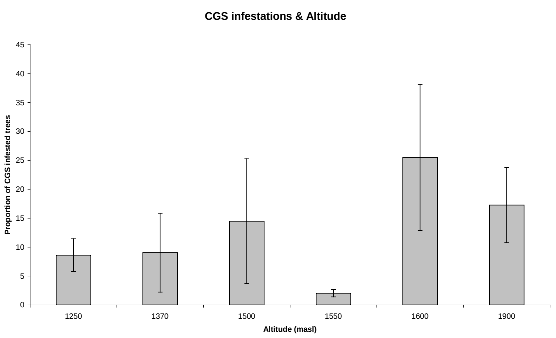
** Correlation is significant at the 0.01 level (2-tailed). * Correlation is significant at the 0.05 level (2-tailed).

In-depth analyses

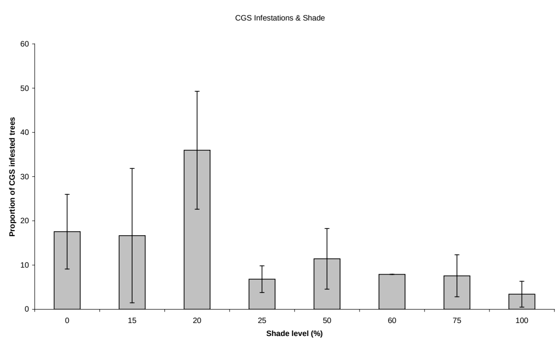
The data was then used to examine the relationships of CGS infestations and the environmental and farm variables measured. A simple method of examining the data is to plot the means with standard errors (+/- 1 s.e.). Figure 11.1.1 shows the means (+/- 1 s.e.) for CGS and environmental / farm variables.

Figure 11.1.1: Mean percent CGS infestations and environmental / farm variables.

Mean percent CGS infestation and altitude (+/- 1 s.e.).



Mean percent CGS infestation and shade level (+/- 1 s.e.).



A measure of significance is required if any meaningful conclusions are to be drawn. Simple descriptive statistics and frequency distribution plots of the data for the two methods were generated and fitted with a normal distribution curve. The data is skewed with a high degree of kurtosis, a positively skewed unimodal distribution. Applying transformations such as log n+1; arcsin and fourth root, made no improvement. As the data represents counts (discrete data) a Poisson type distribution might be expected. However, a Poisson distributions variance/mean ratio equals one, but here the variances are greater than the mean (White & Bennetts, 1996; Quinn & Keough, 2002; Crawley, 2005). The data therefore resembles a negative binomial distribution with a high degree of clumping ($k \leq 1$) as determined by

$$k \approx \frac{\mu^2}{s^2 - \mu}$$

where k measures the degree of aggregation; s is the variance and μ is the mean (Crawley, 2005).

For the analysis the Generalized Linear Model (GENLIN) function in SPSS (version 16) was used. This allowed for the distribution of the dependent variable to be specified as a negative binomial distribution with a log-link function (SPSS, 2008). CGS abundance was used as the response variable and altitude and shade were entered as factors. Additionally, number of trees assessed and total number of trees at a farm was entered as a covariate in this model. All non-significant factors were sequentially removed to simplify the model. Table 11.1.4 shows the final results.

Table 11.1.4: Results of the Generalized Linear Mixed Model on CGS and environmental variables.

	Wald Chi-Square	df	Significance
(Intercept)	1.543	1	.214
Altitude	14.113	5	.015
Shade Level	*11.833*	*7*	*.106*
Total Trees (Covariable)	*1.600*	*1*	*.206*
No. Trees Assessed (Covariable)	11.694	1	.001

Cells in italics are the terms deleted from the main effects model. Non italicised = final terms in the model.

From Table 11.1.4 it can be seen that altitude is a significant driving factor on the number of CGS infested trees. However, this analysis does not take into account other variables measured e.g. ants which have a close relationship with CGS. An additional analysis using the ants as a covariable in addition to tree abundance was conducted (Table 11.1.5). Here, it can be clearly seen that ant abundance tested in the main effects model was the significant driving factor and not shade or altitude.

Table 11.1.5: Results of the Generalized Linear Mixed Model on CGS and environmental variables with the presence of ants as a covariable.

	Wald Chi-Square	df	Significance
(Intercept)	.061	1	.804
Altitude	8.230	5	.144
Shade Level	7.580	7	.371
Total Trees (Covariable)	*.034*	*1*	*.853*
No. Trees Assessed (Covariable)	.263	1	.608
No. with Ants (Covariable)	15.921	1	.000

Cells in italics are the terms deleted from the main effects model. Non italicised = final terms in the model.

Further ecological studies were required to in order to investigate these findings. A full major ecological survey was conducted in November 2007 surveying well over 100 farms.

11.2 Appendix B: Major ecological survey

Introduction

In November 2006 a preliminary survey of 47 farms was conducted in the EHP using a rapid method utilising a diagonal transect across a farm. It was found that the high incidences of CGS were correlated with low levels of shade and an altitude of circa 1500masl. It was also thought that ants maybe promoting the proliferation of CGS by harassing natural enemies associated with CGS. Shade may also be a sink for natural enemies and thus beneficial in reducing CGS levels. However, a more detailed analysis revealed that these factors were not the significant driving factors, but, it appeared to be the presence of ants.

In order to clarify the findings and confirm the accuracy of the diagonal transect method a second survey was conducted in 2007. This was conducted on 128 farms using two survey methods; diagonal transect survey (as before) and a whole block survey.

A diagonal transect walk across the farm was made and every tree encountered was assessed for presence / absence of CGS, ants and predators. The whole block survey included every tree. Table 11.2.1 show the measurements taken.

Table 11.2.1: Variables surveyed with both diagonal transect method and whole coffee block method.

Variable Surveyed:	**Notes:**
Coffee Tree Count.	
Coffee Green Scale Presence.	Pest of coffee.
Ant Presence.	Formicidae – Associated with CGS.
Ladybird Presence.	Coccinellidae – Natural enemy of CGS.
Lecanicillium lecanii Presence.	Entomopathogenic fungus – Natural enemy of CGS.
Altitude.	Several altitudinal zones surveyed.
Shade Level.	Four levels of shade; 0 shade; <30%; 30-75%; >75%.
Farm Size.	Three groups, based of farm size; <500 trees; 500-1000 trees; >1000 trees.
Coffee Age.	Age of coffee trees; young/immature; mature; mix of both.
Garden Site.	Aspect and topographical position; east facing slope; west facing slope; valley; hill; valley and hill.

Simple correlative analyses

The first step in this major survey is to compare both the whole farm diagonal transect (D) and whole coffee block methods (W). A simple correlative procedure using the bivariate procedure Spearman's rank (rho) was used to establish relationships between the two methods. This allows for the analysis of data that might not be normally distributed (Dytham, 1999). It measures how rank orders are related and can be combined with an additional test of significance (two-tailed). Table 11.2.2 clearly shows significant positive correlations between the two methods for all variables.

Table 11.2.2: Spearman Rank (rho) correlations between the two survey methods – a test of the reliability of the diagonal transect method. D = diagonal transect, W = whole coffee block survey method.

	Wtree (N)	Wcgs (%)	Wants (%)	WLbirds (%)	WVertic (%)
Dtree (N)	.535**				
Dcgs (%)		.717**			
Dants (%)			.788**		
DLbirds (%)				.613**	
DVertic (%)					.597**

* Correlation is significant at the 0.05 level (2-tailed); ** Correlation is significant at the 0.01 level (2-tailed).

The next step used the same method to look for simple relationships between the variables surveyed, e.g. relationships between CGS and ant abundance. Table 11.2.3 shows the results for the diagonal and whole block surveys respectively.

Table 11.2.3: Spearman Rank (rho) correlations between and the respective variables measured using the diagonal transect method (D) & whole block count method (W).

	Dtree (N)	Dcgs (%)	Dants (%)	DLbirds (%)
Dcgs (%)	-.039			
Dants (%)	-.071	.786**		
DLbirds (%)	-.026	.276**	.443**	
DVertic (%)	.049	.609**	.502**	.373**
Whole block coffee count method (W).				
	Wtree (N)	Wcgs (%)	Wants (%)	WLbirds (%)
Wcgs (%)	-.095			
Wants (%)	-.170	.795**		
WLbirds (%)	-.128	.496**	.664**	
WVertic (%)	.070	.711**	.603**	.508**
* Correlation is significant at the 0.05 level (2-tailed); ** Correlation is significant at the 0.01 level (2-tailed).				

There appear to be strong positive correlations between CGS and; ants, *Lecanicillium* sp. and ladybirds. Conversely, there is no relationship between the proportion of CGS infested trees and tree abundance. The presence of CGS on the trees may be better explained by other biological, environmental or human mediated factors. The results of these variables and factor correlations are shown in Table 11.2.4.

Table 11.2.4: Spearman Rank (rho) correlations between variables and environmental factors measured from the diagonal transect method (D) & whole block count method (W).

	Altitude	Farm Size	Shade	Age of Trees	Garden Site
Dtree (N)	.178*	.551**	.164	.092	.136
Dcgs (%)	.106	.077	-.242**	.098	-.197*
Dants (%)	-.021	-.027	-.347**	.029	-.259**
DLbirds (%)	-.208**	-.017	-.149	-.063	-.238**
DVertic (%)	-.068	.105	-.176*	-.086	-.051
Whole block coffee count method (W).					
Wtree (N)	-.054	.761**	.356**	.065	.213*
Wcgs (%)	-.059	-.007	-.272**	-.136	-.140
Wants (%)	-.159	-.095	-.322**	-.084	-.209*
WLbirds (%)	-.220**	-.125	-.303**	-.070	-.182*
WVertic (%)	-.246**	.096	-.118	-.119	-.039
* Correlation is significant at the 0.05 level (2-tailed); ** Correlation is significant at the 0.01 level (2-tailed).					

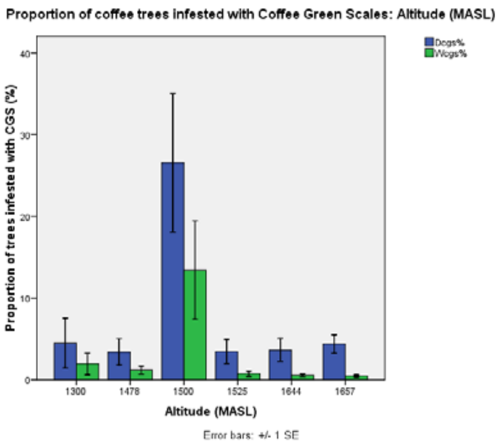
Table 11.2.4 shows a significant positive relationship with farm size and tree abundance which would be expected because large farms have more trees. However, there are no relationships with the proportion of trees infested with CGS and; altitude, farm size and age. There is a significant relationship with the proportion of infested trees and shade. This negative relationship indicates that with increasing levels of shade the proportion of CGS infested trees declines.

In-depth analyses

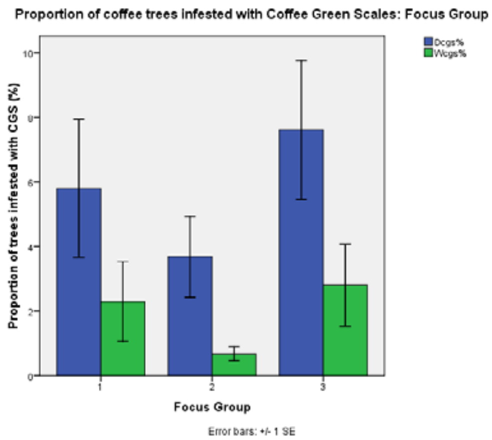
The data was then used to examine the relationships of CGS infestations and the environmental and farm variables measured. A simple method of examining the data is to plot the means with standard errors (+/- 1 s.e.). Figure 11.2.1 shows the means (+/- 1 s.e.) for proportion of trees infested with CGS and environmental / farm variables.

Figure 11.2.1: Mean percentage CGS infestations and environmental / farm variables. Infestation levels are based on the average number of trees infested with CGS per farm.

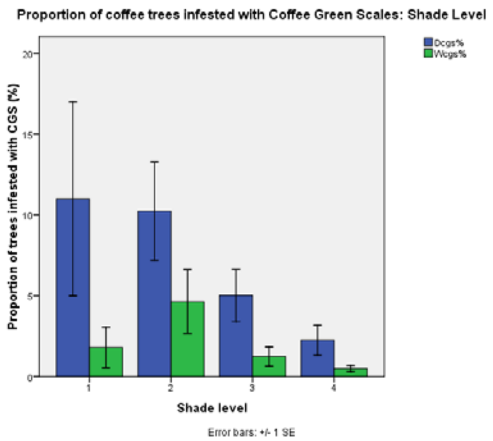
a) Altitude.



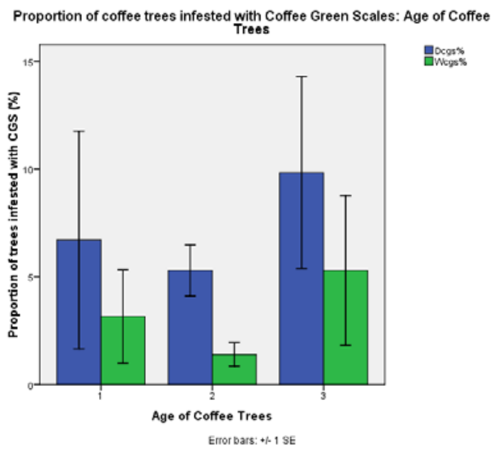
b) Farm Size (=Focus Group).



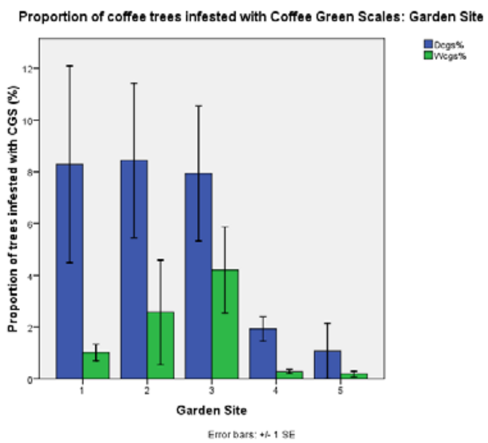
c) Shade.



d) Age of Trees.



e) Garden Site.



Further analysis used the Generalized Linear Model (GENLIN) function in SPSS (version 16) was used. As the CGS data was presence / absence data and resembled a negative binomial distribution the dependent variable (CGS) was specified as a negative binomial distribution with a log-link function (SPSS, 2008). Number of trees with CGS was used as the response variable and altitude, age, garden site, farm size and shade were used as factors. Number of trees was entered as a covariable in the model in order to take account of the differing number of trees surveyed at each farm. All non-significant factors were sequentially removed to simplify the model, table 11.2.5 shows the final results.

Table 11.2.5: Results of the Generalized Linear Mixed Model for Dcgs / Wcgs presence / absence and environmental variables. With number of trees surveyed as a covariable.

	Tests of Model Effects Dcgs Type III			Tests of Model Effects Wcgs Type III		
	Wald Chi-Square	df	Sig.	Wald Chi-Square	df	Sig.
(Intercept)	9.429	1	.002	42.191	1	.000
Altitude	34.860	5	.000	70.260	5	.000
Farm Size	12.899	2	.002	20.571	2	.000
Shade	9.470	3	.024	13.531	3	.004
Age of Trees	*5.691*	*3*	*0.128*	16.225	2	.001
No. of Trees (Covariable)	2.402	1	0.121	3.610	1	0.057

In both data sets, altitude, farm size and shade consistently show significant effects. The most striking is the significantly higher infestations of CGS at 1500m, above and below this, infestations are significantly lower (Figure 11.2.1). Farm size was also significant with large farms (>1000 trees) having greater infestations of CGS. However, this analysis does not take into account other variables measured e.g. ants which have a close relationship with CGS. An additional analysis using the ants as an additional covariable was conducted (Table 11.2.6).

Table 11.2.6 shows no significant differences in the factors recorded in the diagonal transect. However, there are three significant factors associated with CGS in the whole block coffee counts (using tree and ant abundance as covariables). It appears as though farm size, shade level and age of trees are the underlying factors. CGS infestations are highest where there are large farms (>1000 trees), where shade levels are very low and where farms consist of young and immature trees. Additionally, the ant covariable showed significant effects in both survey methods.

Table 11.2.6: Results of the Generalized Linear Mixed Model for Dcgs / Wcgs presence / absence and environmental variables. With number of trees surveyed and presence / absence of ants as a covariable.

	Tests of Model Effects Dcgs Type III			Tests of Model Effects Wcgs Type III		
	Wald Chi-Square	df	Sig.	Wald Chi-Square	df	Sig.
(Intercept)	.303	1	.582	9.558	1	.002
Altitude	*3.891*	*5*	*0.565*	*5.628*	*5*	*0.344*
Farm Size	*2.791*	*2*	*0.248*	7.515	2	.023
Shade	*0.693*	*3*	*0.875*	8.313	3	.040
Age of Trees	*0.865*	*3*	*0.834*	42.066	3	.000
No. of Trees (Covariable)	*1.769*	*1*	*0.183*	*0.113*	*1*	*0.737*
Ants (Covariable)	58.174	1	.000	47.232	1	.000
Cells in italics are the terms deleted from the main effects model. Non italicised = final terms in the model.						

Table 11.2.7 shows the results of a similar analysis using the ants as the response variable with all the factors but also using CGS as a covariate. Here altitude and shade show significant differences in ant abundances in both survey methods.

Table 11.2.7: Results of the Generalized Linear Mixed Model for ant presence / absence and environmental variables. With number of trees surveyed and presence / absence of CGS as a covariable.

	Tests of Model Effects Dcgs Type III			Tests of Model Effects Wcgs Type III		
	Wald Chi-Square	df	Sig.	Wald Chi-Square	df	Sig.
(Intercept)	2.978	1	.084	43.104	1	.000
Altitude	11.405	5	.044	17.740	5	.003
Farm Size	*4.156*	*2*	*.125*	3.965	2	0.38
Shade	17.305	3	.001	11.296	3	.010
Age of Trees	*2.485*	*3*	*0.478*	*0.326*	*3*	*0.955*
No. of Trees (Covariable)	*1.737*	*1*	*0.188*	*0.221*	*1*	*0.638*
Dcgs (Covariable)	41.997	1	.000	21.330	1	.000

Initially, factors influencing CGS appeared to be altitude, farm size and shade level. As ants are thought to provide protection of CGS from natural enemies it was deemed sensible to include their presence in the model as a covariable. However, when the presence of ants is included as a covariable, and thus removing their influences on CGS, most of the environmental variables have no significant effects on CGS presence. Only in the whole coffee block survey were shade, farm size and age of trees found to be significant.

However, conducting the same analysis on ants (i.e. ants as the response variable) reveals that altitude and shade are the significant factors for the presence of ants with CGS as a covariable. Thus the complex intertwined relationship between ants and CGS, and the infestations in the EHP of PNG appear to be influenced by altitude and shade levels. Where ants have favourable conditions CGS will thrive, i.e. where shade is low and at an altitude of 1500masl.List of publications produced by project.

11.3 Appendix C: Ant diversity survey

Introduction

In order to identify possible problematic ant species associated with CGS a survey was commissioned by the project team and conducted by Mr Katayo Sagata (formerly of the World Conservation Society, PNG. The full report is listed in the reference section: Sagata 2008. Four coffee farms were chosen situated along the Okuk Highway, within the EHP. These farms ranged from an altitude of 1410 - 1714masl and were sampled between January and March 2008 Table 11.3.1

Table 11.3.1: Site details of farms surveyed in ant diversity survey.

Name	Location	Farm Size	Altitude
Notofana	S6°00'50.96", E145°21'01.60"	<5ha	1714masl
Umnifentenu	S6°14'57.46", E145°54'58.56"	<5ha	1594masl
Bena Bridge	S6°11'.039", E145°21'01.60",	>5ha	1410masl
Aiyura	S6°19'55.95", E145°54'09.49",	>5ha	1630masl

Methods

To measure ant diversity and abundance and their spatial distribution within coffee farms, five 50m long transects were established at 5m intervals from the adjacent road at each farm.

Surface dwelling ants and arboreal ants

Pitfall traps were used to sample surface dwelling invertebrates which consisted of 7cm diameter cups filled with preserving fluid. A trap was placed every 5m along each of the five 50m transects. After circa five days the traps were collected and the pitfall trap contents sorted and the ants identified.

Sampling arboreal ants used baited traps placed on both coffee and shade trees between 1-1.5m above the ground at 10m intervals per 50m transect. The bait traps were checked every 20 minutes for three hours and the abundance of ants and species identified. Additionally reference samples were collected to confirm field identifications.

Effect of canopy cover and shade tree size on ant diversity and abundance and CGS abundance

Canopy cover and size of shade trees were used as indicators of management regime to test for effects on ant diversity and abundance, and CGS abundance. At 5m intervals along each transect percentage canopy cover was recorded using a spherical concave densitometer (model C, Forest Densitometers, Bartlesville, Oklahoma, USA).

Ants attending CGS

Ant species attending CGS were defined as ants observed patrolling infested parts of trees, building mud tunnels over CGS or nesting on or in the coffee tree. To establish if there is an association between the abundance of attending ants and CGS, six coffee trees infested with CGS were surveyed along each transect. The barrier method Tanglefoot (Tanglefoot® Company, Grand Rapids, Michigan, USA) was applied around the main stem of the infested tree to prevent ants from either leaving or entering the tree. Ants attending CGS from the stem and the foliage of the tree were collected and identified using the following referenced works (e.g. Bolton, 1994).

Pearson product moment correlation was used to explore ant-CGS relationships but only using the most common and abundant ant species. It was thought that these were likely to be most associated with the CGS (e.g. Abbott & Green, 2007).

Effect of road on number of coffee trees infested by CGS

The effect of distance from the road on CGS was investigated by recording the number of infested trees along the transects. Spearman rank was used to test for correlations between distance and the number of coffee trees infested by CGS.

Statistical analyses

The data was arranged in three-way contingency table (age of coffee tree, CGS and the invasive ant species *Pheidole megacephala* and *Anoplolepis gracilipes*) and used log-linear model to examine the effect of these factors on infestation and mortality of coffee branches and shoots. The resulting model in all the log-linear analysis was assessed using the "step" function in "R" (R Development Core Team, 2005). In all the analysis, workers of the most abundant ant species at each farm were used as only workers forage and attend CGS.

EstimateS was used to analyse ant species richness by combining all the methods (Colwell & Coddington, 1994). This allowed for the approximate total richness per site to be calculated based on the richness estimators (Chap 2, Incidence-Based Coverage Estimator (ICE), and Michaelis–Menten Means (MMMeans)). These estimate total richness per site based on observed species richness. Ants are social insects and entire colonies may occur in single samples, thus presence / absence data was used to reduce the effect of spatial aggregation (Longino, Coddington & Colwell, 2002).

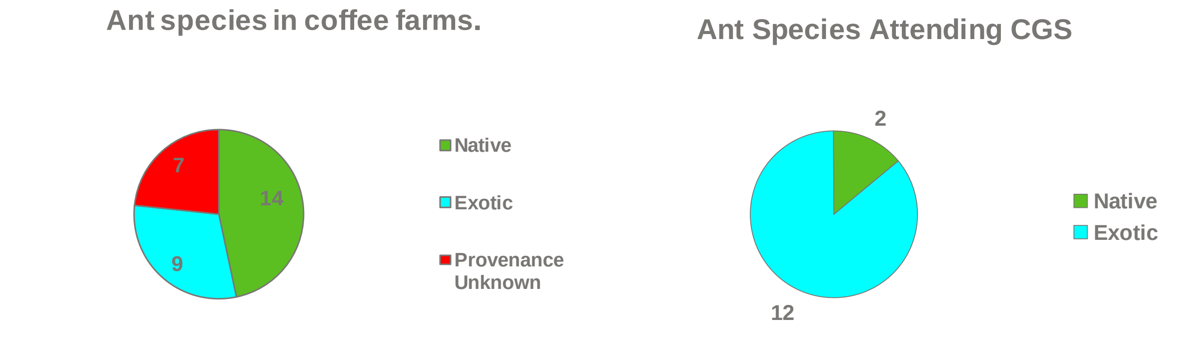
Key results

Species diversity on four coffee farms in EHP was examined together with the effects of ant-CGS association on coffee trees. Ant species varied among the four farms with estimated number of species not exceeding 25 per farm. Combining all the farms resulted in 30 ant species being found. Of these 14 were native, nine were exotic and seven were ant species whose origin is unknown. Of the total number of ant species 14 species attended CGS and of these only two were native (*Rhoptromyrmex* sp1 and *Rhoptromyrmex melleus*). By contrast six exotics and seven species (origin unknown) attended the CGS. See Figure 11.3.1.

Ants attending coffee green scales

Over half the total ant species occurring in the coffee farms attended CGS and almost all of these ant species were exotics or ant species whose origin is unknown. *Rhoptromyrmex sp*1 and *R. melleus* were the only two native ant species that attended CGS. Of the exotic ant species attending the CGS *Anoplolepsis gracilipes* (yellow crazy ant) and *Pheidole megacephala* (African big headed ant) are two of the most invasive ant species with *A. gracilipes* being the most aggressive and invasive (Lowe, Brown & Boudjelas, 2000). These two species together with *Technomyrmex albipes* were most common on the spatial scale and dominated the available resources. This may explain why there were low numbers of native ant species attending the CGS, or it could be that many native ants do not form mutualistic associations with the CGS.

Figure 11.3.1: Number of native and exotic ant species found in Eastern Highlands Province of PNG and number of ant species attending CGS.



The most common and abundant ant species attending CGS (*A. gracilipes* and *P. megacephala*) in this study were not correlated with CGS abundance but it is well known that these ants do attend CGS (Buckley, 1987; Hanks & Sadof, 1990; Buckley & Gullan, 1991; Holway et al., 2002; Abbott & Green, 2007). The failure to find a correlation can be explained by sampling effort. For example, *A. gracilipes* and *P. megacephala* colonies naturally contain many thousands of workers. As such counting all individuals in the survey either attending CGS or foraging for different resources would lead to over representing the actual number of workers attending the CGS.

Infestation of coffee trees by CGS

The factors such as age of coffee tree, CGS abundance, and ant species: *A. gracilipes, P. megacephala* and the interaction of these factors on CGS infestations of whole trees varied among farms. For example, age of coffee tree and interaction between *P. megacephala* did not have an effect on CGS infestations at Bena Bridge, but it did at Aiyura. When these variables were modelled against the mortality of branches and shoots, significant effects of CGS abundance were identified. Again, infestation and subsequent mortality of coffee tree varied greatly among the age of coffee trees and location.

Generally, light infestations were observed to be most prevalent in matured and young coffee trees, heavy infestations were prevalent on shoots and moderate infestations varied among age of coffee trees. Regardless of the infestation level, infestations directly resulted in the mortality of coffee tree branches and shoots but not the whole mature or young trees.

Effect of road on number of coffee trees infested by CGS

Roads are one of the major routes through which exotic species can be introduced and spread (Mack et al., 2000). Invasive ant species colonise disturbed habitats such as towns and roads before native habitats (Kock & Giliomee, 1989; Ward & Harris, 2005). Coffee farms can be considered disturbed habitats, and may have less resilience to invasion (Elton, 1958; Tilman, 1999). For this reason invasive species can establish along the roads where they find readily available resources with less competition from indigenous species. This appears to be the situation with CGS and the exotic ants. CGS readily finds available food resources (i.e. coffee trees) and subsequently *A. gracilipes, P. megacephala, T. albipes, T. melanocephalum* and other invasive ants find resources readily provided by CGS. As a result, more coffee trees are infested near the road than further away from the road.

Observations on ant nesting and CGS attendance behaviour

Where *P. megacephala* occurred, they were observed nesting at the foot of coffee trees and constructing soil tunnels over CGS and connecting them directly to the nest (see plate). This can also provide CGS with protection from natural enemies. The native *Oligomyrmex* sp1 was not observed tending CGS but it did build mud tunnels side-by-side with *P. megacephala*. It is known that *Oligomyrmex* species predate on other ant species broods, and they could be predating on *P. megacephala*.

Technomyrmex albipes have a similar nesting behaviour but they do not build mud tunnels over the CGS and they do not nest in the soil but tend to be arboreal. *Anoplolepis gracilipes* nests in the ground and workers tend CGS. It is the most aggressive species found on coffee farms and apart from its large size it can spray formic acid at prey or indeed natural enemies of CGS. *Anoplolepis gracilipes* is known to out compete large invertebrates (e.g. Christmas Island (O'Dowd, Green & Lake, 2003)) and it is possible that any natural enemies of CGS (e.g. ladybird beetles and parasitoids) are prevented from controlling CGS. Indeed *A. gracilipes* was observed harassing spiders and ladybird beetles on the coffee branches infested with CGS.

Discussion & summary

Although canopy cover and size of shade trees did not have an effect on ant diversity in all the coffee farms, other variables such as litter layer, pruning, use of chemicals, land-use history are perhaps responsible for the difference among the farms. In all the farms, farmers use herbicides and fertilisers (Katayo Sagata, pers. obs.) and it is possible that their use could be having a negative effect on ant richness because an increase in intensity of management can decrease ant species richness in coffee farms (Philpott, Perfecto & Vandermeer, 2006). The presence of invasive ants such as *A. gracilipes, P. megacephala* and *T. albipes* and *T. melanocephalum* can potentially displace less competitive ants because invasive ants are known to displace native ant fauna and invertebrates (Ward, 1987; Holway et al., 2002; Abbott & Green, 2007). The high abundance of *A. gracilipes* and *P. megacephala* observed here were perhaps from a single colony because these ants are known to consist of spatially separated super colonies consisting of thousands of workers (Holway, 1999; Holway et al., 2002). Where they invade, *A. gracilipes* and *P. megacephala* use their numerical abundance and aggressive behaviour resulting in the displacement of native and less aggressive ants (Hoffmann, Andersen & Hill, 1999; Abbott, 2006). Three of the exotic species were spatially most common and abundant (litter, stump and foliage active) were *Anoplolepis gracilipes*, *P. megacephala* and *T. albipes*. These results suggest that spatially common and abundant ants are tracking resources on a spatial structure (e.g. honeydew from CGS). This is not surprising because tracking resource on a spatial structure is a common phenomenon in invasive ants (Abbott & Green, 2007). Domination of space and resources using numerical strength can prevent other ant species from attending CGS or interfere with natural enemies of CGS such as ladybird beetles (Coccinellidae) or biological controls. For example, during the survey *A. gracilipes* and *P. megacephala* were observed “scaring away” spiders, ladybird beetles and other ants that came to parts of the coffee tree infested with CGS.

11.4 Appendix D: Ant exclusion experiment 1

Introduction

Ants are thought to provide both sanitation and protective services to CGS (Bach, 1991; Stadler & Dixon, 2008). The exclusion of ants may reduce scale levels by either: (i) halting the removal of excess honey dew by ants thus increasing the mortality of crawlers – the sanitation hypothesis; or (ii) allowing natural enemies access to scales and thus increasing predation – protection hypothesis. As such, an experiment was set up to test this hypothesis that ants enhance the CGS infestations at CIC Aiyura, PNG.

Materials & methods

To assess the impact of ants on CGS infestation levels a total of 20 young coffee trees, circa two years old, and 20 mature coffee trees were randomly selected within a non shaded coffee block. These trees were then split into two treatment groups, control and ant exclusion. The control allowed ants access to the trees, and the barrier method (Tanglefoot) was used in the ant exclusion treatment. Any differences in scale numbers would therefore be due to either the presence or absence of ants. Regular recordings were conducted from 14th December 2007 to 31st March 2008. Recordings included assessing levels of; live adult scale, parasitised scale, *Lecanicillium lecanii*-infested scale, abundance of ladybirds (coccinellids), number of green cherry and number of internodes.

CGS populations chosen from randomly selected trees should not differ significantly. To test this, the data collected at the start of the experiment was subjected to a one-way ANOVA checking for differences between the treatment groups. Following this all the data was subjected to statistical testing using the repeated measures function in Genstat 11 within the REML package.

Results

Checking of the initial CGS abundance revealed no significant differences between the treatment groups at the start of the experiment. The trends shown in Figure 11.4.1 and Figure 11.4.2 appear to show that when ants are excluded, green scale infestation levels reduce (mature coffee trees) or reduce to zero (young coffee trees).

Figure 11.4.1: Log10 mean CGS infestation levels per tree over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

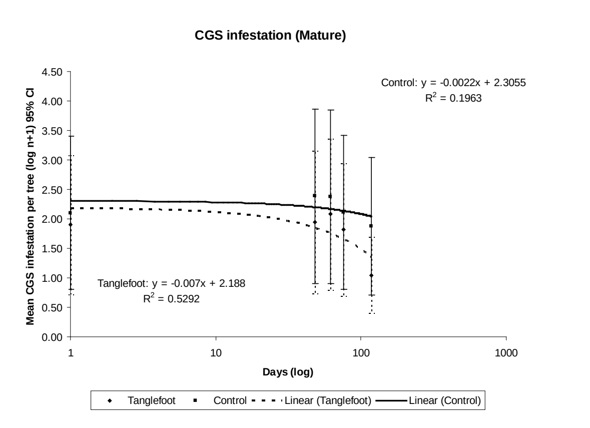


Figure 11.4.2: Log10 mean CGS infestation levels per tree over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

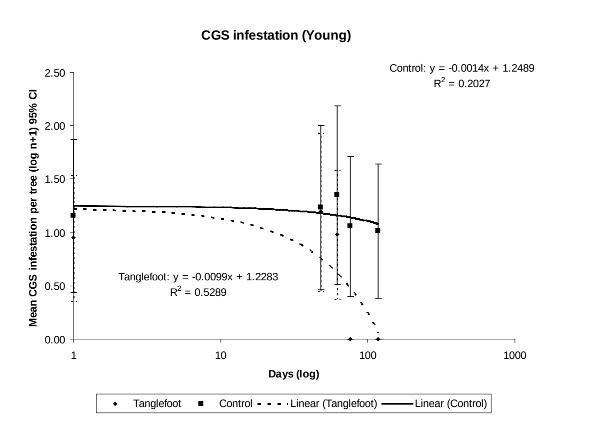


Table 11.4.1 shows the results of the repeated measures test. Both mature and young coffee showed significant differences in CGS abundance between day and treatment. However, there was no significant day × treatment interaction in both cases. Thus making

it difficult to draw any conclusions but the figures suggests a reduction in CGS abundance with ant removal.

Table 11.4.1: CGS infestation levels per tree over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

	Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Mature Coffee:	Day	45.32	4	10.33	25.7	<0.001
	Treatment	6.47	1	6.47	23	0.018
	Day.Treatment	6.6	4	1.5	25.7	0.23
Young Coffee:	Day	15.71	4	3.56	25.2	0.02
	Treatment	7.58	1	7.58	32	0.01
	Day.Treatment	5.73	4	1.3	25.2	0.297

Figure 11.4.3 and Figure 11.4.4 show the percentage healthy CGS, i.e. the proportion of scale that was neither parasitised or infected by fungus. Repeated measures tests revealed significant differences between the control and ant exclusion for the mature coffee, both between treatment, and day × treatment interaction. This suggests a decline in healthy CGS over time, due to ant exclusion. However, the young coffee differed significantly between treatment but there was no day × treatment interaction (Table 11.4.2).

Figure 11.4.3: Mean proportion of healthy CGS (scale that is neither parasitised nor infected with *Lecanicillium* sp.) over time. All values are means with 95% confidence interval error bars.

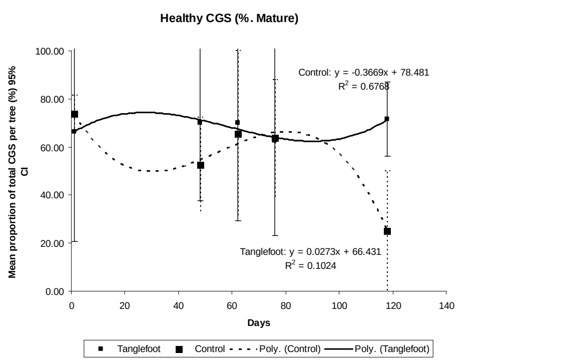


Figure 11.4.4: Mean proportion of healthy CGS (scale that is neither parasitised nor infected with *Lecanicillium* sp.) over time. All values are means with 95% confidence interval error bars.

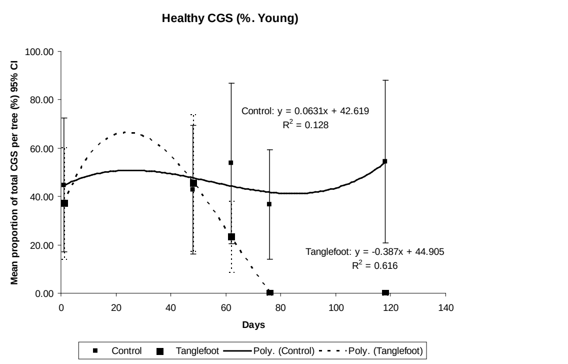


Table 11.4.2: Mean percent healthy scale levels per tree over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was Arcsin transformed.

	Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Mature Coffee:	Day	23.54	4	5.31	23.8	0.003
	Treatment	7.03	1	7.03	21.7	0.015
	Day.Treatment	45.83	4	10.34	23.8	<0.001
Young Coffee:	Day	10.2	4	2.33	26	0.083
	Treatment	13.78	1	13.78	25.5	0.001
	Day.Treatment	8.93	4	2.04	26	0.118

When examining the percent abundance of non-healthy CGS, mature and young coffee, (Figure 11.4.5 and Figure 11.4.6), there was no significant day × treatment interaction (Table 11.4.3).

Figure 11.4.5: Mean proportion of non-healthy CGS (scale that is either parasitised or infected with *Lecanicillium* sp.) over time. All values are means with 95% confidence interval error bars.

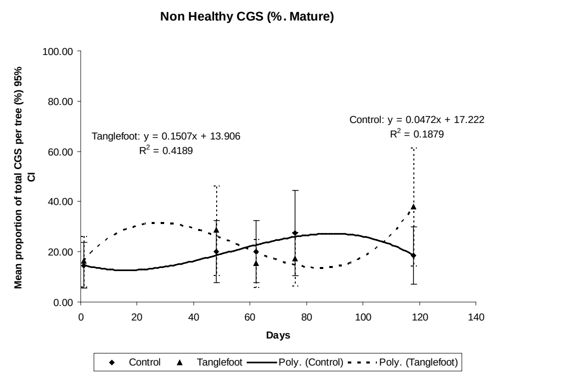


Figure 11.4.6: Mean proportion of non-healthy CGS (scale that is either parasitised or infected with *Lecanicillium* sp.) over time. All values are means with 95% confidence interval error bars.

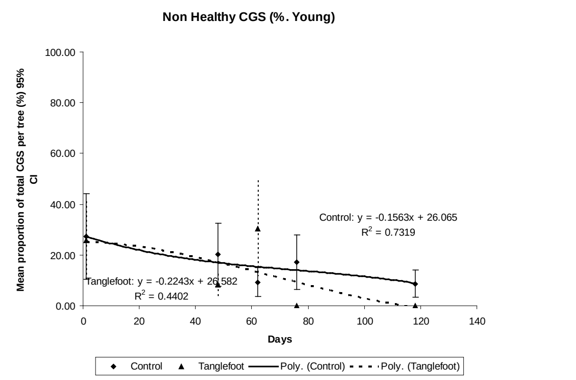


Table 11.4.3: Mean percent non-healthy scale levels per tree over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was Arcsin transformed.

	Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Mature Coffee:	Day	19.73	4	4.45	23.7	0.008
	Treatment	0.33	1	0.33	23.6	0.571
	Day.Treatment	11.15	4	2.51	23.7	0.069
Young Coffee:	Day	17.69	4	4	24.7	0.012
	Treatment	6.86	1	6.86	30.5	0.014
	Day.Treatment	10.27	4	2.32	24.7	0.085

The two components of the non-healthy scale were the proportions of CGS either parasitised or infected with fungi. Closer examination of the abundance of CGS parasitised only, shows that there is a difference between the treatment which is significant, i.e. there are fewer parasitised scales where ants are present (Table 11.4.4). However, this is not the case for the young coffee (Figure 11.4.8). The results were the same for entomopathogenic fungi, there was a day × treatment interaction but no difference between treatments (Figure 11.4.9, Figure 11.4.10).

Figure 11.4.7: Log10 mean parasitised CGS per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

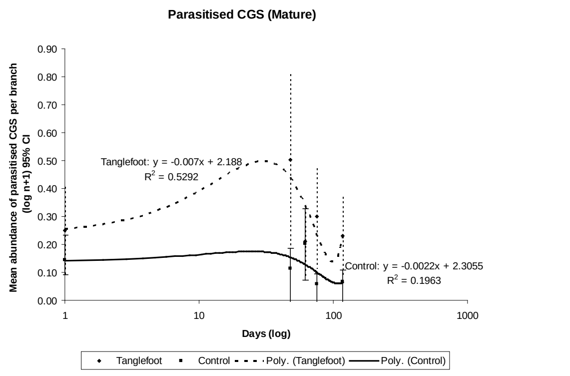


Figure 11.4.8: Log10 mean parasitised CGS per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

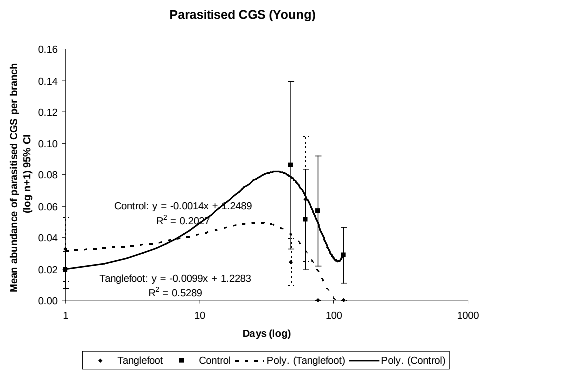


Figure 11.4.9: Log10 mean fungus infected CGS per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

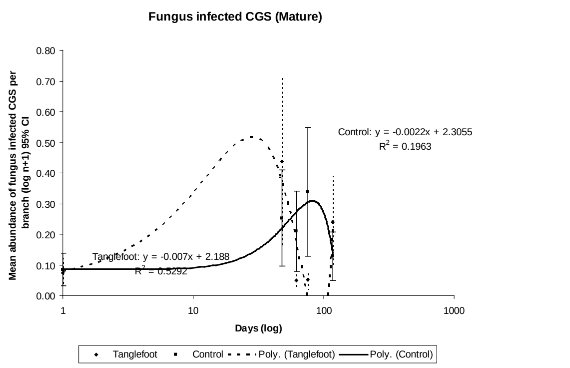


Figure 11.4.10: Log10 mean fungus infected CGS per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

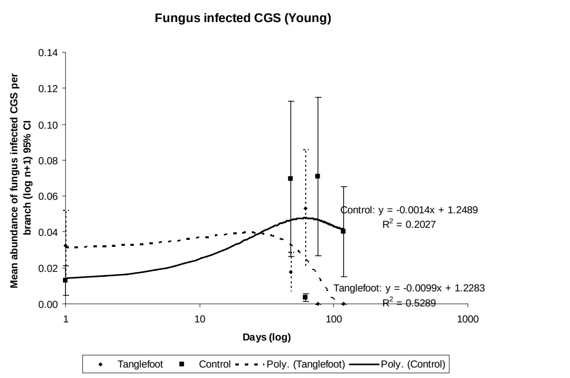


Table 11.4.4: Mean parasitised CGS per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

	Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Mature Coffee:	Day	10.16	4	2.3	25.1	0.087
	Treatment	24.68	1	24.68	30.4	<0.001
	Day.Treatment	31.64	4	7.17	25.1	<0.001
Young Coffee:	Day	6.22	4	1.4	24.2	0.264
	Treatment	1.96	1	1.96	36.9	0.17
	Day.Treatment	5.3	4	1.19	24.2	0.339

Table 11.4.5: Mean *Lecanicillium* infected CGS per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

	Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Mature Coffee:	Day	27.91	4	6.3	24.3	0.001
	Treatment	0.92	1	0.92	31.4	0.345
	Day.Treatment	24.25	4	5.47	24.3	0.003
Young Coffee:	Day	3.8	4	0.86	24.9	0.502
	Treatment	0.55	1	0.55	30.2	0.464
	Day.Treatment	12.01	4	2.72	24.9	0.052

In both mature and young coffee, green cherry production, the treatments did not differ significantly (Figure 11.4.11, Figure 11.4.12 and Table 11.4.6).

Figure 11.4.11: Log10 mean green cherry production per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

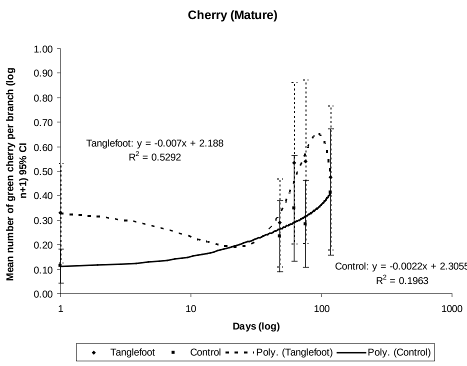


Figure 11.4.12: Log10 mean green cherry production per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

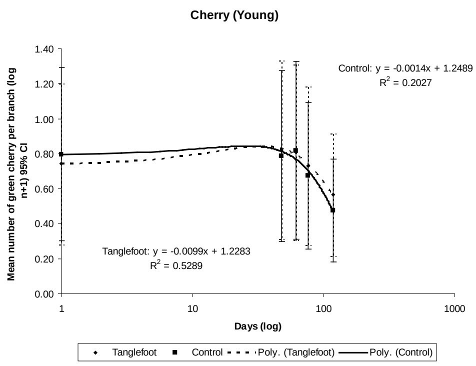


Table 11.4.6: Mean green cherry per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

	Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Mature Coffee:	Day	11.59	4	2.63	24.5	0.059
	Treatment	3.68	1	3.68	17.8	0.071
	Day.Treatment	9.58	4	2.18	24.5	0.102
Young Coffee:	Day	63.93	4	14.49	24.9	<0.001
	Treatment	0.2	1	0.2	26.3	0.655
	Day.Treatment	2.3	4	0.52	24.9	0.721

Discussion & conclusion

There is plenty of evidence for scale reducing yield but in this experiment no differences were found, and here the experiment centred primarily on removing ants and thus scale. This experiment was only conducted for four months and is likely to have been too short to detect differences. For this to be measured, field trials of at least one coffee growing season would be required.

Whilst there were no significant reductions in CGS levels in terms of the day × treatment interaction, as a result of the ant exclusion method, it is clear that there were significant differences in the proportion of healthy scale populations over time. This suggests that the treatment may have subtle effects but it is not clear what is responsible for this decrease in the proportion of healthy scale. The data does suggest that increased non-healthy scale levels e.g. parasitised by species such as *Metaphycus baruensis* or entomopathogenic fungi such as *Lecanicillium* sp. might be the cause, again this is not clear-cut.

11.5 Appendix E: Ant exclusion experiment 2

Introduction

Ants are thought to provide both sanitation and protective services to CGS (Bach, 1991; Stadler & Dixon, 2008). The exclusion of ants may reduce scale levels by either: (i) halting the removal of excess honey dew by ants thus increasing the mortality of crawlers – the sanitation hypothesis; or (ii) allowing natural enemies access to scales and thus increasing predation – protection hypothesis. As such, an experiment was set up to test this hypothesis at CIC Aiyura, PNG.

Materials & methods

To assess the impact of ants on CGS infestation levels a total of 20 coffee trees were selected within a non shaded coffee block. These trees were then split into two groups, control and ant exclusion. The control allowed ants access to the trees, and the barrier method (Tanglefoot) was used in the ant exclusion treatment. Any differences in scale numbers would therefore be due to either the presence or absence of ants. Regular recordings were conducted from 22nd April 2008 to 28th August 2008. Recordings included assessing levels of; live adult scale, parasitised scale, *Lecanicillium lecanii*-infested scale, abundance of ladybirds (coccinellids), number of green cherry and number of internodes.

Results

Trends in Figure 11.5.1 and Figure 11.5.2 show that when ants are excluded, CGS infestations and the proportion of healthy scale are eventually reduced to zero. The results of the repeated measures in Genstat 11 revealed significant reductions in CGS levels and healthy scale between treatments and over time (day × treatment interaction), see Table 11.5.1 and Table 11.5.2. Therefore, excluding ants reduces healthy scale and overall scale abundance over time.

Figure 11.5.1: Mean CGS infestation level per tree over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

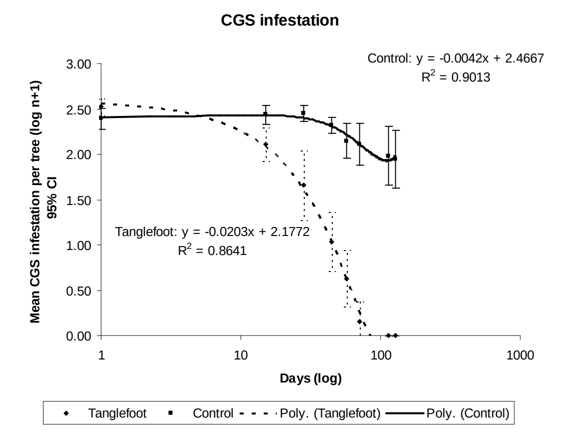


Figure 11.5.2: Mean proportion of healthy CGS (scale that is neither parasitised nor infected with *Lecanicillium* sp.) over time. All values are means with 95% confidence interval error bars.

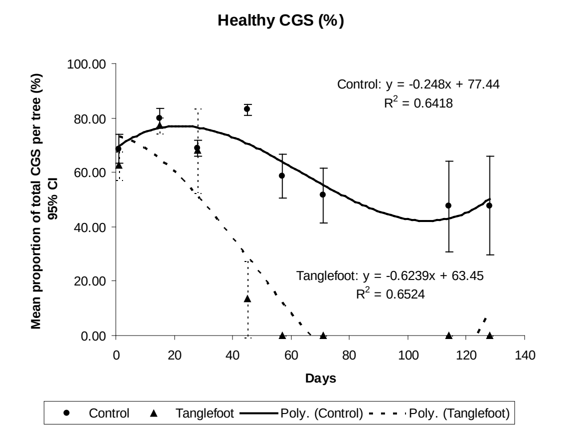
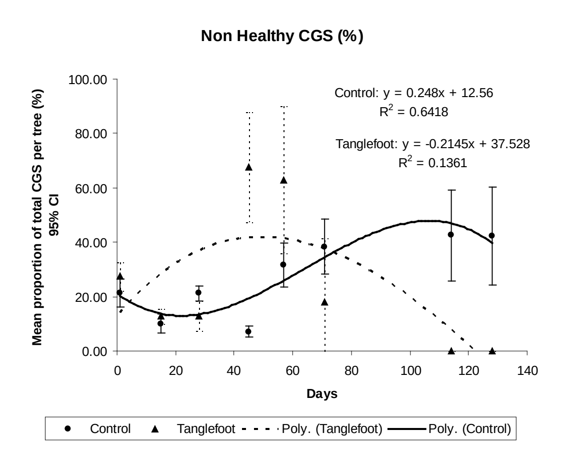


Figure 11.5.3: Mean proportion of non-healthy CGS (scale that is either parasitised or infected with *Lecanicillium* sp.) over time. All values are means with 95% confidence interval error bars.



When comparing the non-healthy CGS (Figure 11.5.3), there is a significant day × treatment interaction indicating a change over time. It can be seen that the proportion of non-healthy scale increases where ants are excluded, but then decreases as all the CGS die. However, there is no overall difference between the treatments which can be explained by this rise and fall of CGS in the ant exclusion treatment.

Table 11.5.1: CGS infestation levels per tree over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	331.14	7	40.9	31.2	<0.001
Treatment	17.52	1	17.52	21.6	<0.001
Day.Treatment	188.61	7	23.3	31.2	<0.001

Table 11.5.2: Mean percent healthy scale levels per tree over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was Arcsin transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	363.85	7	44.88	31.4	<0.001
Treatment	142.45	1	142.45	31.4	<0.001
Day.Treatment	274.33	7	33.84	31.4	<0.001

Table 11.5.3: Mean percent non-healthy scale levels per tree over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was Arcsin transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	76.61	7	9.48	31.3	<0.001
Treatment	0.12	1	0.12	18	0.728
Day.Treatment	88.95	7	11.01	31.3	<0.001

The two components of the non-healthy scale were the proportions of CGS either parasitised or infected with fungi. On closer examination of the abundance of CGS parasitised only, Figure 11.5.4 shows that there is a significant difference between treatments and a day × treatment interaction. Over time the there are fewer parasitised scale where ants are present because the CGS is declining in abundance. Where the entomopathogenic fungi are concerned, there was a day × treatment interaction but no difference between treatments suggesting an overall decline in the fungus (Figure 11.5.5). Likewise, Figure 11.5.6 shows a significant day × treatment interaction but no overall difference between treatments. There does appear to be an increase in coccinellid abundance after the ant exclusion treatment is applied, but this declines over time to similar levels to the control.

Green cherry production did not differ significantly between treatments (Figure 11.5.7).

Figure 11.5.4: Log10 mean parasitised CGS per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

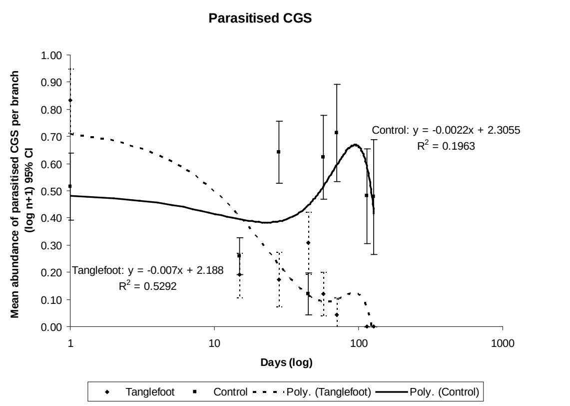


Figure 11.5.5: Log10 mean fungus infected CGS per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

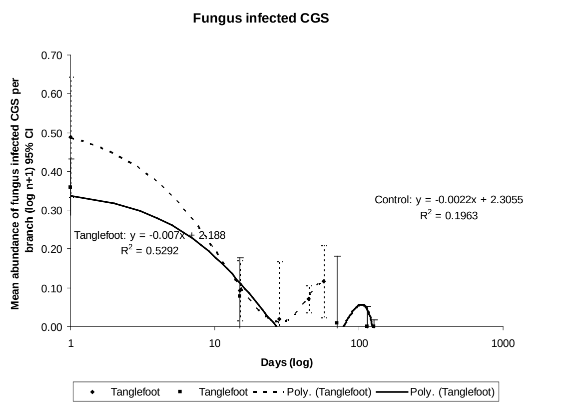


Figure 11.5.6: Log10 mean coccinellid abundance per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

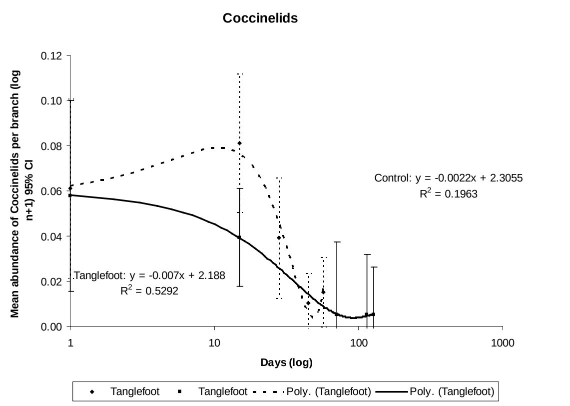


Figure 11.5.7: Log10 mean green cherry production per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. All values are means with 95% confidence interval error bars.

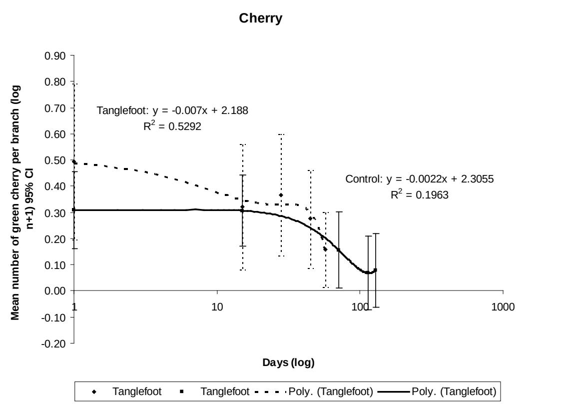


Table 11.5.4: Mean parasitised CGS per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	141.87	7	17.63	32.6	<0.001
Treatment	3.27	1	3.27	29.9	0.081
Day.Treatment	127.4	7	15.83	32.6	<0.001

Table 11.5.5: Mean *Lecanicillium* infected CGS per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	77.24	7	9.64	33.6	<0.001
Treatment	2.59	1	2.59	33.4	0.117
Day.Treatment	55.6	7	6.94	33.6	<0.001

Table 11.5.6: Mean coccinellids per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	23.37	7	2.88	31.2	0.019
Treatment	2.48	1	2.48	39.8	0.123
Day.Treatment	20.05	7	2.47	31.2	0.039

Table 11.5.7: Mean green cherry per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	19.54	7	2.43	32	0.041
Treatment	0.83	1	0.83	18.2	0.374
Day.Treatment	6.49	7	0.81	32	0.589

Discussion & conclusion

There were significant reductions in CGS levels and in the proportion of healthy scale populations over time as a result of the ant exclusion method. This suggests that the there are natural enemies present which, if ants are controlled, can control CGS. It is not fully clear what is responsible for this control but the data does suggest that increased parasitism by *Metaphycus baruensis* or entomopathogenic fungi such as *Lecanicillium* sp. might be the cause. It is also possible that the highly mobile coccinellids may also be responsible.

There is plenty of evidence for scale reducing yield but in this experiment no differences were found, and here the experiment centred primarily on removing ants and thus scale. This experiment was only conducted for four months and is likely to have been too short to detect differences. For this to be measured, field trials of at least one coffee growing season would be required.

11.6 Appendix F: Ant exclusion experiment 3

Introduction

Ants are thought to provide both sanitation and protective services to CGS. The exclusion of ants may reduce scale levels by either: (i) halting the removal of excess honey dew by ants thus increasing the mortality of crawlers – the sanitation hypothesis; or (ii) allowing natural enemies access to scales and thus increasing predation – protection hypothesis. An experiment was set up to test this hypothesis at CIC Aiyura, PNG.

Materials & methods

Due to Tanglefoot not being entirely effective as a barrier to ants in the first experiment (experiment 1), a second experiment was conducted (see experiment 2) which showed that the exclusion of ants decreased CGS levels. A third experiment was set up to compare the use of an ant bait insecticide and Tanglefoot. The experiment was set up at CIC Aiyura (Block 248) to assess the two methods of ant control on CGS infestation levels on coffee. A total of 30 trees were selected within a non shaded coffee block. These trees were then split into two treatment groups and a control. The control allowed ants access to the trees, and the barrier method (Tanglefoot) was used in the ant exclusion treatment 1. Tanglefoot may restrict some natural enemies access as it acts as a barrier around the basal part of the tree and can also act as a trap. The second treatment consisted of the ant bait Amdro which does not restrict natural enemies access to the tree. Any differences in scale numbers would therefore be due to either the presence or absence of ants. Regular recordings were conducted from 9th October 2008 up to 22nd December 2008. Recordings include assessing levels of; live adult scale, parasitised scale, fungal infected scale (*Lecanicillium lecanii*), abundance of ladybirds (coccinellids), number of green cherry and number of internodes. Table 11.6.1 shows the treatment combinations within a block of recycled coffee.

Table 11.6.1: Ant exclusion study 3: Amdro vs Tanglefoot – Aiyura coffee, Block 248.

Treatment Codes	Treatment Description	Treatment Notes
Control	Control	This is the control, no restricted access.
1	Ant Exclusion (Tanglefoot)	Simple ant exclusion - denies ants access to CGS.
2	Ant Exclusion (Amdro)	Baited ant exclusion - should control all ants thus keeping CGS free of ants.

Results

Trends in Figure 11.6.1 appear to show that when ants are excluded, CGS infestations are reduced. The results of the repeated measures test in Genstat 11 revealed significant reductions in CGS levels over time for the ant exclusion methods ($p \leq 0.05$). Additionally, there was a significant overall difference between the two ant exclusion treatments compared to the control ($p \leq 0.05$). Amdro treated trees had significantly lower CGS levels compared to the control and Tanglefoot did not differ significantly from Amdro.

Figure 11.6.2 shows the proportion of healthy CGS and Figure 11.6.3 the proportion of non-healthy CGS. In these two graphs there were no significant between-treatment effects but there was a significant interaction with treatment over time. Excluding ants therefore reduces healthy scale and Amdro reduces overall scale infestations over time. When comparing the non-healthy CGS, there is a significant interaction between non-healthy scale and time but no overall significant difference between treatments, i.e. a significant change over time between treatments but no overall difference between the treatments as a whole. In effect there is a significant decrease in non-healthy scale in the Tanglefoot treatment compared to a significant increase for the control. For full statistical results see annex 11.6.5.

Figure 11.6.1: Log10 mean CGS infestation level per tree over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. Line of best fit using linear regression model. All values are means with 95% confidence interval error bars.

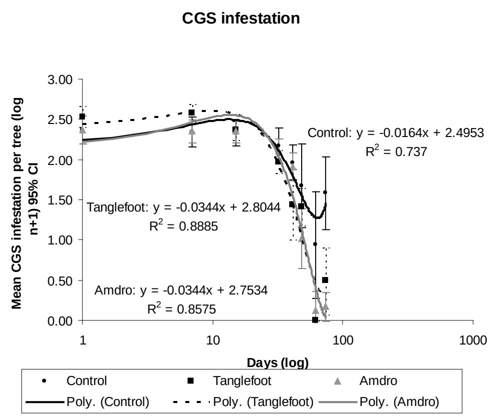


Figure 11.6.2: Mean proportion of healthy CGS (scale that is neither parasitised nor infected with *Lecanicillium* sp.) over time. All values are means with 95% confidence interval error bars.

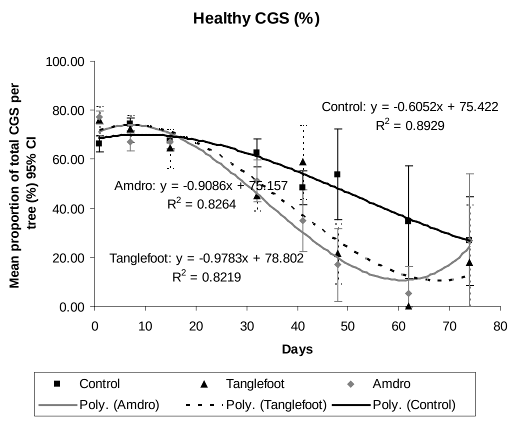
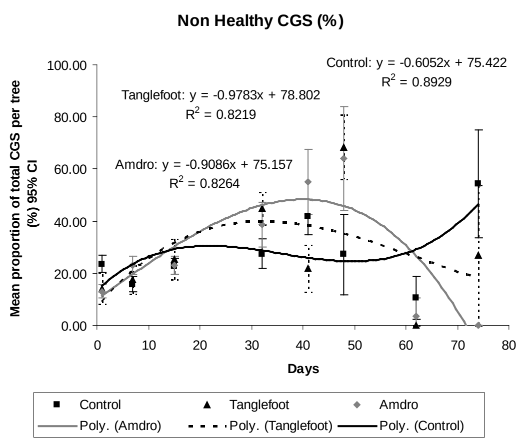


Figure 11.6.3: Mean proportion of non-healthy CGS (scale that is either parasitised or infected with *Lecanicillium* sp.) over time. All values are means with 95% confidence interval error bars.



The two components of the non-healthy scale were the proportions of CGS either parasitised or infected with fungi. Closer examination of the abundance of CGS parasitised only, Figure 11.6.4, shows that there is no significant difference between treatments but there is a significant interaction over time. Over time there are fewer parasitised scales where ants are not present. Where the entomopathogenic fungi are concerned, there was an interaction with treatment and time (Figure 11.6.5).

Figure 11.6.4: Log10 mean parasitised CGS per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. Line of best fit using linear regression. All values are means with 95% confidence interval error bars.

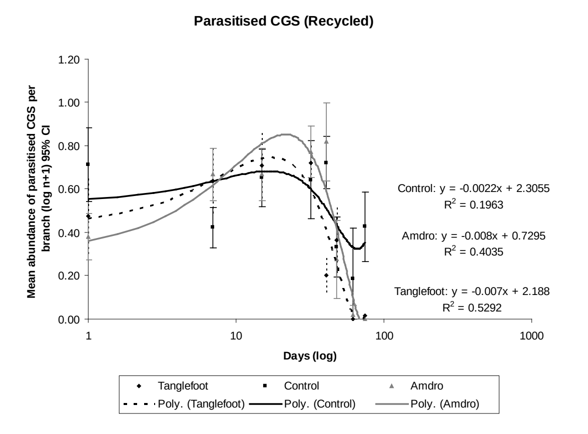


Figure 11.6.5: Log10 mean fungus infected CGS per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. Line of best fit using linear regression. All values are means with 95% confidence interval error bars.

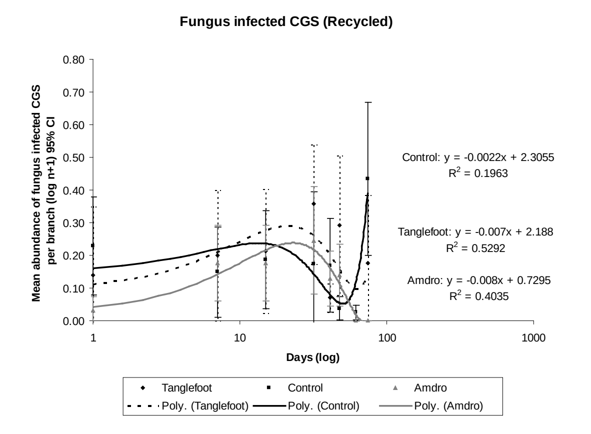


Figure 11.6.6: Log10 mean coccinellid abundance per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. Line of best fit using linear regression. All values are means with 95% confidence interval error bars.

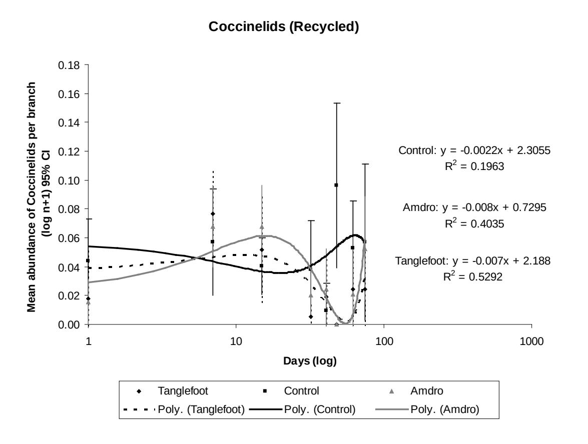
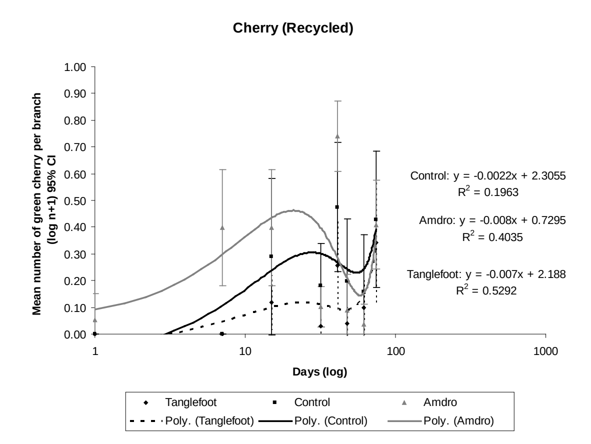


Figure 11.6.7: Log10 mean green cherry production per branch over time (log scale). Comparison between control and Tanglefoot ant exclusion treatments. Line of best fit using linear regression. All values are means with 95% confidence interval error bars.



The abundance of coccinellids (Figure 11.6.6) was also examined and there was no significant difference between treatments but there was a significant interaction with time. Green cherry production treatments did not differ significantly although cherry production over time did (Figure 11.6.7).

Discussion & conclusion

There is plenty of evidence for scale reducing yield but in this experiment no differences were found, and here the experiment centred primarily on removing ants and thus scale. This experiment was only conducted for four months and is likely to have been too short to detect differences in productivity. For this to be measured, field trials of at least one coffee growing season would be required.

There were significant reductions in CGS levels and in the proportion of healthy scale populations over time as a result of the ant exclusion method. This suggests that the there are natural enemies present which if ants are controlled, can control CGS. It is not fully clear what is responsible for this control but the data does suggest that increased parasitism by *Metaphycus baruensis* or entomopathogenic fungi such as *Lecanicillium* sp.

might be the cause. It is also possible that the highly mobile coccinellids may also be responsible.

It is suggested that an additional trial is conducted with an additional treatment using ant bait, plus sleeves to exclude ladybirds but allow parasites of CGS in.

Annex

Table 11.6.2: CGS infestation levels per tree over time. Comparison between control, Tanglefoot ant exclusion and Amdro treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	440.5	7	57.72	53.4	<0.001
Treatment	7.72	2	3.86	36	0.03
Day.Treatment	77.32	14	5.01	73.7	<0.001

Table 11.6.3: Mean percent healthy scale levels per tree over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was Arcsin transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	327.69	7	42.83	51.9	<0.001
Treatment	0.17	2	0.08	29.1	0.919
Day.Treatment	111.58	14	7.21	71.5	<0.001

Table 11.6.4: Mean percent non-healthy scale levels per tree over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was Arcsin transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	260.69	7	34.1	53.1	<0.001
Treatment	4.4	2	2.2	48.6	0.122
Day.Treatment	127.28	14	8.23	73.7	<0.001

Table 11.6.5: Mean parasitised CGS per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	556.33	7	73.08	55	<0.001
Treatment	28.27	2	14.14	44.2	<0.001
Day.Treatment	265.41	14	17.25	75.8	<0.001

Table 11.6.6: Mean *Lecanicillium* infected CGS per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	650.49	7	84.96	52.4	<0.001
Treatment	43.9	2	21.95	47.8	<0.001
Day.Treatment	218.98	14	14.14	72.9	<0.001

Table 11.6.7: Mean coccinellids per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Day	22.48	7	2.92	50.6	0.012
Treatment	6.77	2	3.38	63.4	0.04
Day.Treatment	27.66	14	1.78	71	0.06

Table 11.6.8: Mean green cherry per branch over time. Comparison between control and Tanglefoot ant exclusion treatment using repeated measures. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.					
Fixed term	Wald statistic	d.f.	Wald/d.f.	chi pr	
Day	125.88	6	20.98	<0.001	
Treatment	0.01	2	0	1	
Day.Treatment	29.9	12	2.49	0	

11.7 Appendix G: Ant exclusion experiment 4

Introduction

Previous experiments have shown that the removal of ants significantly reduces the abundance of CGS on coffee trees compared to trees where ants have access to CGS. The findings strongly suggest that ants play a crucial role in CGS infestations (Bach, 1991). As such an experiment was set up to investigate the impact of specific natural enemies on CGS abundance, as it is still unclear what reduces these infestation levels, e.g. is the reduction due to lady birds or parasites, or a combination? The main two questions addressed in this experiment were:

1. What is the effectiveness of parasitoid natural enemies in the presence / absence of ants?
2. What is the effectiveness of the larger coccinellid (ladybird) natural enemy predators in the presence / absence of ants?

Materials & methods

The experiment was set up at CIC Aiyura (Block 248) to both assess the two methods of ant control on CGS on coffee and determine the impact of the respective natural enemies on CGS abundance. In total 120 coffee trees were selected and split into ten blocks of 12 trees and randomly assigned a treatment in each block (see Table 11.7.1 and Figure 11.7.1).

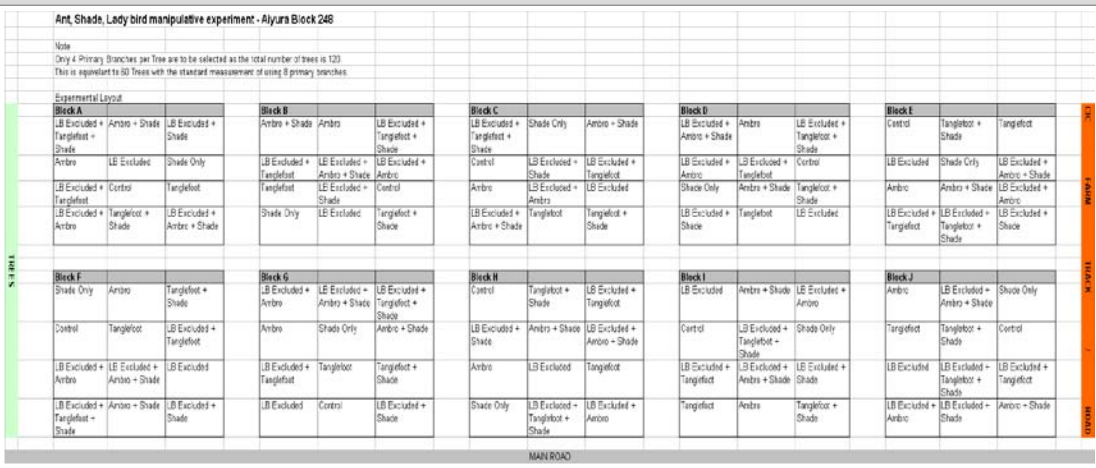
As in previous experiments the control allowed ants access to the trees, and the barrier method (Tanglefoot) was used to exclude ants. Tanglefoot may restrict some natural enemies access as it acts as a barrier around the basal part of the tree and can also act as a trap. Additionally, the ant bait Amdro which does not restrict natural enemies access to the tree was also used as a treatment. Any differences in scale numbers would therefore be due to either the presence or absence of ants.

In contrast to previous experiments, 2mm mesh sleeves were used in order to exclude coccinellids on branches on half the trees. This would allow for changes in CGS abundance to be directly related to either parasitism or fungal infection. Additionally, half the treatments also had artificial shade applied (70% shade), a shade method often used in nurseries.

The experiment was set up in October 2008 in order to coincide with increasing CGS abundance, and initial recordings of CGS, natural enemy, and ant abundance were taken. However, the experiment did not start until 1st May 2009, and ended 22nd September 2009. This is after peak abundance and where CGS declines. Recordings were taken twice a month, including assessing levels of; live adult scale, parasitised scale, fungal infected scale (*Lecanicillium lecanii*), abundance of ladybirds (coccinellids) and ants as well as their identity.

Table 11.7.1: Ant, shade, ladybird, parasitoid manipulative experiment – Aiyura coffee, Block 248.		
Treatment Codes	Treatment name	Treatment description
1	Control	This is the control, no restricted access.
2	Ant Exclusion (Tanglefoot)	Simple ant exclusion - denies ants access to CGS.
3	Ant Exclusion (Amdro)	Baited ant exclusion - kills ants keeping CGS free of ants.
4	Ladybird Exclusion (Mesh Sleave)	This allows ants and parasitoids access to CGS but denies ladybirds and other large predatory invertebrates access to CGS.
5	Ladybird Exclusion (Mesh Sleave) + Tanglefoot	This allows parasitoids access to CGS but denies ants, ladybirds and other large predatory invertebrates access to CGS.
6	Ladybird Exclusion (Mesh Sleave) + Amdro	This allows parasitoids access to CGS but denies ants, ladybirds and other large predatory invertebrates access to CGS.
7	Shade Only	Shaded coffee tree with a single mesh of circa 70% shade. Will be draped over the tree and pinned to the ground. Some sides will be open to allow invertebrates access to CGS. It will resemble an A framed ridge tent, no actual frame will be required, but it should be positioned so that it shades the coffee tree for the whole day.
8	Ant Exclusion (Tanglefoot) + Shade	As in Treatments 2 - 6 but with shade as in treatment 7.
9	Ant Exclusion (Amdro) + Shade	As above
10	Ladybird Exclusion (Mesh Sleave) + Shade	As above
11	Ladybird Exclusion (Mesh Sleave) + Tanglefoot + Shade	As above
12	Ladybird Exclusion (Mesh Sleave) + Amdro + Shade	As above

Figure 11.7.1: Experimental layout



Results & discussion

Figure 11.7.2 shows the mean CGS abundance per branch on ant excluded versus the control trees (ants present). It appears as though there are no differences between CGS abundance on the ant excluded and ants present trees. This goes against previous findings where ant exclusion successfully reduces CGS abundance. In order to understand these results possible causes were investigated:

- Amdro had reduced ants within the experimental field to such an extent that CGS populations declined on both ant excluded and ant present treatments.
- A natural decline in CGS abundance, greater than that of the affect of excluding ants.

In previous studies the exclusion of ants increased the proportion of non-healthy over that of healthy CGS, either parasitised or fungus infected. This was again plotted for this experiment, and if Amdro had reduced ants to such an extent in the whole field then this pattern would be expected throughout the treatments, including the control. As can be seen in Figure 11.7.3 to Figure 11.7.5, this pattern does not exist.

Data on CGS phenology, supplied by CIC, suggests the peak period of CGS abundance is April. A natural decline in CGS abundance could be the cause, and as stated in the methods section the original planned timing of the experiment was to coincide with increasing CGS abundance and include the peak period. However, the experiment was delayed by some six months starting in April and ending in September (see Figure 11.7.6). It is very probable that this was the cause of the inconsistent findings. The results from coccinellid exclusion and shade elements also showed the same patterns (see annex 11.7.4 for results).

Additionally, a list of the coccinellid predators was compiled throughout this study (see Table 11.7.2, annex 11.7.4).

Figure 11.7.2: Mean CGS infestation level per branch over time. Comparison between control (ants present), Tanglefoot (ant barrier exclusion) and Amdro (ant bait exclusion) treatments. All values are means +/- 1 standard error.

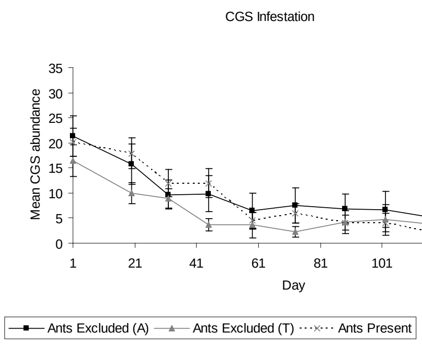


Figure 11.7.3: Mean proportion of healthy versus non-healthy CGS per branch, over time. Control trees, ants present. All values are means +/- 1 standard error.

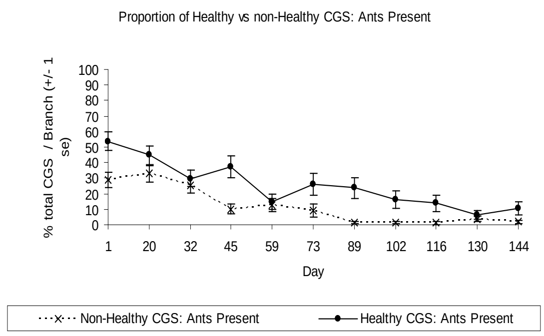


Figure 11.7.4: Mean proportion of healthy versus non-healthy CGS per branch, over time. Ant barrier exclusion trees (Tanglefoot). All values are means +/- 1 standard error.

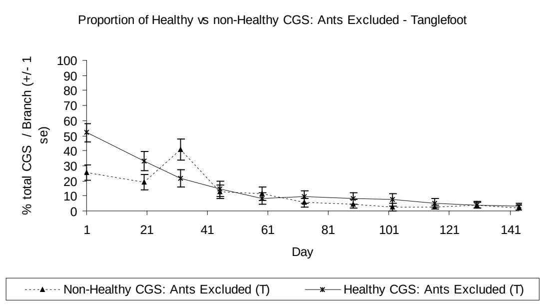


Figure 11.7.5: Mean proportion of healthy versus non-healthy CGS per branch, over time. Ant bait exclusion trees (Amdro). All values are means +/- 1 standard error.

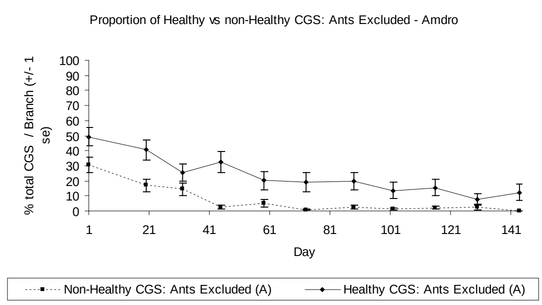
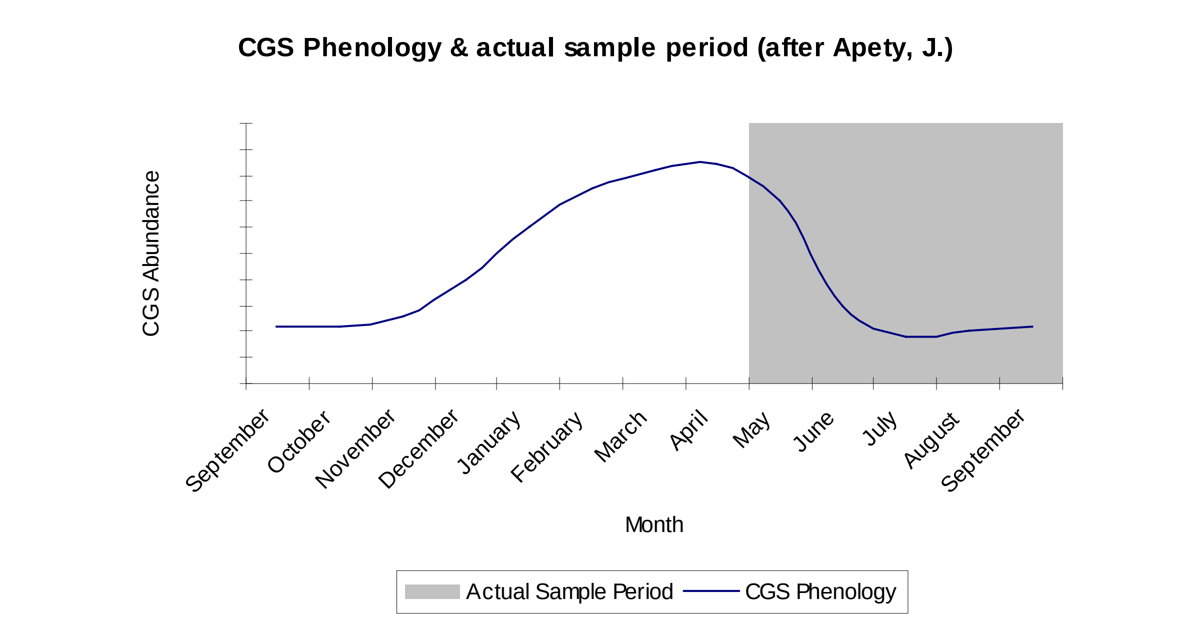


Figure 11.7.6: CGS phenology throughout the year. Data supplied by CIC, recreated after Apety, J.



Annex

Table 11.7.2: List of coccinellid predators found in the Eastern Highlands Province of PNG. Additional notes are provided from the literature.

CIC ID	Status	Distribution	Predator of	Additional Notes
Orcus sp.	Predator	Papua New Guinea (Szent-Ivany 1958)	*Coccus viridis* (Green), Papua New Guinea (Szent-Ivany 1958) *Pulvinaria psidii* (Maskell), Papua New Guinea (Szent-Ivany 1958) *Saissetia coffeae* (Walker), Papua New Guinea (Szent-Ivany 1958)	
Callineda sp.	Predator	Papua New Guinea (Szent-Ivany 1958)	*Coccus viridis* (Green), Papua New Guinea (Szent-Ivany 1958) *Pulvinaria psidii* (Maskell), Papua New Guinea (Szent-Ivany 1958) *Saissetia coffeae* (Walker), Papua New Guinea (Szent-Ivany 1958)	*Harmonia* sp. near *testudinaria* Mulsant Synonyms: *Callineda* sp. near *testudinaria* Mulsant)
Menochilus sp.	Predator	Papua New Guinea (Szent-Ivany 1958), India (Reddy et al 1990),	*Coccus viridis* (Green), India (Reddy et al 1990), Papua New Guinea (Szent-Ivany 1958) *Pulvinaria psidii* (Maskell), Papua New Guinea (Szent-Ivany 1958)	*Cheilomenes sexmaculata* (Fabricius) Synonyms: *Menochilus sexmaculatus* Fabricius
Cryptolaemus sp.	Predator	Papua New Guinea (Szent-Ivany 1963)	*Planococcus minor* (Maskell), Papua New Guinea (Szent-Ivany 1963)	*Cryptolaemus affinis* Crotch

Cryptolaemus sp.	Predator	Australia (B. Pinese In litt. 1999), Hawaii (Reimer Cope & Yasuda 1993), Indonesia Java (Ultee 1929), Indonesia Sulawesi (Leefmans 1928), Kenya (James 1932a), New Caledonia (Chazeau 1981), Puerto Rico (Wolcott 1951), South Africa (James 1932a)	*Coccus viridis* (Green), Hawaii (Reimer Cope & Yasuda 1993), New Caledonia (Chazeau 1981) *Dysmicoccus brevipes* (Cockerell), Hawaii (Illingworth 1929) *Ferrisia virgata* (Cockerell), Indonesia Java (Ultee 1929) Not recorded, Australia (B. Pinese In litt. 1999) *Planococcus citri* (Risso), Indonesia Java (Betrem 1932) *Planococcus kenyae* (Le Pelley), Kenya (James 1932a), South Africa (James 1932a) *Pulvinaria psidii* (Maskell), Puerto Rico (Wolcott 1951) *Rastrococcus iceryoides* (Green), Indonesia Sulawesi (Van der Goot 1948) *Rastrococcus spinosus* (Robinson), Indonesia Java (Leefmans 1928), Indonesia Sulawesi (Leefmans 1928)	*Cryptolaemus montrouzieri* Mulsant

Figure 11.7.7: Mean CGS infestation level per branch over time. Comparison between control (ants present), Tanglefoot (ant barrier exclusion) and Amdro (ant bait exclusion) treatments. Coccinellids (LB) were excluded from the trees with the use of mesh 2mm sleeves. All values are means +/- 1 standard error.

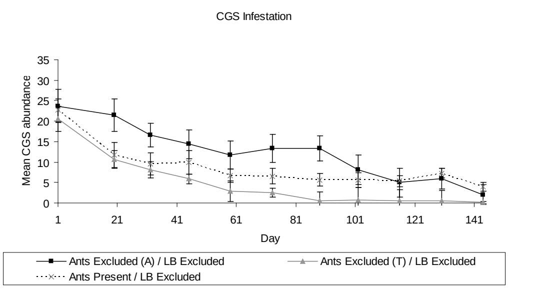


Figure 11.7.8: Mean proportion of healthy versus non-healthy CGS per branch, over time. Control trees, ants present. Coccinellids (LB) were excluded from the trees with the use of mesh 2mm sleeves. All values are means +/- 1 standard error.

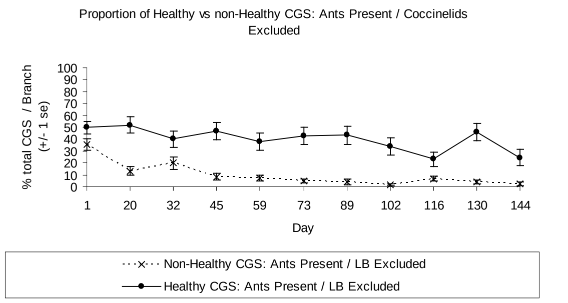


Figure 11.7.9: Mean proportion of healthy versus non-healthy CGS per branch, over time. Ant barrier exclusion trees (Tanglefoot). Coccinellids (LB) were excluded from the trees with the use of mesh 2mm sleeves. All values are means +/- 1 standard error.

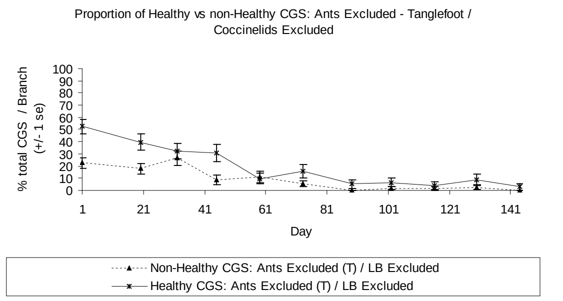


Figure 11.7.10: Mean proportion of healthy versus non-healthy CGS per branch, over time. Ant bait exclusion trees (Amdro). Coccinellids (LB) were excluded from the trees with the use of mesh 2mm sleeves. All values are means +/- 1 standard error.

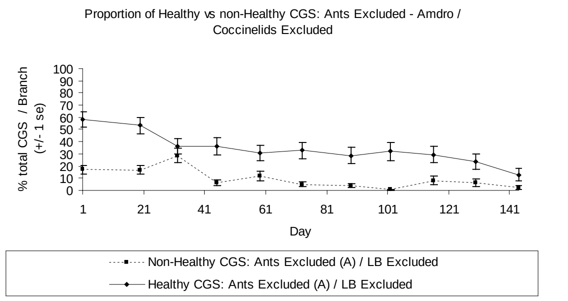


Figure 11.7.11: Mean CGS infestation level per branch over time. Comparison between control (ants present), Tanglefoot (ant barrier exclusion) and Amdro (ant bait exclusion) treatments. Coccinellids (LB) were excluded from the trees with the use of mesh 2mm sleeves. Artificial shade was also used. All values are means +/- 1 standard error.

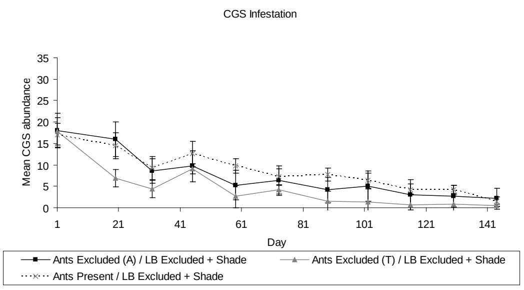


Figure 11.7.12: Mean proportion of healthy versus non-healthy CGS per branch, over time. Control trees, ants present. Coccinellids (LB) were excluded from the trees with the use of mesh 2mm sleeves. Artificial shade was also used. All values are means +/- 1 standard error.

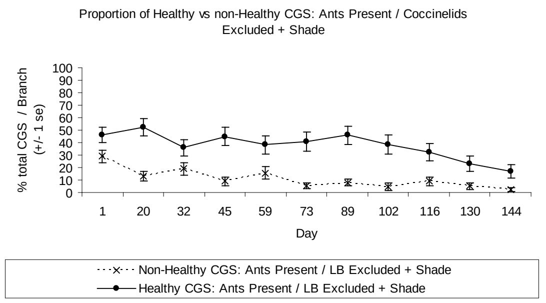


Figure 11.7.13: Mean proportion of healthy versus non-healthy CGS per branch, over time. Ant barrier exclusion trees (Tanglefoot). Coccinellids (LB) were excluded from the trees with the use of mesh 2mm sleeves. Artificial shade was also used. All values are means +/- 1 standard error.

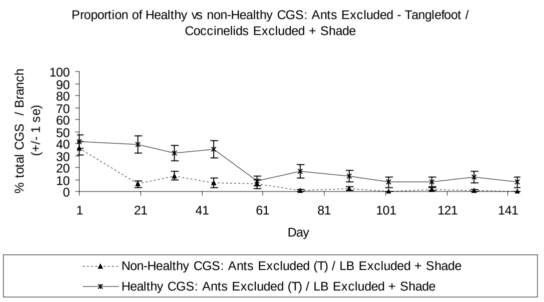


Figure 11.7.14: Mean proportion of healthy versus non-healthy CGS per branch, over time. Ant bait exclusion trees (Amdro). Coccinellids (LB) were excluded from the trees with the use of mesh 2mm sleeves. Artificial shade was also used. All values are means +/- 1 standard error.

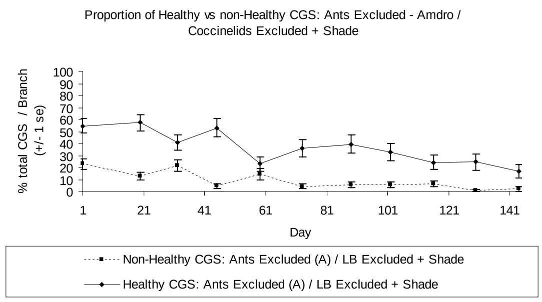


Figure 11.7.15: Mean CGS infestation level per branch over time. Comparison between control (ants present), Tanglefoot (ant barrier exclusion) and Amdro (ant bait exclusion) treatments. Artificial shade was used. All values are means +/- 1 standard error.

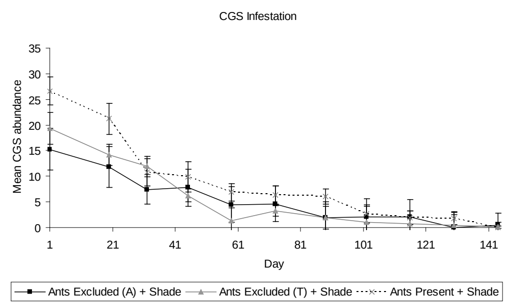


Figure 11.7.16: Mean proportion of healthy versus non-healthy CGS per branch, over time. Control trees, ants present. Artificial shade was used. Artificial shade was also used. All values are means +/- 1 standard error.

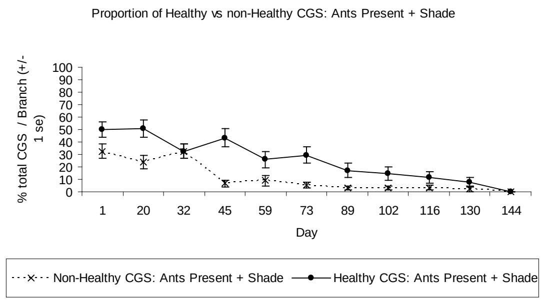


Figure 11.7.17: Mean proportion of healthy versus non-healthy CGS per branch, over time. Ant barrier exclusion trees (Tanglefoot). Artificial shade was used. All values are means +/- 1 standard error.

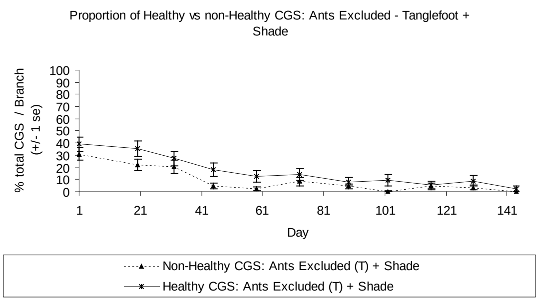
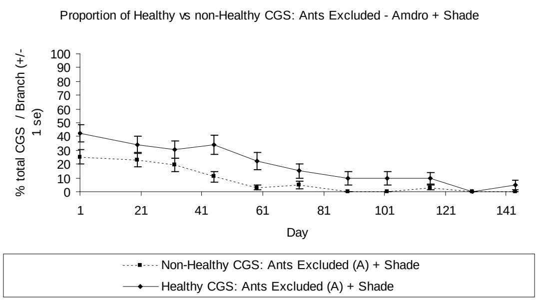


Figure 11.7.18: Mean proportion of healthy versus non-healthy CGS per branch, over time. Ant bait exclusion trees (Amdro). Artificial shade was used. All values are means +/- 1 standard error.



11.8 Appendix H: Infestation of coffee by CGS and the role of ants

Introduction

Anecdotal field observations have recorded the transportation of CGS by ant species. The mutualistic relationship between ants and CGS may encompass the movement by ants of CGS to differing parts of the plants, or new plants. As yet there have been limited studies into the role that ants play in facilitating the process of CGS infestations via transportation. The question is; do ants facilitate CGS infestation of Coffee Arabica by CGS?

Ho: Ant species facilitate the movement of CGS to new plants.

H1: Ant species do not facilitate the movement of CGS to new plants.

Methods

Plants were chosen from the CIC nursery representing clean plants, infested plants without ants, and infested plants with ants. Each seedling was re-potted within a plastic pot of approximately 15cm diameter with a depth of 15cm. Plants were placed in a large plastic bowl (circa 25cm diameter) with the rim smeared with tangle foot. These were placed within a sealed mesh cage. Clean plants were paired with either CGS plants with ants or without ants, i.e. two plants per cage. A wooden bridge (30cm long, 4mm diameter) was then placed between the top green shoots of each plant potentially allowing CGS and ants access to the clean plant. Table 11.8.1 shows the experimental design.

Table 11.8.1: Experimental design of ant facilitation of CGS infestations of coffee seedlings

Clean Plant – no ants no CGS	CGS infested plant with ants
Clean Plant – no ants no CGS	CGS infested plant with no ants

During monitoring the presence of CGS on the clean plants was recorded. As CGS reproduces, any CGS found on clean plants were counted and then removed. Thus only CGS originally from the infected plants was counted and not their progeny. This removal allows the rate of infestation from the infected plants to be determined.

In total three species of ant were used in three separate experiments, these were: *Technomyrmex albipes* (white footed ant), *Pheidole megacephala* (African big-headed ant) and *Anoplolepis gracilipes* (yellow crazy ant). Monitoring of CGS and ants took place at regular intervals, initially every day for the first week.

Results

The rate of addition of CGS to clean plants was measured and compared between ants present and ants absent. Figure 11.8.1 to Figure 11.8.3 clearly shows that the presence of ants does not alter the rate at which CGS infest coffee tree seedlings. Repeated measures ANOVA showed no significant differences between the presence and absence of ants in the rate of infestation of coffee seedlings (see annex 11.8.5 for statistical results). This was the case for all three species.

Figure 11.8.1: Mean cumulative juvenile CGS infestation in the presence and absence of *Technomyrmex* sp. All values are log n+1, +/- 1 standard error.

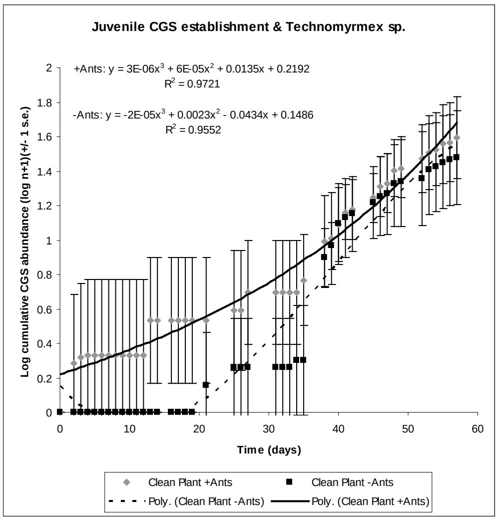


Figure 11.8.2: Mean cumulative juvenile CGS infestation in the presence and absence of *Pheidole* sp. All values are log n+1, +/- 1 standard error.

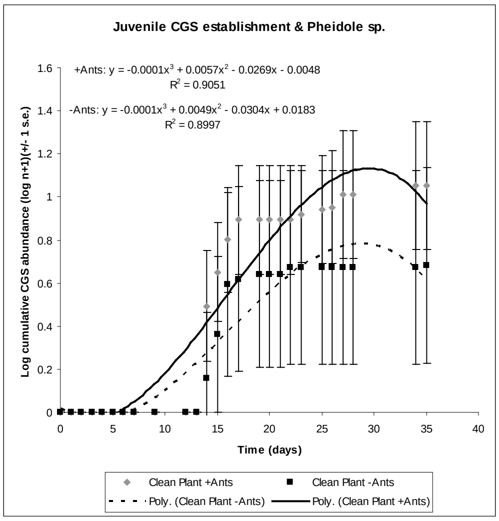
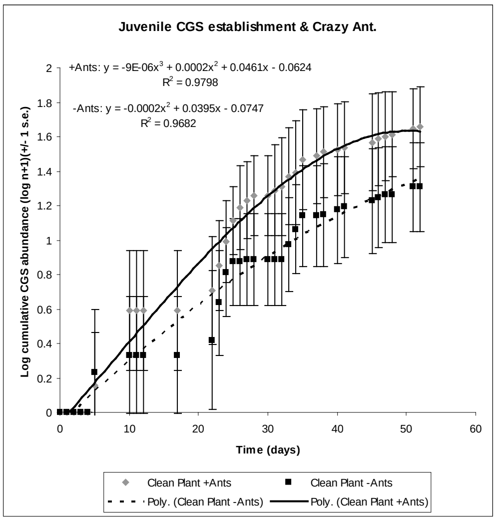


Figure 11.8.3: Mean cumulative juvenile CGS infestation in the presence and absence of *Anoplolepis* sp. All values are log n+1, +/- 1 standard error.



Discussion & conclusion

In all three ant species no significant differences were found in the rate of spread from infested to clean plants in the presence or absence of ants. Transportation of CGS as noted in the field may be occurring but at such low levels that it does not enhance infestation rates. As we know from the ant exclusion experiments, ants do reduce the mortality of CGS and are therefore more likely to be indirectly, rather than directly, enhancing the spread of CGS, by protecting them from natural enemies. This implies that the presence of the invasive exotic ant species benefits CGS by protecting them from natural enemies but they do not actively transport CGS. As a consequence the spread of these species may enhance CGS survival and thus indirectly encourage the spread of CGS.

It should be remembered that due to materials available it was only possible to have three replicates for each of the experiments.

Annex

Table 11.8.2: Juvenile CGS infestation levels per tree over time. Comparison between presence and absence of the ant species *Tecnomyrmex* sp. using repeated measures ANOVA. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.					
Fixed term	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	1	4.66272	4.66272	2.5	0.189
Residual	4	7.46774	1.86694	51.84	
Day	42	69.25322	1.64889	45.79	<0.001
Treatment.Day	42	2.05324	0.04889	1.36	0.091
Residual	168	6.05021	0.03601		
Total	257	89.48713			
Table 11.8.3: Juvenile CGS infestation levels per tree over time. Comparison between presence and absence of the ant species *Pheidole* sp. using repeated measures ANOVA. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.					
Fixed term	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	1	1.06764	1.06764	0.73	0.442
Residual	4	5.8657	1.46643	29.48	
Day	25	23.03502	0.9214	18.53	<0.001
Treatment.Day	25	0.84165	0.03367	0.68	0.868
Residual	100	4.97347	0.04973		
Total	155	35.78348			
Table 11.8.4: Juvenile CGS infestation levels per tree over time. Comparison between presence and absence of the ant species Crazy Ant (*Anoplolepis* sp.) using repeated measures ANOVA. Any significant differences are where p ≤ 0.05. All data was log10 n+1 transformed.					
Fixed term	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	1	3.38411	3.38411	3.06	0.155
Residual	4	4.42574	1.10643	48.34	
Day	32	51.00574	1.59393	69.64	<0.001
Treatment.Day	32	0.9655	0.03017	1.32	0.143
Residual	128	2.92958	0.02289		
Total	197	62.71065			

11.9 Appendix I: Impact of artificial shade on CGS abundance

Introduction

In March 2008 a manipulative shade study was planned and started in late 2008, at CIC Aiyura. The aim was to assess the influence of artificial shade levels on the development of CGS on seedlings, mimicking the variety of artificial shade found at nurseries.

Methods

Simple wooden frames were constructed to allow netting of different shade levels to be applied over coffee seedlings. Three levels of shade were used, 0% shade, 50% shade and 70% shade.

Light levels outside and inside the cages were measured using a light meter. All light levels were measured in Klux units. It was noted that the meter expresses units in both Klux and lux depending on the amount of light, e.g. it measures in Klux for high light values and lux for low light values, e.g. 29.21 Klux = 29210 lux. Table 1 shows the percentage light of each cage.

Table 11.9.1: Experimental design of effects of artificial shade on coffee growth.

Number of Cages	Shade Category	% light inside cage
10	0% Control	100% (no cage)
10	50% shade	49.77%
10	70% shade	31.26%

CGS infested plants were acquired from the CIC nursery and randomly allocated the three shade levels. Cages were laid out in a randomised design near the CIC Aiyura laboratory to avoid interference from local people. Each cage was placed at least 1m apart from the neighbouring cage in order to limit shading of one treatment by another. Plant height (cm) and CGS abundance measurements were collected approximately every two weeks for 204 days.

Results

Plant growth, as measured by plant height, was not significantly affected by increased shade levels over time, Figure 11.9.1 (Table 1: Treatment × Time interaction $p=0.144$). However, there were differences in height between the shade levels overall, indicating different initial plant sizes. Figure 11.9.2 shows mean CGS abundance under differing shade levels. High shade appeared to show significant increases in CGS abundance but this did not take into account the differences in plant size (see Table 11.9.2 and Table 11.9.3 statistical results). The fine mesh artificial shade used would have acted as a barrier to natural enemies.

Figure 11.9.1: Coffee seedling growth under different levels of artificial shade: control = no shade, 50% shade, 70% shade. Growth was measured by using plant height (cm). Data was log transformed and displayed with mean values with estimated standard errors.

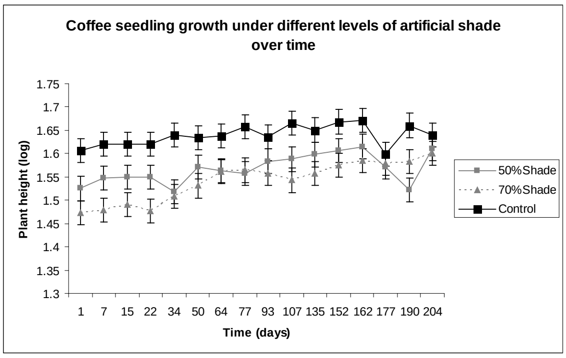


Figure 11.9.2: CGS abundance under different levels of artificial shade: control = no shade, 50% shade, 70% shade. Total CGS abundance was measured by counting all scales present. Data was log n+1 transformed.

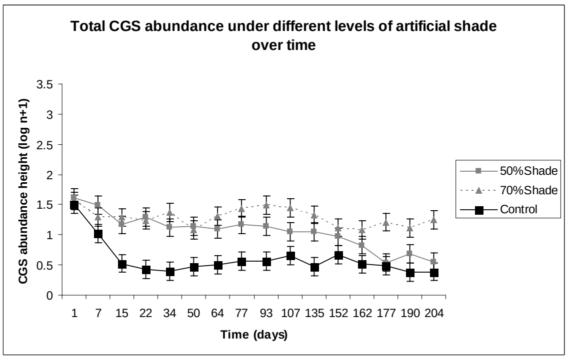


Table 11.9.2: Repeated Measures Analysis of variance: Variate: plant height					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block	9	1.67422	0.18602	1.21	
Treatment	2	2.45353	1.22677	7.95	0.003
Residual	18	2.777	0.15428	1.05	
Time	15	1.00193	0.0668	6.02	<0.001
Treatment.Time	30	0.49518	0.01651	1.49	0.144
Residual	1303	14.44965	0.01109		

Table 11.9.3: Repeated Measures Analysis of variance: Variate: CGS abundance					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block	9	238.5914	26.5102	3.59	
Treatment	2	118.8762	59.4381	8.05	0.003
Residual	18	132.9251	7.3847	2.24	
Time	15	59.5506	3.97	17.99	<0.001
Treatment.Time	30	27.0054	0.9002	4.08	<0.001
Residual	1303	287.4688	0.2206		

11.10 Appendix J: Soil nutrient experiment – effect of SOA on CGS abundance

Introduction

Within crop production, improved plant nutrition changes many plant characteristics, e.g. leaf area and number of stems. These changes may facilitate pest infestations both directly and indirectly. Currently, seedlings grown at the CIC nursery have inputs of sulphate of ammonia, (($(NH_4)_2SO_4$; 21% N & 24% S) (SOA). Here we trial differing fertiliser regimes on coffee seedlings and the responses of CGS to the soil nutrients.

In October 2007 it was planned to use an existing fertiliser trial to examine the influence of soil fertility on CGS development. However, given lack of knowledge of previous history of the experimental fields, it was proposed to carry out a small-scale study within a greenhouse. Primarily the aim was to investigate the effects of soil nutrients on CGS on coffee seedlings.

Methods

Glasshouse experiments were conducted at CIC research facility at Aiyura, EHP, PNG during 2008. Within the greenhouse coffee seedlings were assembled into fully randomised treatment blocks with seven replicates of two nitrogen treatments.

Nitrogen treatments

The seedlings preceding the experiment were given zero fertiliser inputs. The nitrogen treatments used were: two levels of SOA, 40 kg/ha, 80 kg/ha, and a control of zero input. The standard field rate used at Aiyura is 80 kg/ha. Each replicate consisted of five plants grown in circa 13cm pots. These were placed on individual double layered plastic sheets so as to stop any run-off from other treatments influencing the experiment. Fertiliser was applied at regular intervals according to standard CIC field practice.

Plant measurements

Clean plants were grown in plastic plots and plant growth measurements were made prior to CGS infestations and the application of treatments. After treatment application these measurements were repeated when scales were recorded.

CGS assessments

CGS populations were assessed prior to treatments being imposed, to establish baseline numbers. After treatment imposition, populations were assessed on a weekly basis. On each sampling date, on each plant, measurements were taken in a non-destructive manner. CGS were counted and the number killed by entomopathogenic fungi, the number parasitised and any natural enemies were recorded.

Results & discussion

Plant growth, as measured by plant height, was not significantly increased by the addition of SOA over time, Figure 11.10.1 (Table 11.10.1; $p=0.216$). There were no significant differences in height between the SOA treatments. Figure 11.10.2 shows CGS abundance in the differing SOA levels; there were no significant effects of SOA on CGS abundance.

In this experiment it does not appear that soil nutrients are important in CGS development. However, the plants did not appear to grow over the period of 427 days which is a questionable finding. It would be expected for plants to grow over such a length of time irrespective of nutrient addition. These results should be treated with caution.

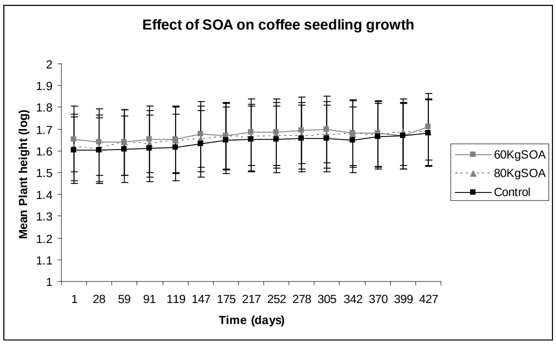
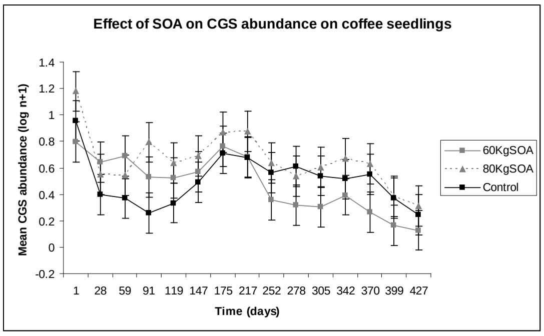
Figure 11.10.1: Effect of SOA on coffee seedling growth. Mean plant height, log n+1 transformed (+/- 1 s.e.)	Figure 11.10.2: Effect of SOA on CGS abundance over time. Mean CGS abundance, log n+1 transformed (+/- 1 s.e.)
	

Table 11.10.1: Repeated Measures Analysis of variance: Variate: plant height					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block	6	0.222596	0.037099	0.65	
Treatment	2	0.275575	0.137787	2.43	0.13
Residual	12	0.681266	0.056772	0.98	
Time	14	0.777302	0.055522	29.72	<0.001
Treatment.Time	28	0.07045	0.002516	1.35	0.216
Residual	1422	2.656383	0.001868		
Total	1568	9.493043			
Table 11.10.2: Repeated Measures Analysis of variance: Variate: CGS abundance					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block	6	252.1254	42.0209	5.12	
Treatment	2	10.3002	5.1501	0.63	0.55
Residual	12	98.4405	8.2034	4.69	
Time	14	47.692	3.4066	12.15	<0.001
Treatment.Time	28	14.2862	0.5102	1.82	0.057
Residual	1428	400.312	0.2803		
Total	1574	970.0774			