

An alternative baiting method of Yellow Crazy Ants (*Anoplolepis gracilipes*) on Christmas Island, Indian Ocean

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Abstract

The Aim of the research project was to trial a low toxicity bait containing Boron in the form of Disodium octaborate tetrahydrate (DOT) in a new bait delivery system to determine the baits effectiveness in controlling Yellow Crazy Ant colonies of high to low densities, and to test whether the new bait delivery system has minimal impact on non-target species, especially native and endemic vertebrates and medium-sized and large invertebrates on Christmas Island in the Indian Ocean. Three trial and control sites were identified. The sites were selected according to the density of Yellow Crazy Ants from high to low density. All sites were monitored for six weeks prior to baiting to ensure there were no impacts on native species. Ant counts for density analysis and monitoring of other invertebrates was undertaken at each site. Positive results were obtained with a decline in Yellow Crazy Ant density in all three baited sites compared to the control sites. Yellow Crazy Ants were absent from the Greta baited site for a period of more than 9 months, indicating possible local area eradication. From the results it is very clear that the bait has had a significant impact on Yellow Crazy Ants. As Yellow Crazy Ant density decreased in the baited area there was an increase in density of Yellow Crazy Ants in each of the controls. Analysis of the data has shown strong evidence that boric acid is an effective bait for the control of Yellow Crazy Ants, and that it is a safer and significantly more environmentally friendly approach to control Yellow Crazy Ants compared to the present fipronil baiting regime on Christmas Island.

Keywords: Boric acid, bait stations, synthetic honeydew, non-target species, fipronil.

1. Introduction

Christmas Island is an Australian Territory located in the Indian Ocean (Figure 1) 500km south of Jakarta and 2800 km north west of Darwin (10°30'S 105°40'E). The island is approximately 135 sq km in area and is the summit of a marine mountain of volcanic origin with a central plateau at 361 metres above sea-level. The main economic activity on the island is phosphate mining and 63% of the island has been set aside as a National Park.

There are 54 recognised tramp ant species on Christmas Island with no endemic species (Framenau and Thomas, 2008). Of these different ants the Yellow Crazy Ants (*Anoplolepis gracilipes*) (YCA) representing the greatest threat to Christmas Island's biodiversity (Thomas, Becker, Abbott and Feldhaar, 2010). A major concern is the threat they pose to the Red Crab (*Gecarcoidea natalis*), which play a key role in the maintenance of the island's forest ecosystem. The crab population size has been reported to be approximately 120 million in 1990's (Environment Australia, 2011). The Christmas Island National Park (CINP) survey data indicates that the Red Crab population has declined by 25-50% to about 43 million in 2001 (Adamczewska and Morris, 2001) and that numbers have continued to decline since then with between 10 and 15 million of the crabs having been killed by YCA over the past several years (O'Dowd *et al*, 2003). Populations of many vertebrate species such as the Christmas Island Pipistrelle (*Pipistrellus murrayi*) and the Blue-tailed Skink (*Cryptoblepharus egeriae*) have also decreased significantly over the same period (Algar *et al*, 2011) while several species are now listed under the Environmental Protection and Biodiversity Conservation Act (EPBC Act) as critically endangered. Of particular concern is the rapid decline and acknowledged extinction of the Pipistrelle(Lumsden, 2009; Beeton *et al*, 2010; Algar *et al*, 2011), as the last known living individual was last recorded in late 2009 with none of these bats recorded since then (G. Richards *pers com*, 22nd November 2010).



FIGURE 1: Location of Christmas Island in the Indian Ocean, which is 500km south of Jakarta, and 2800km north-west of Darwin.

As a result of their impact on the environment and ability to form multi-queened colonies with high ant densities referred to as “supercolonies”, YCA control is a priority conservation management objective of CINP. An aerial baiting program conducted in 2002 dropped 12 tonnes Fipronil bait over 2400 ha of the island (Parks Australia North, 2002). Annual follow-up ground surveys and local baiting have been conducted since then. Despite this intensive baiting campaign, the distribution of YCA’s has continued to increase (DSEWPC. 2010). Supercolonies now occur throughout the island although the overall YCA density has decreased in most areas (CINP Referral July 2009). Due to this increase in YCA numbers and density on Christmas Island a further aerial baiting program was undertaken by CINP where an additional 8 tonnes of AntOff Ant Bait was dropped during September 2009 over 2000 hectares (CINP Referral July 2009), at a rate of 4kg/ha containing 0.01g/kg Fipronil. The number of mortalities due to the baiting operation on non-target species (NTS) are not known by the authors, however it is understood that Robber Crabs were used as a surrogate to assess Fipronil impacts on NTS.

Fipronil is a non-selective insecticide that effectively kills YCA however there is evidence that YCA may return to previously colonised areas (personal observations). This probably occurs because baiting is conducted only on supercolonies. At supercolony densities it is assumed that fipronil baiting of supercolony densities will not impact negatively on Red Crabs or other non-target species (NTS) because the ants have already killed or displaced them. Baiting is not conducted in the adjacent lower ant density areas because Fipronil could impact non-target-species. These sites therefore provide the source of founder ants to build up to supercolony densities, and/or reinvade the previously occupied super-ant colony sites. Although success is achieved to some extent with the baiting of Fipronil, it is important that other techniques are also tested and evaluated to produce more ecologically friendly results that could have less of an impact on the total ecosystem especially non-targeted species.

This paper is aimed at describing an alternative non-toxic baiting system using Boron-containing bait in a new bait delivery system to control YCA colonies of low and high densities and to test whether the new bait delivery system has minimal impact on non-target species, especially native and endemic vertebrates and medium-sized and large invertebrates. The trial described in this paper used 1% disodium tetrahydrate octaborate (DOT) in a solution of synthetic honeydew (Gourmet Liquid Ant Bait (GLAB)), which mimics the honeydew produced by scale insect. It was delivered in KM AntPro® ant bait stations, which have a small gap through which only ants can enter, which in turn were housed inside wire-cages to prevent disturbance by other fauna such as the large and powerful robber crab. It is proposed that this approach to controlling YCA is a safer as the DOT has no known adverse effects on the environment and is thus potentially a more attractive control method than Fipronil. When placed in an aqueous solution DOT breaks down into borax and boric acid, substances that are benign and common in the environment (Argust, 1998; Gentz and Grace 2006).

Pre-baiting monitoring commenced at the start June 2009 and ran for three weeks, followed by a four week sugar bait trial and baiting using DOT commenced in the last week of July 2009 at all three sites (dates as stated under Results below). The trial continued to the end of December 2010, with a post-baiting monitoring phase that run for four months from January to the End of April 2011. The trial ran for a total of 22 months.

2. STUDY SITES

An on-island survey was conducted from 4 January to 21 January 2009 to identify suitable ant bait trial sites. Three sites were identified (Figure 2) and shown to the Attorney General's Department (AGD). During the field trips YCA colonies with low, medium and high density ant colonies were identified. The standard EPBC referral process was followed and an application made to the Department of Environment, Water, Heritage and the Arts to undertake such a baiting trial (EPBC 2009/4763). A decision was made that the action was not a controlled action under the EPBC Act (1999) and that the baiting trial using disodium octaborate tetrahydrate may proceed with certain additional provisions. An initial pre-baiting trial using sugar water and Arenga palm commenced on 10 June to 29 July 2009 (6 weeks) so that cages designed to protect bait stations from damage by Robber Crabs could be evaluated.

The ant baiting trial sites were:

2.1.1. Smith Point Trial Site A.

This site had a high density YCA colony in scrub forest (DSEWPC, 2011a), which included exotic vegetation in disturbed areas along the track. There was evidence of high Red Crab mortality caused by YCAs.

2.1.2. Greta Beach Trial Site B.

This site had low to medium density YCA colonies in coastal terrace vegetation with Arenga/terrace forest (DSEWPC, 2011b) species that showed evidence of scale insects in the canopy due to sooty mould present on surrounding vegetation. There were resident Christmas Island Thrush, and Robber and Red Crabs noted near or within the site.

2.1.3. South Point Trial site C.

This site had a low to medium density YCA colony with a high density of Red Crabs, and was situated on the secondary limestone terrace within open forest (DSEWPC, 2011c). There was evidence of dieback among tall evergreen trees and scale insects were present on a number of trees. There were resident birds such as Red Footed Booby, Brown Booby, Christmas Island Thrush, plus Robber and Red Crabs.

Other ant species were noted within the area, with one unknown species carrying scale insects into the trees.

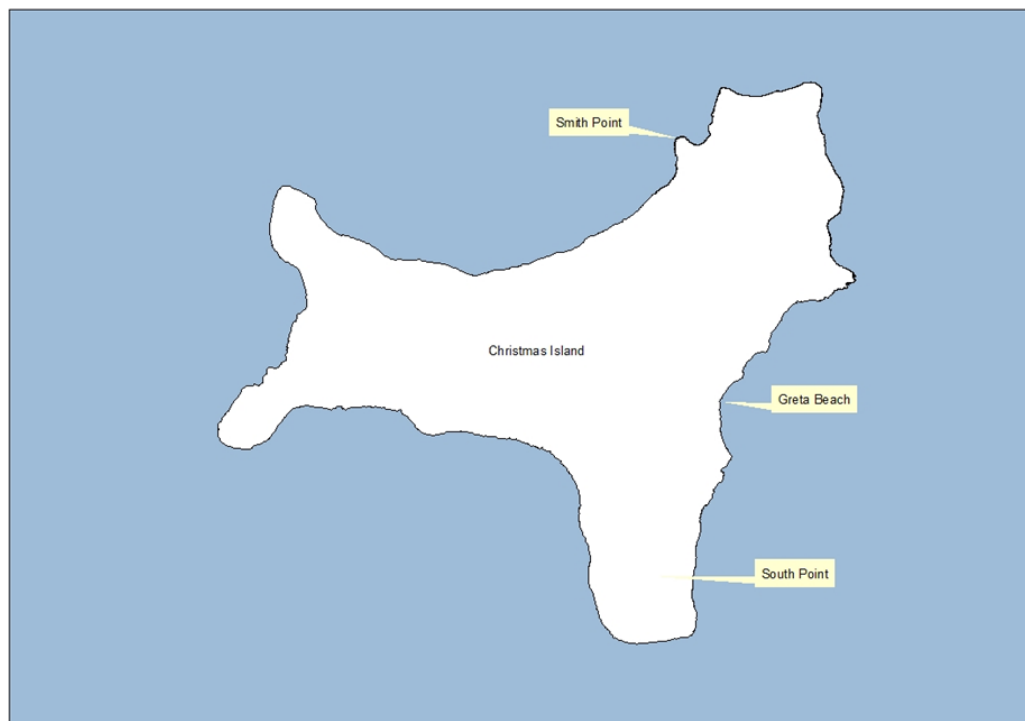


FIGURE 2: The location of the three trial sites on Christmas Island. Site A at Smith Point on the northern side of the island, Greta Beach area on the east coast and South Point on the south east coast of the Island.

3. Methods

A total of 200 bait stations were placed 10 metres apart (200m x 200m over an area of 0.04 ha) for sites B and C and 5 metres apart (100m x 100m over an area of 0.02 ha) for site A, All bait stations were set along a predetermined grid of “best fit”. A fixed coordinate start point was identified, and an adjacent similar-sized area was established as an untreated control area at each study site.

The KM AntPro® bait stations were obtained from KM AntPro® LLC PO Box 967 Nokomis KL 34275 USA. Steel mesh cages (gauge 3mm, approx. 300x300x300mm, mesh square 25mm) were manufactured in the CIP workshop and overseas. One bait dispenser was placed in each cage. In the field, the bait dispenser was leveled and pegged to the ground and the mesh cage secured to a star picket or tree trunk. The cages had lockable lids which enabled operators to refill poison in the dispensers when required.

3.1. Setting KM AntPro® Bait Stations and bait requirements

KM AntPro® bait stations were placed in metal mesh cages to prevent access by non-target species such as Robber Crabs and were fixed into place by spike, zip-tie or strap to prevent removal and ensure secure anchorage. Where vegetation was required to act as an anchor, care was taken to ensure no damage would be sustained by the plant due to strapping or ties used. No nails or invasive anchor methods were used. Bait was stored and transported in 1L containers to reduce total spill volume should this unlikely event have occurred. KM AntPro® bait station recharge or refill was accomplished by using 1L refill plastic bottles. All refill/recharge 1L bottles were recorded when leaving the store and on return to the store to ensure no bottles were left in the field. Provision was made for all accidental spills or leaks to be recorded and reported to CINP and all other appropriate island authorities. No spills occurred during the trial. The number, location and use of KM AntPro® bait stations were recorded in the field. In general, 600 sequentially numbered KM AntPro® bait stations, spaced at 10m and 5m were used at three field sites. Three additional control sites were established adjacent to each trial site. The control sites were setup to ensure that if there was a dramatic population crash in the treated (poisoned) site, which this was not due to some other (unknown) factor(s).

Methods to monitor ant density were similar to those used by CINP, viz: A 50m transect was established within each of the replicates (giving a total 6 transects). Along each 5m of a transect, litter was cleared with one hand swipe, a plastic card with inscribed 10 x 10cm squares, was placed on the cleared ground, the observer waited 15s, then ants were recorded on one randomly selected square for a period of 30s, Initially, ant densities were recorded on the 6 transects weekly for 2 weeks prior to baiting and then every month following the baiting, with further monitoring for 6 or so weeks after bait was removed.

Ultra-violet light traps developed by Australian Entomological Supplies for collecting night-flying insects were used to monitor invertebrate densities. A trap was placed at a fixed point in each of the paired trial (baited) and control (unbaited) sites at each study area, and samples were collected monthly. Further back-to-back sampling of invertebrates at two fipronil aerial baited sites and the trial and control sites at South Point; were taken over two nights for a comparative study of invertebrates.

4. Results

4.1 General summary

The data indicate a positive decline in ant density in all three baited sites compared to the control sites. YCA were absent from the Greta trial site from 19 August 2010 to April 2011, a period of 9 months. Smith Point data also show a reduction of YCA in the trial site ant density compared to that in the control site, from 21 October 2010 to the end of December 2010. As YCA density decreased in the baited area there was a significant increase in YCA in the control. There was also a reduction in YCA density at the South Point trial site, with a marked increase in YCA counted within the control site. It thus seems that the bait

has had a negative impact on YCA within the baited areas at each site. Further discussion on each site follows.

4.2. Smith Point (trail site A)

There have been no recorded or observed impacts or mortalities to any native invertebrate or vertebrate non-target-species. No Robber Crab damage or breakage of any bait station, or cages was observed. Disodium octaborate tetrahydrate bait 1% in solution (Gourmet Liquid Ant Bait) was introduced into the bait stations on 30 July 2009. A 14 day pre-baiting trial from 30 June to 13 July was carried out where sugar water and Arenga Palm pieces were used as lures to identify if any non-target-species would be attracted to the sugar bait or were able to impact or damage the bait stations or cages within the trial site. In August 2010 the bait stations were found to be contaminated with a mould, which resulted in the YCAs refusing to take any bait. Consequently all 200 hundred bait stations were replaced with new ones and filled with fresh bait. Following bait replacement, a marked increase in YCA activity was observed in and around the bait stations. It appeared that increased bait uptake was correlated with the frequency of bait replacement. Recordings of ant density over time are shown in Figure 3.

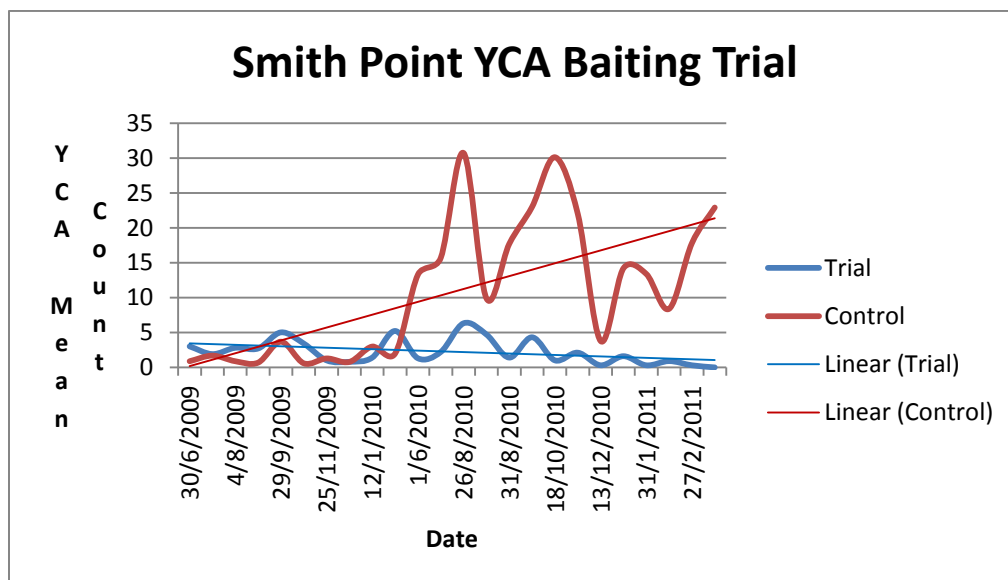


FIGURE 3: Mean YCA counts in the trial and control sites at Smith Point. Although there was a low decline in YCA numbers over time, it is important to note that YCA exploded almost six-fold in the untreated control area. The bait stations ran empty of bait early in the trial, which allowed YCA to have eliminated all bait from their systems. Further bait was found to be contaminated and had to be replaced, but density nevertheless was suppressed well below that of the control area.

During the trial period YCA numbers in the control site increased from a mean of 2 to over 30 per sample, which is an exponential growth rate of YCA in the non-baited site. In comparison the trial or baited site data indicate a decrease from a mean number of YCA of over 6 to less than 1 (>1). This was a significant difference in reduction in ant density and significant between the treated and control sites (FIGURE 4).

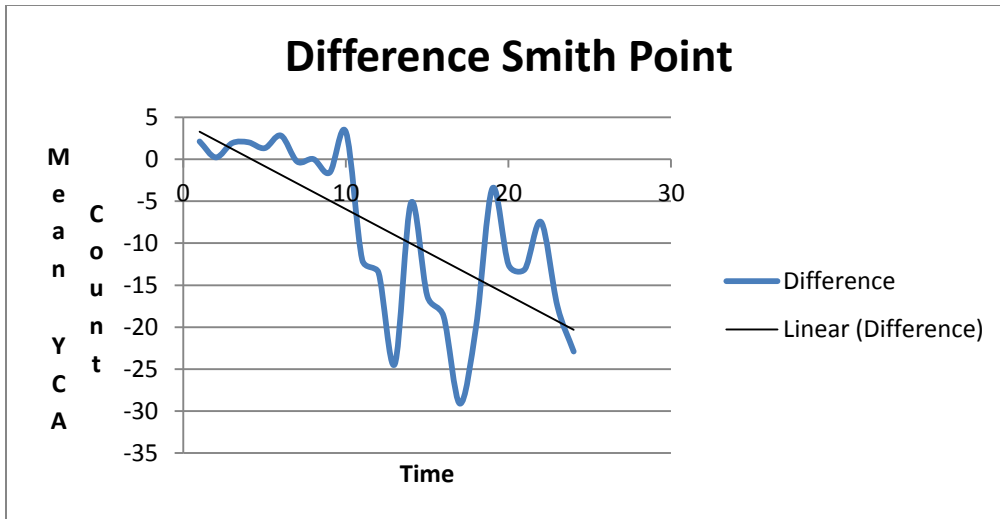


FIGURE 4: Difference between the mean YCA trial and control count over time, where mean control sample was subtracted from mean trial sample to indicate differences at any one point in time during the baiting. A linear regression through the samples shows the progressive decrease in YCA density in the baited area at Smith Point.

Analysis of insect data showed a moderate to high positive trend in weight over time in the trial site and invertebrates trapped in the control site (FIGURE 5). It can be seen that where YCA numbers were suppressed through baiting, there was a significant increase in invertebrate populations in the period December 2009 to January 2010, assumed to be from successful breeding during the wet season. Notably, at the same time, invertebrate numbers decreased a response likely to be a result of predation by YCA of eggs, larvae and pupae in the control.

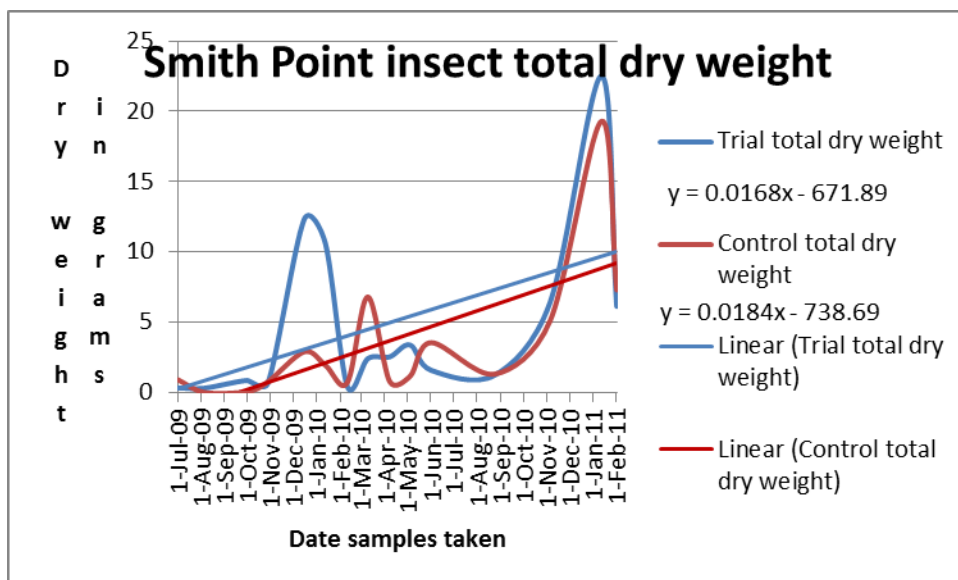


FIGURE 5: Insect dry weight showing a strong positive trend in invertebrate weight that has increased over time within the Gourmet Liquid Ant Bait area compared and the control site.

Correlation between YCA density and climatic conditions may reveal activity trends (an analysis of climate data is currently being undertaken), as field observations recorded a decrease in YCA foraging during wet or rainy periods, with an increase in warm or sunny days.

4.3. Greta Beach (Trial site B)

There were no recorded or observed impacts or mortalities to any native invertebrate or vertebrate non-target-species. Nor was there any Robber Crab damage or breakage of any bait station or cages at this site during the trial.

This site is a known Robber Crab habitat and provided an ideal area to test if the bait station cages could be damaged by this species. Robber Crabs were observed near the Arenga palm placed in the selected cages but they were unable to damage or open any of the cages. Sugar water and Arenga Palm pieces were placed in bait stations and cages for a period of 21 days between 7 and 27 July as lures for non-target-species. No evidence was found that any non-target-species were attracted to the bait stations.

The Greta Beach data showed a marked absence of YCA within the baited area from August 2010, but there was an increase of YCAs in the control (non-baited site) over most of the observation period (FIGURE 6). A spike in YCA numbers occurred in October 2009 in the trial and this thought to be the result of YCA being attracted to the bait. Notably, as with the Smith Point site, when a significant YCA outbreak occurred, as measured on 26 August 2010, YCA densities remained suppressed at zero at the same time.

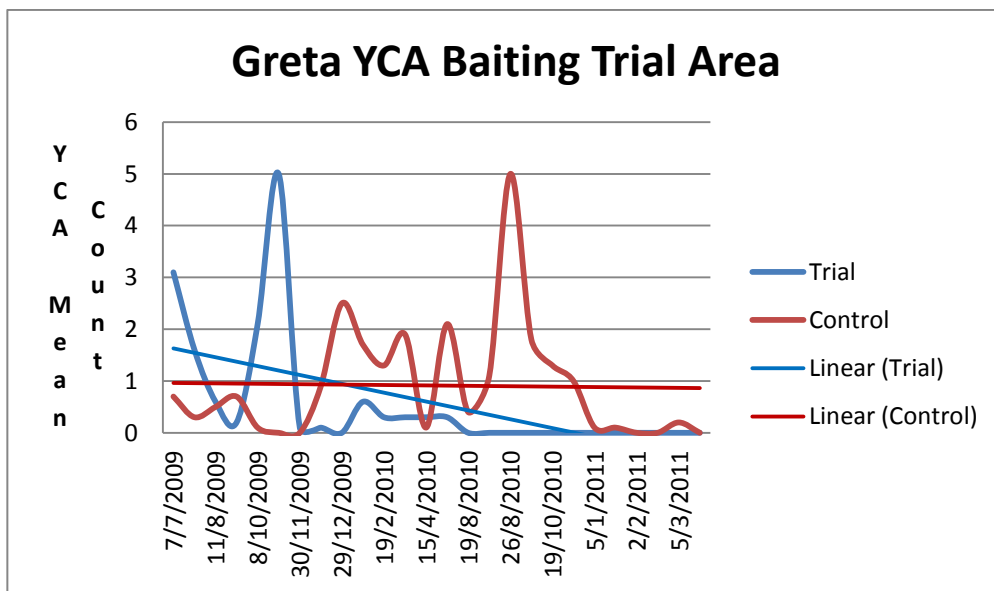


FIGURE 6: The data show a substantial decrease in YCA numbers within the DOT trial site, compared

with an increase in YCA numbers within the control area, showing positive results with the use of Gourmet Liquid Ant Bait.

The difference between the trial and control areas at Greta Beach was also significant (FIGURE 7). A noticeable increase in the number of Red Crabs was recorded (both adult and juvenile) in the treated site. Observations of Robber Crabs, Red Crabs and Blue Crab populations indicated they also increased in density in the treated site (personal observation). In particular there was an observable increase in crab activity and sightings during the migration period, and a substantial number of returning juvenile crabs within 27 days post migration through the trial site. No juvenile crab deaths were recorded in or at any of the bait stations and the returning crabs were not attracted to the bait stations for the Gourmet Liquid Ant Bait.

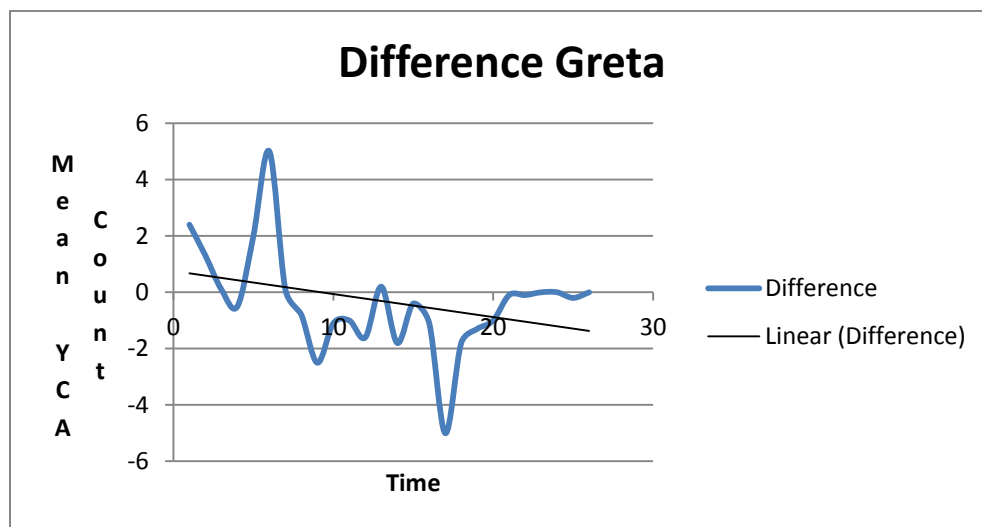


FIGURE 7: Linear regression showing a decrease in YCA density within the trial area using the difference in mean ant counts between the trial and control sites at Greta.

Insect weight data show a corresponding increase in numbers that were light-trapped, both within the trial and control areas (FIGURE 8). There is variation in monthly weight statistics. The trend shows an increase in invertebrate numbers within the trial and the control. There was also an observed reduction of scale insects and an improvement of canopy cover in the treated site (personal observations).

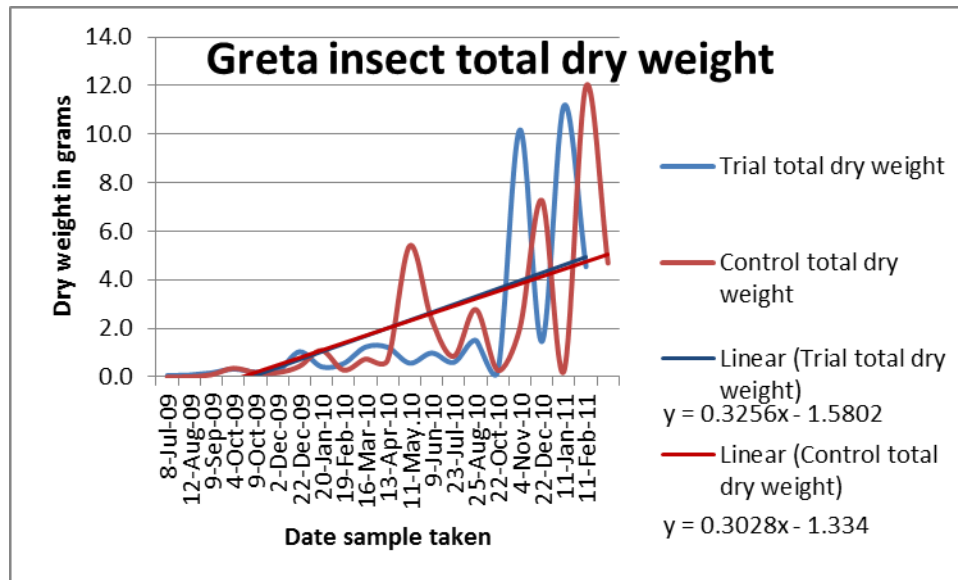


FIGURE 8: Total insect weights at Greta showing the trends for both trial and control areas with a gradual increase in invertebrate weights over time.

With the absence of YCA and an increase in invertebrate numbers occurring within the trial and the reciprocal (opposite) of this within the control, there is strong evidence that boric acid baits work as a control and suppression method of YCA.

4.4. South Point Trial Site C

Pre-trial sugar water and Arenga palm bait was undertaken as required under the conditions of the Referral for 17 days from 13 to 29 July 2009. No non-target-species showed any interest in either bait. Disodium octaborate tetrahydrate bait at 1% solution was placed in the bait stations from 30 July and no non-target-species had been recorded to take any of this bait. There were no recorded or observed impacts or mortalities to any native invertebrate or vertebrate non-target-species. No Robber Crab damage or breakage of any bait station, or cages occurred.

The monitoring data showed a decrease in YCA density (FIGURE 9) within the trial or baited site, with an identifiable increase in crab numbers noted within the trial site. The data show a decrease in YCA mean numbers within the trial over time FIGURE 10, which is similar to results found in the other two trial sites and indicative that the bait is effective in reducing YCA density within these areas.

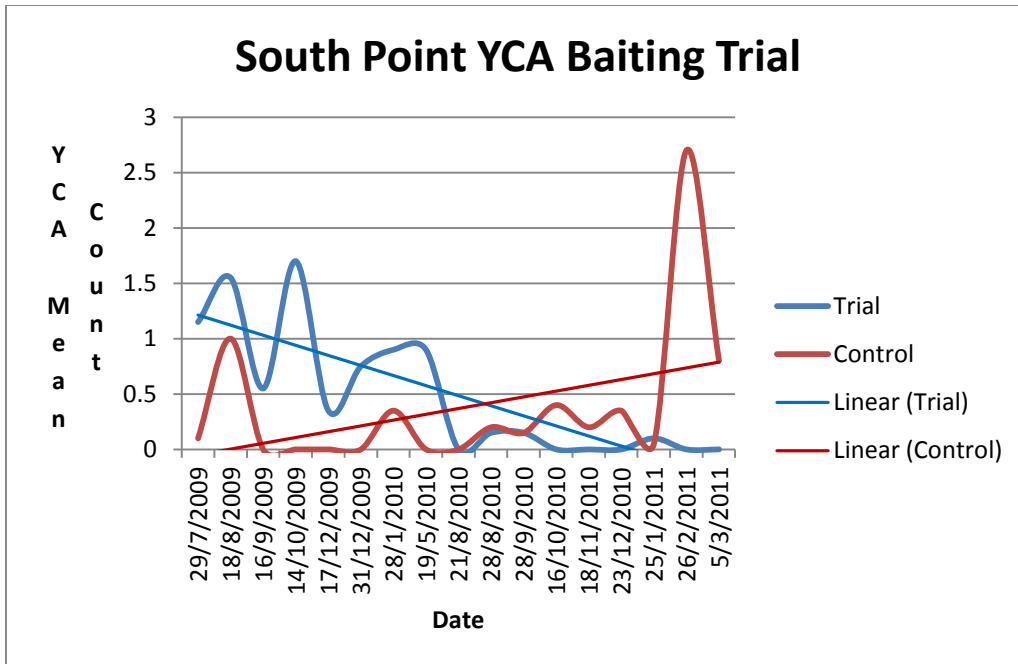


FIGURE 9: Although not obvious from this graph, there was a decreasing trend in YCA numbers within the trial. The control site was not well chosen as there were low YCA numbers compared to the trial, however over time the number of YCA in the control increased.

The insect data show an increasing trend in invertebrate numbers within the trial site (FIGURE 11).

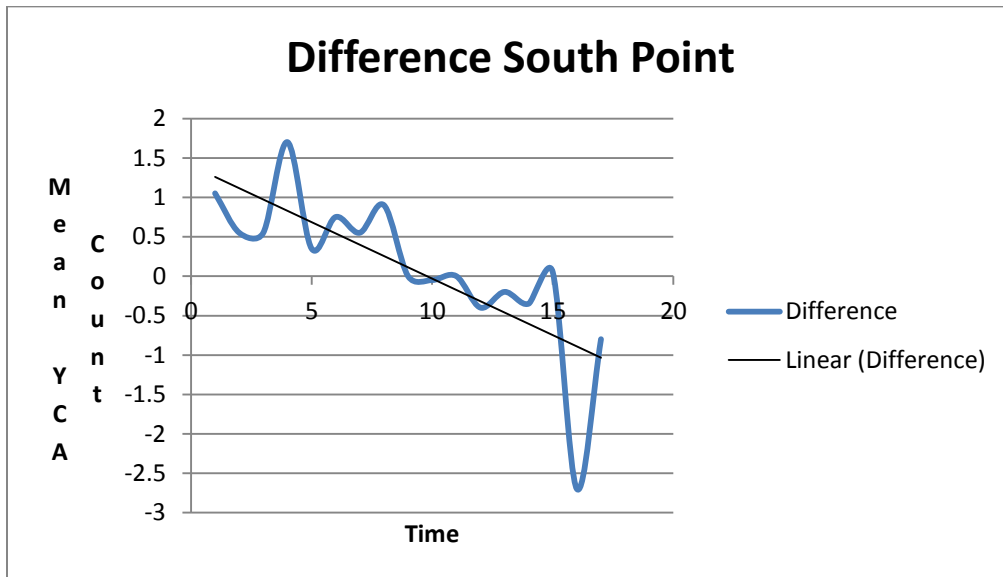


FIGURE 10: Mean YCA count difference between the trial and control sites at South Point showing a decrease in YCA density over time

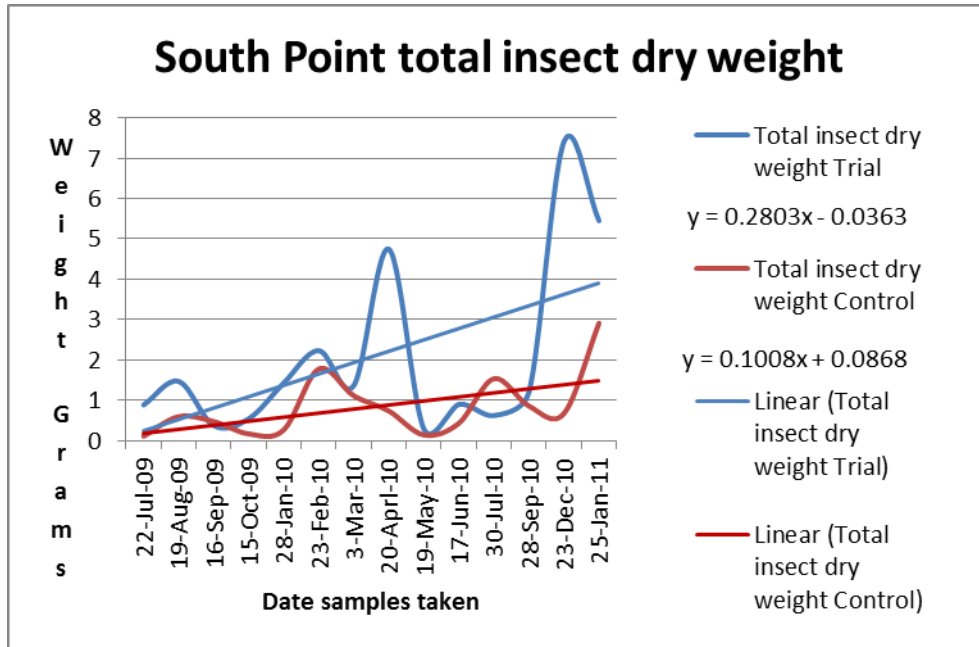


FIGURE 11: Insect dry weight showed an increase in biomass in the South Point trial site, with positive trends (increases) in insect weight with a decrease in YCA numbers over time. The trial site showed the greatest increase in invertebrate weights compared to the control.

Red Crab numbers increased during the monitoring, especially during the migration period, when an increase in Red Crab activity was recorded within both the trial and control area.

4.5 Back-to-back sampling of South Point Trial Site and fipronil aerial baited sites.

During the monitoring period, a number of light-trap samples were obtained from areas that had been aerial baited with Fipronil, at the same time that samples were collected from the trial and control sites. Sample data showed that major differences between invertebrate counts at the trial sites compared to the control sites (almost double) whilst the aerial baited (Fipronil) sites were lower again (TABLE 1). Of note from this sampling is that where Fipronil is used indiscriminately to control YCA, there appears to be a concomitant loss of insect prey available to vertebrates. For example, on 24 March 2010 there were roughly half the number of flying insects at the two fipronil-baited areas than at the South Point control area where YCA were not baited. Further, where boric acid was used with the appropriate lure and in an appropriate delivery system, there were significantly higher numbers of insects available as vertebrate prey.

TABLE 1: Back-to-back sampling of two fipronil aerial baited sites (Fipronil sites 1 & 2 = Fip 1 & Fip 2) compared to South Point Trial and Control areas.

Date of sampling	Trial	Control	Fip 1	Fip 2	
3-Mar-10	681	278	124	0	
24-Mar-10	310	146	73	80	
	Total	991	424	197	80
	mean	495.5	212	98.5	80
	Sum of all	886			
	mean of all	221.5			
	Ratio		2.3	5	6.2
	Percentage value	55.9255079	23.92777	11.11738	9.029345
		0.559255079	0.239278	0.111174	0.090293

5. ADHERENCE TO PROTOCOLS, CONDITIONS, AND REQUIREMENTS

The YCA baiting trial was initiated by Christmas Island Phosphates for two main reasons;

1. To test the viability of bait stations as a safe and environmentally sound method for bait delivery, and
2. To trial a boric acid bait (disodium octaborate tetrahydrate % in solution of a synthetic honey dew) that is more environmentally friendly than Fipronil.

The following measures were taken to ensure that the system being trialed had no adverse effect on the Island ecosystem. Specifically

- i. Listed threatened species and communities.
 - ii. Listed migratory species.
 - iii. Commonwealth land.
1. If any listed, protected or threatened species showed any interest in the bait or would be impacted by any activity associated with the trial, then that trial at that site would cease immediately and all baits would be removed.
 2. If any adverse event occurred it would be reported to the relevant Commonwealth Departments.
 3. Prior to the placement of the boric acid baits the following pre-trial tests would be carried out:
 - a. Arenga palm seeds and pieces be placed in 50 randomly selected bait station cages for 2 weeks to test the cage integrity against Robber Crabs (*Birgus latro*)
 - b. Sugar water be placed in all other bait stations for two weeks
 - c. Bait stations to be checked daily for two weeks determine if any non-target-species were impacted or interested.
 - d. That monitoring of the three sites start one month before commencement of the trials

- e. If there was any interest by non-target species of the sugar water, no boric acid baits would be used.
4. Monitoring for non-target species be undertaken daily for the first two weeks and then weekly for the duration of the trial.

Data monitoring sheets were used throughout the pre-trial and trial periods, where all relevant information and measurements or counts were recorded. All manually recorded data was then transferred to an electronic data base, with the hard copies filed. After the first three months an interim report was drafted and submitted to the Commonwealth Department of Environment, Water, Heritage and the Arts.

All monitoring was in keeping with the project design as submitted to, and accepted by the above Department and all staff involved in the monitoring of the trial were made aware of all the requirements of the project and project design (ANNEXURE 1). Training was provided to staff on the methodology of YCA counts by Parks Australia on island. Co-ordination of the project was by the environmental section of Christmas Island Phosphates.

7. DISCUSSION

Analyses of the data indicate:

- i. There has been a decrease in YCA density in the treated sites at both Smith point and South Point.
- ii. A total absence of YCA at the Greta trial area at the end of the trial.
- iii. An increase in invertebrate numbers occurred in all three trial sites.

There is evidence in the data that show a relationship between YCA density and other invertebrate numbers in that where there has been a reduction in YCA density there has been a corresponding increase in other invertebrate density in general with an increase in invertebrate size classes (Richards. 2010). This finding has potential significance for the survival of any insectivore on Christmas Island through feeding competition between them and YCA (starvation).

The data from this study indicate that boric acid baits suppress YCA, without impacting negatively on other invertebrates, but rather allows for the re-colonisation of native insects in the trial site. Further the data show that there was a decrease of YCA numbers in all three baited sites with an absence of YCA in the Greta area from August 2010.

With reference to the back-to-back sampling at South Point trial and Fipronil aerial baited sites the data to date indicates that the South Point boric acid trial site has a 2.3 fold more invertebrates than the control site and, to 5 to 6.2 times more invertebrates than either the Fipronil aerial baited sites. Analysis of the

preliminary insect data collected at the two aerial baited areas compared to the trial and control areas at South Point trial site show that there may be evidence that Fipronil baited areas have a lag time for re-colonisation by native invertebrates of the area.

The preliminary data from this trial also indicates that in the Fipronil aerial baited sites there is a significant lag in the time before re-colonisation of invertebrates after baiting has ceased. From the study of the two Fipronil aerial baited sites which were baited on 15 September 2009, the data (collected six months after the fipronil baiting) show that between 5 and 6.2 fold more invertebrates are found within the DOT baited trial site in South Point, compared to that baited with Fipronil. This could be caused by the residual adverse effect of Fipronil on invertebrates. This is a significant observation. If it is confirmed in subsequent sampling it would indicate that Fipronil baiting could be having an adverse effect on Christmas Island ecosystems. However according to a report commissioned by the Director of Parks Australia on monitoring of the aerial baiting using Fipronil in 2009 the conclusion was that there was no noticeable evidence of any impacts on non-target invertebrates (Weeks and McColl, 2011).

8. CONCLUSION

From the analysis of the trial data there is strong evidence that boric acid is an effective bait for the control of YCA, and that it is a safer and significantly more environmentally friendly approach to control YCA compared to the present Fipronil baiting regime on Christmas Island. Invertebrate counts show increases in numbers at all three trial sites, all sites showed a suppression or reduction in YCA density, with a total absence of YCA from the Greta Beach trial site. Observations of the Greta Beach baited site suggested that there has been a substantial decrease in scale insect numbers (including some trees showing mortality of the scale insect on branches) with an increase in leaf cover. This is assumed to be due to the removal of the YCAs from the area and the disruption of the primary mutualistic relationship (Davis *et al*, 2009; Von Holle, 2011) between YCA and scale insects.

So far there is an indication that boric acid bait is readily taken up by foraging YCA and that the bait has a knock-down effect on the colony's density. This reduction in YCAs density allows for the re-colonisation of other invertebrates, which were previously suppressed due to YCA predation and dominance in the trial area.

This experiment has yielded sufficient results to continue on a longer basis since it has led to a decrease in YCA and an increase in native invertebrates with no other known animal deaths. If YCAs are out-competing native animals for invertebrate prey, starvation could be the cause for the decline and loss of native species on the island.

The interim results of the back-to-back sampling of two Fipronil aerial baited sites with that of the control and trial sites at South Point indicate that there is a substantial difference in invertebrate numbers between the boric acid trial sites and the aerial baited sites. This suggests it is possible that Fipronil has a prolonged impact on non-target- invertebrates within the Fipronil aerial baiting sites and possible residual effects on the environment, preventing invertebrates from re-colonising these areas after removal of the YCA.

9. RECOMMENDATIONS

1. From the above finding it is recommended that boric acid ant baits be utilised as a management tool in suppressing and controlling YCA on Christmas Island, with the purpose of protecting the islands native invertebrate and vertebrate animals, their food sources and habitats.
2. The cost of using fixed bait stations which are very expensive and require constant re-baiting would make it impractical to use the same system used in this trial for control of YCAs on a broad scale on Christmas Island. However, current trials of a continuously replenishing low cost bait delivery system indicate that costs of baiting significant areas of the Island could be reduced markedly and it is recommended that an operational trial of this system proceed.
3. That this trial be replicated at any future continuously replenishing low cost bait delivery system site to further confirm the above conclusions.
4. The lag effect observed during the trial, where YCA numbers continued to decline after bait was removed, and with a concomitant increase in insect prey, would be worthy of further investigation. It is recommended that the three sites are re-sampled every six months to determine how long it would take for YCA to return to original densities, which would also provide information for the operation of the low cost delivery system.
5. Present and continued control methods using fipronil may result in the loss of additional invertebrate prey species due to the non-selectiveness of this insecticide, further compounding a possible critical food shortage for native species on Christmas Island and contributing to their extinction .While it is not possible to conclude that fipronil is having an adverse effect on the Island ecosystem until further studies are undertaken we believe there is sufficient information from this study to invoke the “precautionary principle” and cease use of this non selective insecticide until it is demonstrated that it is not having an adverse and possibly permanent effect on the Christmas Island ecosystem.

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Annexure 1: Experimental design

Objective	Site	Description	Location	No. of replicates	Bait stations		YCA monitoring		NTS monitoring				Searches		Light-traps	
					No. per replicate	Total	Transects		Pit-traps		Anabats		No. per replicate	Total	No. per replicate	Total
							No. per replicate	Total	No. per replicate	Total	No. per replicate	Total				
1	CLP	Workshop	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2	3	High density YCA colony	AGD VCL	Two: 2ai 2aii	100	200	1 x 50m	2	10	20	1	2	1 x 50m	2	1	2
	2	Mid Density YCA colony	AGD VCL	Two: 2bi 2bii	100	200	1 x 50m	2	10	20	1	2	1 x 50m	2	1	2
	1	Low - mid Density YCA colony	AGD VCL	Two: 2ci 2cii	100	200	1 x 50m	2	10	20	1	2	1 x 50m	2	1	2
3	1 to 3	Control sites	AGD VCL	Two each	0	0	1 x 50m	2	10	20	1	2	1 x 50m	2	1	2
4	All	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Totals				12		600		8		80		8*		8		8*

YCA Yellow crazy Ant

NTS Non-target species

AGD Attorney Generals Department

VCL Vacant Crown Land

*Effectively 2 units required because of sampling schedule (Table 2)

Annexure 1: Monitoring frequency.

Method	Month 1	Month 2				Month 3				Month 4				Month 5				Month 6	Month 7	Month 8	Month 9	Month 10	
	W4	W 1	W 2	W 3	W 4	W 1	W 2	W 3	W 4	W 1	W 2	W 3	W 4	W 1	W 2	W 3	W 4	W1-4	W1-4	W1-4	W1-4	W1-4	
YCA	2ai 2aii 2bi 2bii 2ci 2cii	2ai 2ai i 2bi 2bi i 2ci 2ci i	PL 	2ai 2ai i 2bi 2bi i 2ci 2ci i	2ai 2ai i 2bi 2bi i 2ci 2ci i	2ai 2ai i 2bi 2bi i 2ci 2ci i	2ai 2ai i 2bi 2bi i 2ci 2ci i	2ai 2ai i 2bi 2bi i 2ci 2ci i	2ai 2ai i 2bi 2bi i 2ci 2ci i														
Pit-traps & Search	Inst all	3ai 3ai i	3bi 3bi i	3ci 3ci i		3ai 3ai i	3bi 3bi i	3ci 3ci i	PL	3ai 3ai i	3bi 3bi i	3ci 3ci i		3ai 3ai i	3bi 3bi i	3ci 3ci i		3abci 3abci i	3abci 3abci i	3abci 3abci i	3abci 3abci i	3abci 3abci i	
Anabat & Light-trap		3ai 3ai i	3bi 3bi i	3ci 3ci i		3ai 3ai i	3bi 3bi i	3ci 3ci i		3ai 3ai i	3bi 3bi i	3ci 3ci i		3ai 3ai i	3bi 3bi i	3ci 3ci i							

W week

PL poison laid and maintained over duration of trial

YCA: Along each 5m of a transect, clear any litter with 1 hand swipe, place a plastic card with inscribed 10 x 10cm squares, wait 15s, then record ants on one randomly selected square over 30s; input data to computer

Pit-traps: Install: PVC pipe, approx. 5cm diameter; dig 10 holes/replicate spaced 10m, chop pipe to fit (approx 20cm), insert floor, tag & number pit site; Monitor: Day1: open & clean pit, Day3: record invertebrates etc. & cover/lid pit-trap; input data to computer

Search: Install: establish 1 x 50m transect per replicate, tag route. Monitor: 2 observers conduct non-destructive search along transect (approx. 10m wide) recording invertebrates including crabs, reptiles, forest birds. On return along transect count red crab holes. Replace logs, rocks, litter etc; input data to computer

Anabat: Day1: establish in robber crab-free position, in approx. middle of replicate, before dark; Day2: remove Anabat and download data to computer. If rain/strong wind, repeat another night

Light-trap: Day1: establish in robber crab-free position, in approx. middle of replicate; Day2: remove trap and identify captures, input data to computer. If rain/strong wind, repeat another night.

