IMPACT OF RED IMPORTED FIRE ANTS ON INSECT ABUNDANCE AS A FOOD SOURCE FOR BROODS OF THE CRITICALLY ENDANGERED ATTWATER'S PRAIRIE-CHICKEN

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Project title:

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Introduction

The endangered Attwater's prairie-chicken (APC) (*Tympanuchus cupido attwateri*) is on the brink of extinction (Morrow et al. 1996, 2004, Silvy et al. 2004). Currently, fewer than 100 individuals remain in 3 geographically separated populations in Texas (Attwater Prairie Chicken National Wildlife Refuge (APCNWR) near Eagle Lake, Texas City Prairie Preserve, private land in Goliad County) (Morrow et al. 2004). These populations have been supplemented with captive-reared birds since 1996 (Morrow et al. 2004). Through the use of predator management and predator-deterrent fences around nest sites, 2001 – 2005 nesting success averaged 61% (APCNWR, unpublished data), compared to the historic average of

32% (Peterson and Silvy 1996). However, poor brood survival has been identified as a one of the major limiting factors affecting Attwater's prairie-chicken recovery (APCNWR, unpublished data). Like most gallinaceous birds, prairie-chicken chicks are primarily insectivorous during the first weeks of life (Lehmann 1941, Jones 1963, Thomas 1987, Johnson and Boyce 1990). Preliminary data collected collaboratively by APCNWR and the Society of *Tympanuchus Cupido Pinnatus* suggest that availability of insects as a food source for newly hatched chicks is contributing to the observed poor brood survival. Comparisons of insect abundance in APC brood habitat (APCNWR) with that from an increasing Minnesota greater prairie-chicken (GPC) (*T. c. pinnatus*) population found that while total insect biomass did not differ between the 2 areas, APC brood habitat supported < 30% of insect numbers compared to the GPC brood habitat (Pratt et al. 2003, unpublished report). Based on these preliminary data we argue that the red imported fire ant (RIFA), *Solenopsis invicta* Buren, might contribute to the reduction of insect diversity and abundance at APC brood habitat.

Disruptive impacts of RIFA on native insect communities are relatively well documented. In a Texas study, Porter and Savignano (1990) reported that species richness and abundance of non-ant arthropods were respectively 30% and 75% lower in sites infested by RIFA. In another study, Calixto et al. (2007) investigates how the reduction of RIFA benefited other ant species resulting in increases of up to 25%. Allen et al. (2001) described how RIFA altered the abundance of the loggerhead shrike by decimating insect abundance, the main source of food for this species.

Although experimental work links RIFA to negative impacts on arthropods, claims about the impact of this species on vertebrates are built mostly on anecdotal reports rather than on experimental evidence (Allen et al 2004, Tschinkel 2006). To cite a few examples, RIFA are linked to the decline of horned lizards in Texas where it is believed to competitively exclude the red harvester ant, *Pogonomyrmex sp.*, considered by many the primary food source of the horned lizard (*Phrynosoma cornutum*) (Taber 1998). Barlett (1997) speculates that snake declines in certain areas is due to egg predation by RIFA. In yet another descriptive study, Stake and Cimprich (2003) conclude that the survival of the federally endangered black capped vireo (*Vireo atricapillus*) is severely compromised by the presence of RIFA. In this study, video monitoring of 142 black-capped vireo nests identified RIFA as the primary nest predator on Fort Hood, TX. The impact at the population level remains unclear. Despite the poor understanding of RIFA impacts on vertebrates there are a few studies that merit mention. Drees (1994) conducted a removal experiment using broadcast baits on a spoil island off the Texas coast where he investigated the breeding success of several species of water birds. He found that survival of water birds was 90% lower in untreated areas compared to those where RIFA was reduced. Allen et al. (1995) studied the impact of RIFA on bobwhite quail (*Colinus virginianus*), white-deer tailed deer *(Odocoileus virginianus*) and loggerhead shrike (*Lanus ludovicianus*) populations. In this study bobwhite survival was significantly higher in areas were RIFA was removed.

The lack of manipulative experiments and appropriate replication, limits the inference that can be drawn from these experiments. These are strong justification for studying the impact of RIFA on food resources of the endangered Attwater's prairie-chicken in Texas. Understanding the effects of RIFA on food sources for APC broods is essential for the successful reintroduction and establishment of this endangered species in their native areas. Long-term comprehensive ecological studies with a larger scope are needed, conducted with untreated controls and adequate temporal and spatial replication to provide an understanding of the effects of RIFA on vertebrate and invertebrate populations.

The goal of this preliminary study was to collect baseline data that would help in the future to determine the impacts of RIFA on insect populations at the APCNWR. To address this goal, we attempted to determined the impacts of RIFA on 1) insect abundance and 2) insect diversity.

Materials and Methods

Study site

This study was conducted at the Attwater Prairie Chicken National Wildlife Refuge (NWR) located approximately 60 miles west of Houston, Texas. It is one of the largest remnants of coastal prairie habitat remaining in southeast Texas and home to one of the last populations of the critically endangered Attwater's prairie-chicken. All activities were conducted on coastal prairie grassland during the APC brood rearing period 15 April – 30 June, 2006, 2007 and 2008, respectively).

Experimental Design

In this study we used an Impact-Reference design with match pairs with 5 sets of replicates in space and 6 replicates in time (15 April – 30 June) (site by time factorial design).

Experimental plots. Plots consisted of two-hectare (5-acre) plots as experimental units. The software ArcGIS (ESRI) was used for selecting plots. Plots were located in the field using a Trimble GPS (submeter accuracy). To reduce variation, pairs of plots were blocked according to the habitat observed both in the field and on aerial photographs (**Figure 1**).

Treatments. Consisted of 1) RIFA reduced using broadcast bait (Extinguish® Plus "hopper blend, consisting of Extinguish®, 0.5% s-methoprene and Amdro®, 0.73% hydramethylnon, labeled for pastures) and 2) untreated controls. The treatments were assigned to the experimental units at random. Bait treatments were applied March (2006, 2007 and 2008), and retreated in November (2006, 2007) to maintain 80%+ fire ant control. These products have shown low residual effects and no adverse impact on non-target arthropods, especially in areas where fire ants are abundant and rapidly remove all the insecticide material back to the colony (Calixto 2008).

Sampling Methods. We used three complementary sampling techniques 1) sweep net sampling, 2) pitfall traps and 3) fatty food lures (1.3-cm slices of hot dogs) on each experimental plot. Samples were collected on a weekly basis from 15 April-30 June 2006 (APC brood rearing period) and processed during the fall and winter.

1) *Sweep net sampling*: This method was used to estimate the abundance and species composition of active insects in lower vegetation (Southwood 1978). The sweep net measured 38 cm (15 inches) in diameter and was attached to a 1-m wooden pole via a steel ring. The net was constructed of sailcloth. We took samples at four points at 25 m from the plot center. To minimize sample variation, the same individual who swept at the treated plot also swept at the untreated plot on the same day. A sweep consisted of one 180° arcs through the vegetation, quickly turning and reversing direction at the end of the first arc. A sample consisted of 25 sweeps taken on each of four sub-plot quadrants, walking at a constant speed.

Each sweep-net sample was then transferred to a one gallon plastic bag and labeled. Bags were stored in a freezer until processed.

2) *Pitfall traps*: This method was used to estimate the abundance and species composition of ground active insects (Southwood 1978). A trap consisted of a 120 ml plastic cup filled with propylene glycol (commercial non-toxic antifreeze). We deployed 5 uniformly distributed traps (one at the plot center, 4 at 25 m from the center) on the ground of each plot (systematic sampling provides uniform coverage of the whole of the population of interest according to Morrison et al. (2001)) (**Figure 2**). In 2007–2008, we left the units open for 7 days before samples were collected and returned to the laboratory for processing. In 2006, samples were collected after 48 hours.

3) *Fatty food lures ("hot dogs")*: This method was used to estimate and examine RIFA activity and behavior in the ground (Bestelmeyer et al 2000). We assessed pre- and post-treatment RIFA surface activity on each 2 hectare plot by determining the number of RIFA/30-45 minutes attracted to 10 food lures (1.3-cm hot dog slices) distributed uniformly within 25 m of the plot center (Porter and Tschinkel 1987, Calixto et al 200X). We estimated the number of ants per slice using a scale system ranging from 0 to 100 on increments of 10 (Pereira and Porter 2005, Calixto et al. 200X).

Sample processing. Insects and other arthropods were frozen immediately after collection. Later, specimens were sorted out from each sample and placed in vials with alcohol. Impact of RIFA on relative abundance of insects and other arthropods (ants not included) were estimated by counting all insects caught using the two sampling methods per experimental unit (plot), and specimens were identified to order level. Selection of RIFA on certain size classes of insects and other arthropods (ants not included) were estimated by measuring each insect/arthropod collected using both sampling methods and recorded at order level. Total biomass (to nearest 0.01 g) of insects and other arthropods was determined for both pitfall and sweep net samples .

Data Analysis

Analyses focused on 3 aspects; 1) the effect of RIFA reduction (treated vs untreated) and time on insect and arthropod relative abundance, 2) the effect of RIFA reduction on insect and arthropod relative biomass, and 3) the effect of RIFA reduction on insect and arthropod size. Data obtained from the different sampling methods were analyzed using a Linear Mixed Model (Repeated Measures - Type III sum of squares and diagonal repeated covariance) comparing the effect of different treatments on the response variable (i.e., insect abundance, biomass) per sampling interval. In this model, treatment and sampling interval are considered fixed factors and plot as random factor. SPSS 16.0 (SPSS Inc. 2007) was used to perform these analyses. Values were considered significantly different at $P < 0.05$.

Results

Fire ant density: Data collected using pitfall traps and food lures confirmed an effective RIFA reduction as a result of the insecticide bait treatments. RIFA density was significantly lower on bait treated plots (>%80) compared to those left untreated (**Figure 3**). Relative fire ant density remained low throughout the sampling periods (April–June of each year 2006-2008). Samples were not collected at other times of the year except 2008.

Impact on insect and other arthropod relative abundance: Numbers of insects and other arthropods caught using pitfall traps showed no significant differences between RIFA reduced sites versus those untreated (**Figure 4**). However relative abundance of Hemipterans (i.e., leafhoppers) and Hymenopterans (i.e., bees, non-ant specimens) was significantly higher on untreated plots only for year 2006. Isopods (pill bugs) were the only group to appear significantly higher on treated plots for the three years of the study. Numbers of insects and other arthropods caught using sweep nets showed no significant differences between RIFA reduced sites versus untreated (**Figure 5**). However, relative abundance of Hemipterans (i.e., leafhoppers) was significantly higher on untreated plots for year 2006 even though these results appear inconclusive.

Impact on insect and other arthropods sizes: Size of insects and arthropods caught using pitfall traps and sweep nets did not differ between treated and untreated sites (**Figure 6** and **7**). However, Isopods (pill bugs) caught using pitfall traps were significantly larger (mm) in RIFA reduced areas compared to those untreated.

Impact on insect and other arthropod relative biomass: Overall dry weight of insects and arthropods caught using pitfall traps and sweep nets did not differ between treated and untreated sites for the three years of the study (**Figure 8** and **9**).

Discussion

This preliminary study has shown that for the 2006-2008 seasons, removal of RIFA did not exert significant impacts on insect guilds and arthropods (ants not included). Further analyses at the order/family level and size classes might be necessary to understand the impacts of RIFA on specific insect/arthropod assemblages. Pill bugs (Isopoda) were the only group that appeared significantly affected by RIFA because they were significantly more abundant and larger in areas where RIFA was reduced.

Modifications of methods used in this 3 year study are warranted in future efforts to assess the impact of RIFA on insect/arthropod fauna. Plots, larger than the 2 hectare (5-acre) plots in this trial, should be considered because highly mobile winged insects may have migrated into untreated plots. Additionally, while in general we did not sample insects or RIFA outside the April–June focus of our study, samples collected in late summer/fall 2008 indicated that RIFA had already begun to re-invade treated plots in substantial numbers. It is possible that reduction in RIFA numbers was not being achieved during time periods critical to insect reproductive success. Finally, more detailed analysis of samples (by order/family) would be needed to effectively determine the impact of RIFA on insect/arthropod abundance. In this study, samples were processed to order level. Analysis should be performed to establish the effects of RIFA on insect communities at a smaller scale, possibly focusing on insect family or group(s) of species thought to serve as prominent Attwater prairie chicken food. If differences were found, research hypotheses could be developed where effect sizes (magnitude of the effect) were used to determine biological significance of the impact of RIFA on specific insect assemblages. This information would better tailor strategies for the management of RIFA on this area and for the restoration and conservation of the Attwater's Prairie-Chicken.

Conclusions

This preliminary study failed to document consistent and significant impacts of RIFA foraging on insects and other arthropods with the exception that numbers of pill bugs (Isoptera) were significantly higher and larger (mm) in RIFA reduced plots. It is possible that the study design for this preliminary study was inadequate to determine impacts on arthropod communities, especially with respect to duration of RIFA reduction as it relates to the life history of other arthropods. Because of the preliminary nature of this study, plot size and number of replications may have been inadequate to document effects of RIFA control Additionally, factors other than RIFA (e.g., weather, poor drainage, genetic isolation, pesticide drift, etc.) may have contributed to low insect/arthropod abundance in this area.

A more elaborated study that accounts for these design constraints should be considered for future efforts. Classification to lower taxonomic levels may be necessary for understanding RIFA impacts. RIFA and insect/arthropod assessments through the year, particularly in late fall and early spring, could perhaps better document the relationship between these groups and timing of RIFA reduction and subsequent re-invasion. Finally, the development of target-specific treatments for RIFA, particularly in areas where RIFA population densities are low in order to assure minimization of potential secondary impacts of insecticide baits on non-target species, would possibly improve the likelihood of documenting the effects of RIFA on local insect/arthropod assemblages and support implementation of RIFA as a management practice in this sensitive environment.

Products resulting from this Texas Parks and Wildlife Department supported effort:

Presentations (Posters)

2009 A. Calixto, B. Drees, M. Morrow, D. Roach, J. Johnson, T. Catanach and C. Botero. Impact of fire ants on Attwater's prairie chicken's diet. Annual Red Imported Fire Ant Research Conference. Oklahoma City, Oklahoma.

2009 A. Calixto, B. Drees, M. Morrow, D. Roach, J. Johnson, T. Catanach and C. Botero. Impact of fire ants on Attwater's prairie chicken's diet. 57th meeting of the Southwestern Branch of the Entomological Society of America. Stillwater, Oklahoma.

2008 A, Calixto, B. Drees, M. Morrow, D. Roach, J. Johnson, T. Catanach and C. Botero. Impact of *Solenopsis invicta* on prairie insects essential for the survival of the endangered Attwater's prairie-chicken. Entomological Society of America, Annual Meeting. Reno, Nevada.

2008 A, Calixto, B. Drees, M. Morrow, D. Roach, J. Johnson, T. Catanach and C. Botero. Impact of *Solenopsis invicta* on prairie insects essential for the survival of the endangered Attwater's prairie-chicken. The Wildlife Society, Annual Meeting. Miami, Florida.

2008 A, Calixto, C. Botero, B. Drees, M. Morrow, D. Roach, J. Johnson and M. Harris. Red Imported Fire Ant impact on insect community as a food source for the endangered Attwater's Prairie Chicken. Annual Red Imported Fire Ant Research Conference. Charleston, South Carolina.

2008 C. Botero, A, Calixto, B. Drees, M. Morrow, D. Roach, J. Johnson and M. Harris. Red Imported Fire Ant impact on insect community as a food source for the endangered Attwater's Prairie Chicken. 56th meeting of the Southwestern Branch of the Entomological Society of America. Fort Worth, Texas.

Proceedings

Botero, C., B. Drees, A. Calixto and M. K. Harris. 2008. Red imported fire ant impact on insect community as a food source for the endangered attwater's prairie-chicken. Pp. 15. *In* proceedings 56th meeting of the Southwestern Branch of the Entomological Society of America. Fort Worth, Texas.

Extension Publication

Drees, B., A. Calixto, C. Botero, M. Morrow, D. Roach and J. Johnson. 2008. Preliminary Assessment of the Impacts of Red Imported Fire Ants on Communities of Insects that Serves as Major Food Source for Broods of the Endangered Attwater's Prairie Chicken. Pp 152-159. *In* Urban IPM Program Summary Report 2007, Texas AgriLife Extension Service.

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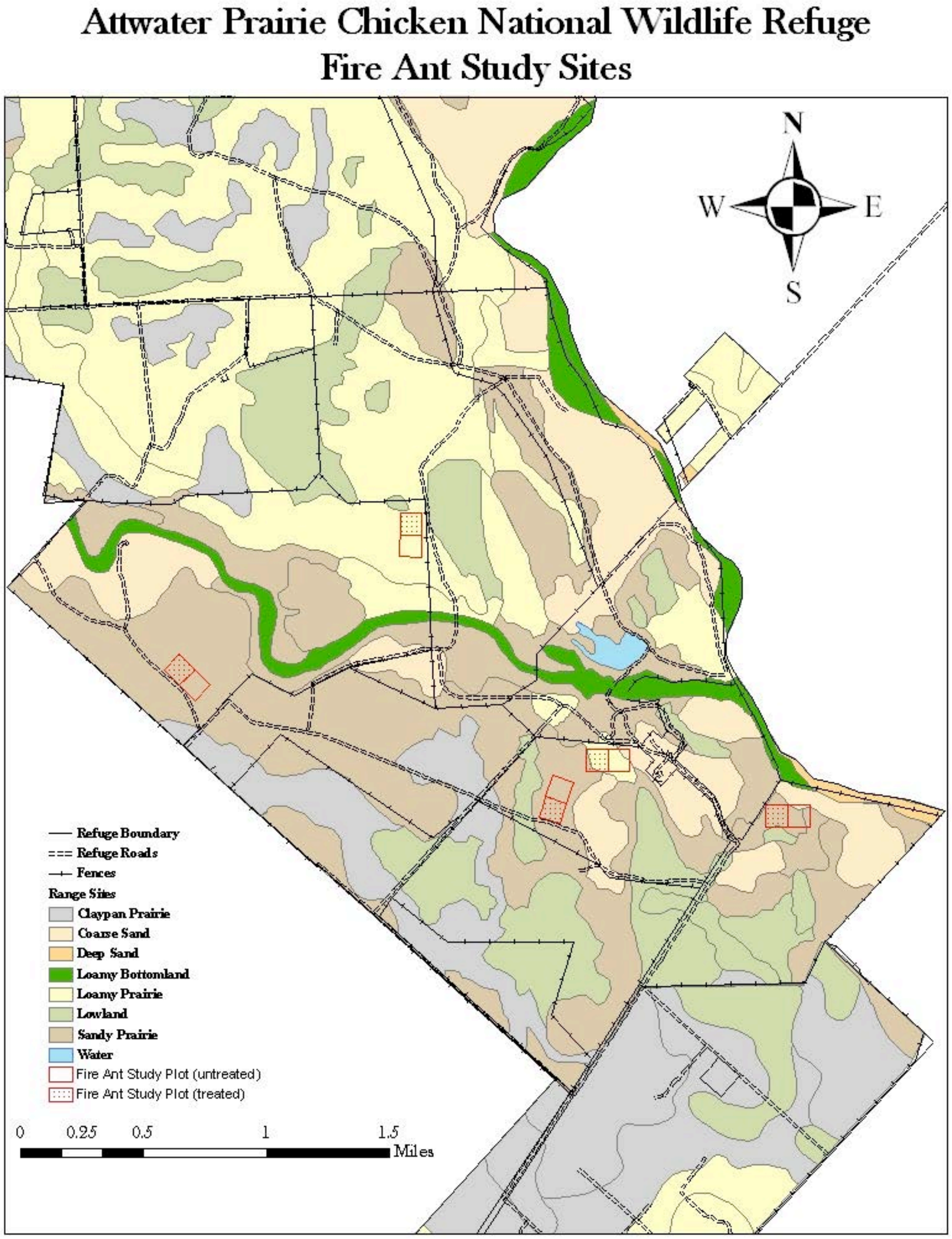

Figure 1. Plots location at APCNWR.

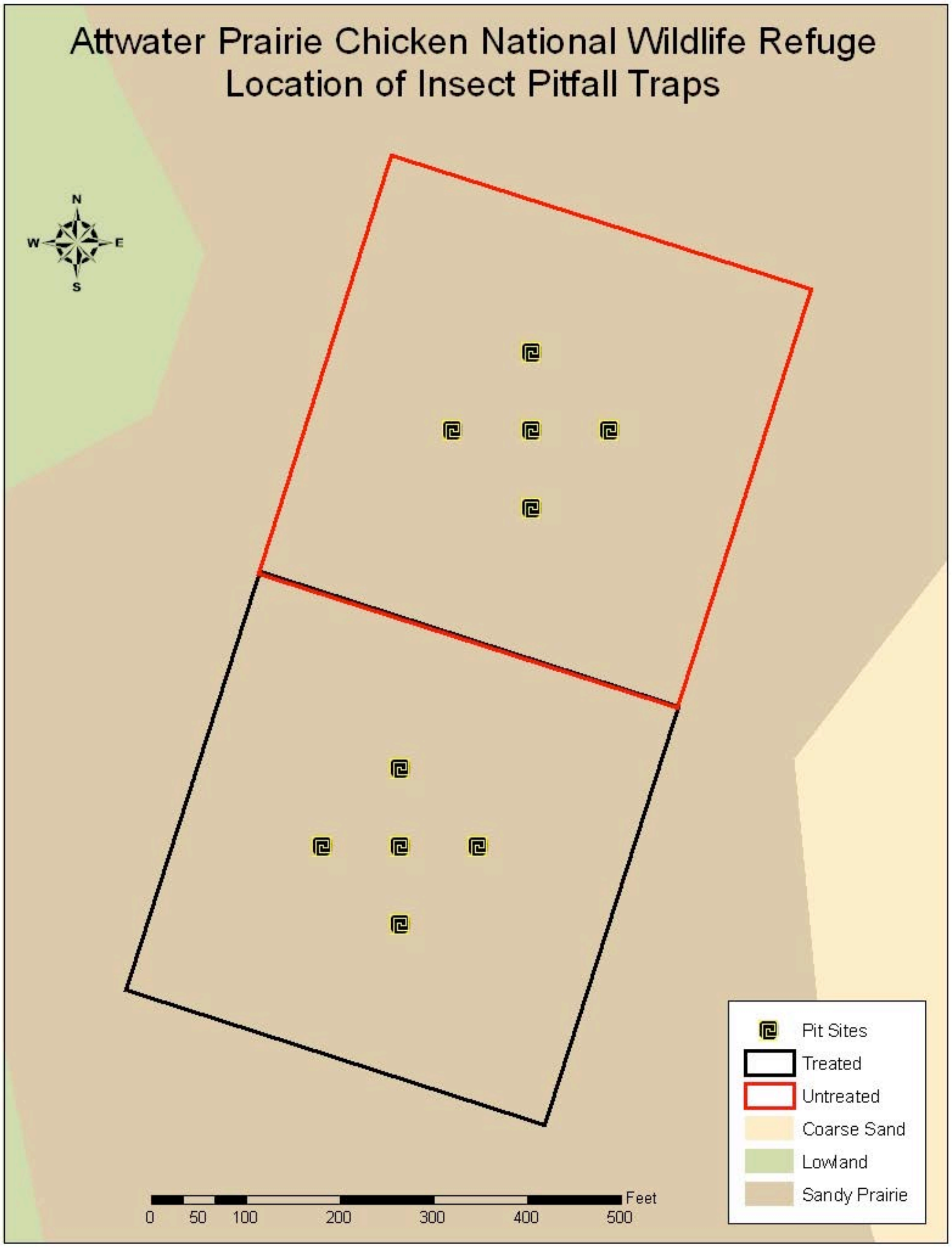

Figure 2. Pitfall traps and food lures location (systematic sampling).

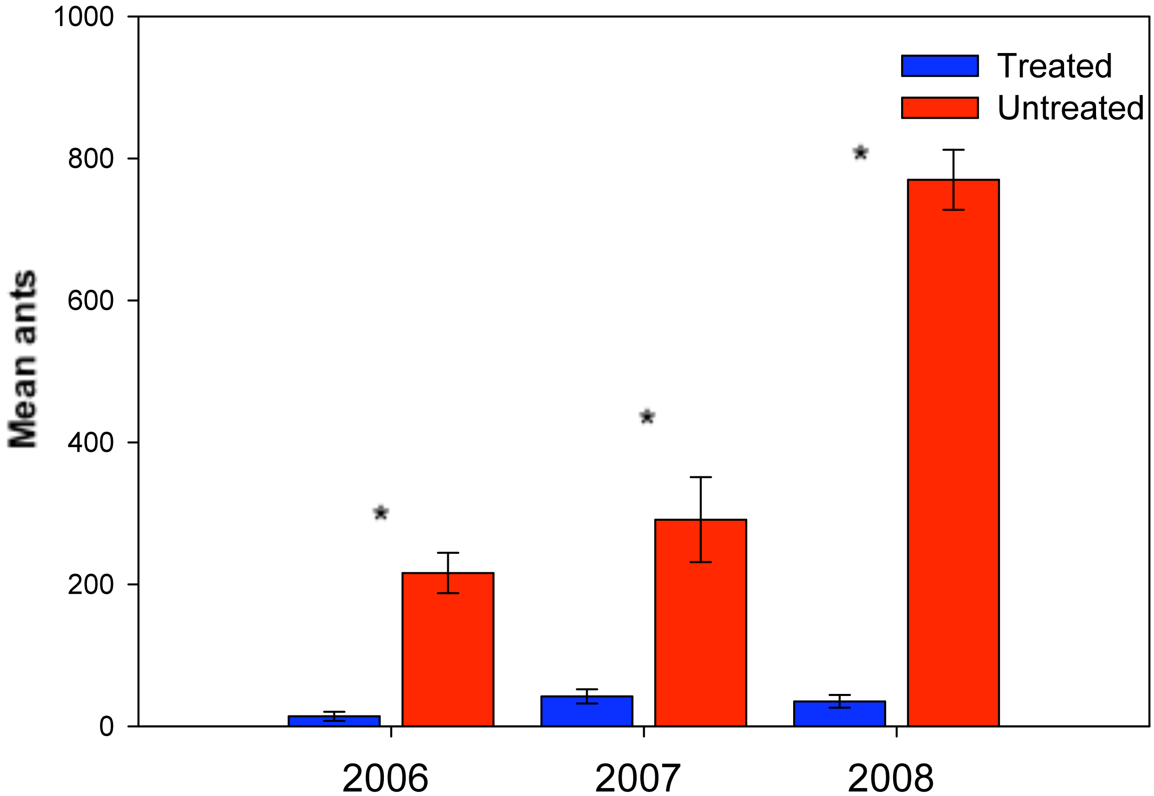

Figure 3. Fire ant density (mean±se) in bait treated and untreated plots based on pitfall traps and food lures, 2006-2008. (Mixed Linear Model (MLM), * indicates differences at $P < 0.05$).

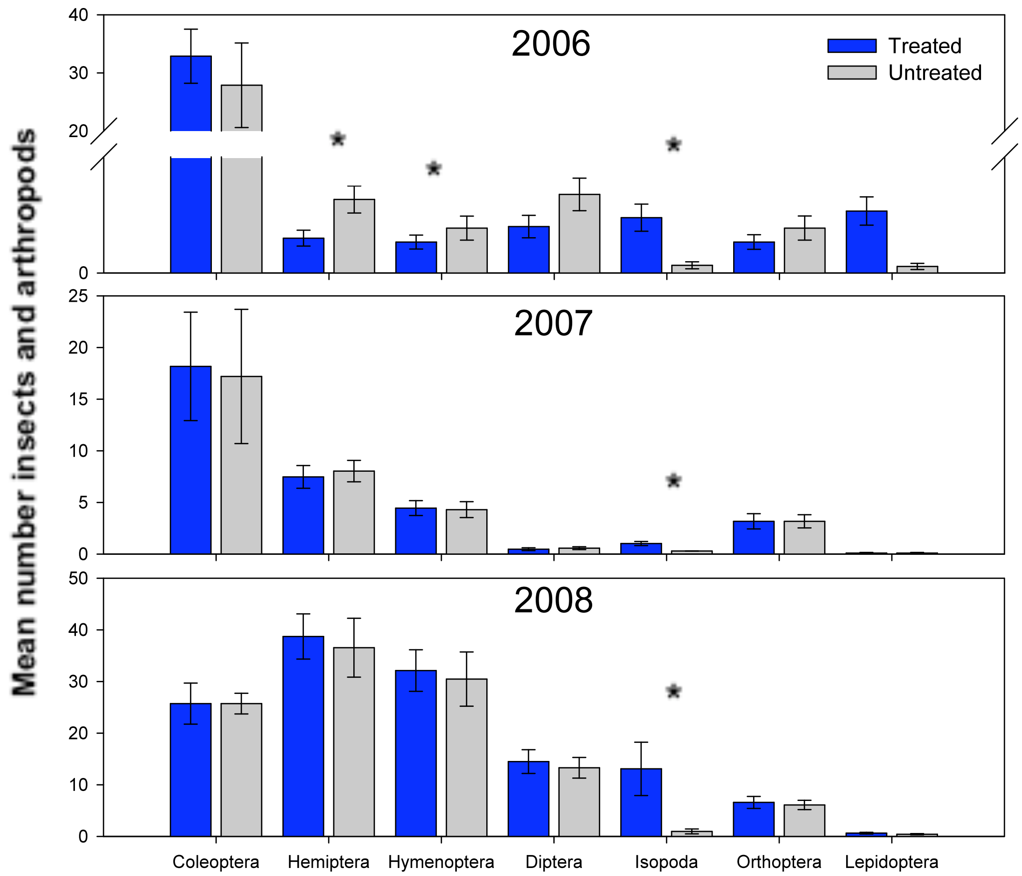

Figure 4. Insect and arthropods relative abundance in RIFA reduced and untreated plots based on pitfall traps, 2006-2008. (Mixed Linear Model (MLM), * indicated differences at $P < 0.05$).

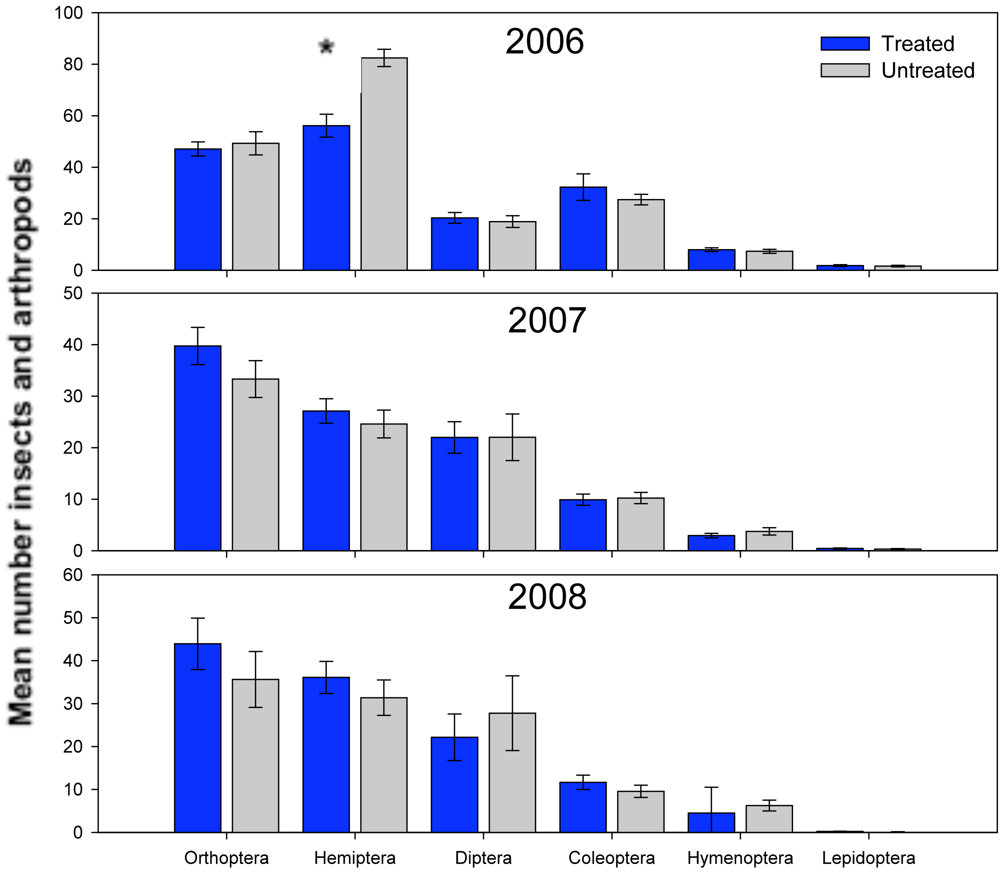

Figure 5. Insect and arthropods relative abundance in RIFA reduced and untreated plots based on sweep nets, 2006-2008. (Mixed Linear Model (MLM), * indicated differences at $P < 0.05$).

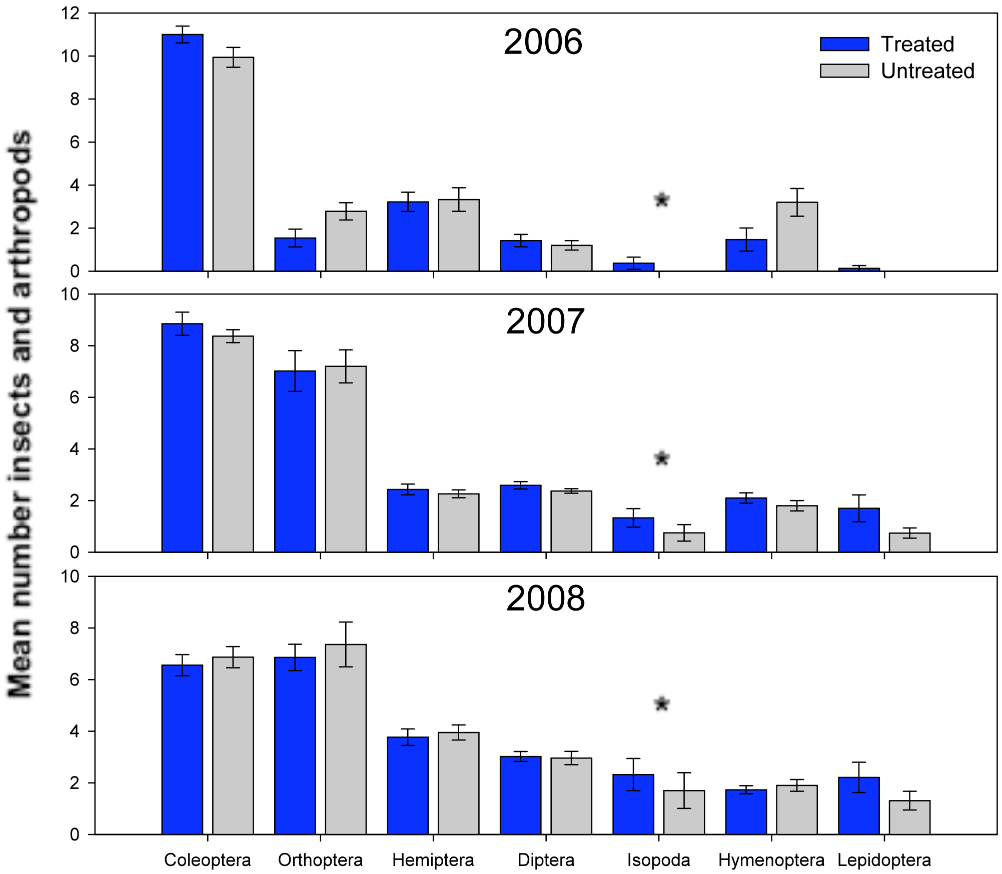

Figure 6. Insect and arthropod sizes in RIFA reduced and untreated plots based on pitfall traps, 2006-2008. (Mixed Linear Model (MLM), * indicated differences at $P < 0.05$).

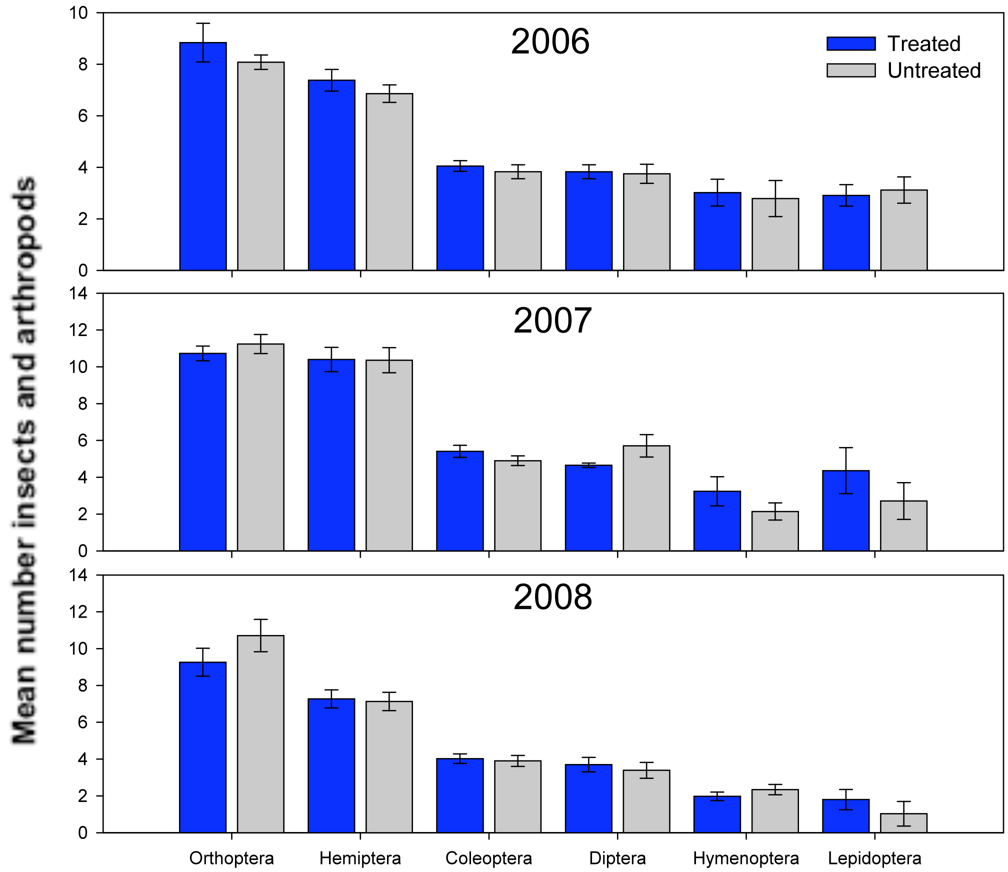

Figure 7. Insect and arthropod sizes in RIFA reduced and untreated plots based on sweep nets, 2006-2008. (Mixed Linear Model (MLM), * indicated differences at $P < 0.05$).

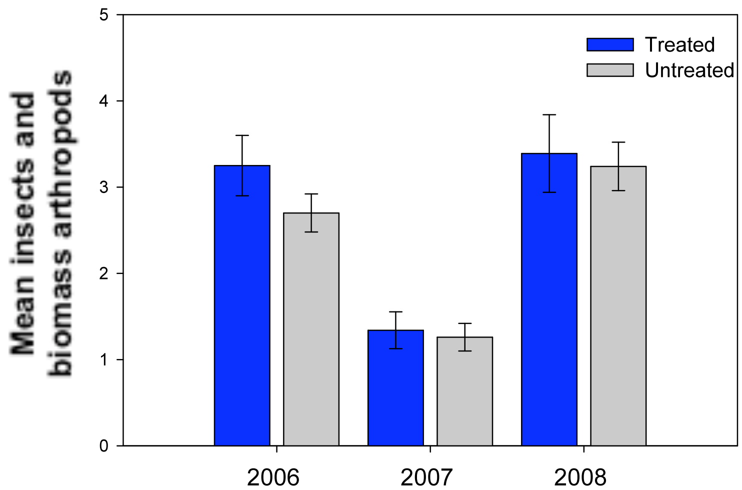

Figure 8. Insect and arthropod relative biomass in RIFA reduced and untreated plots based on pitfall traps, 2006-2008. (Mixed Linear Model (MLM), * indicated differences at $P < 0.05$).

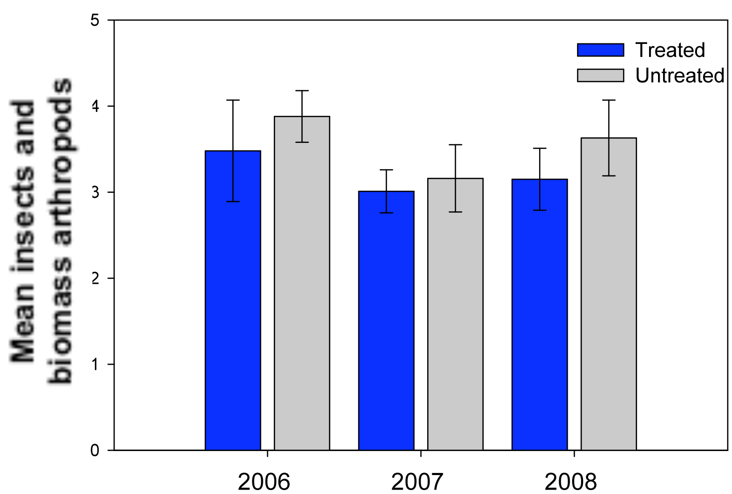

Figure 9. Insect and arthropod relative biomass in RIFA reduced and untreated plots based on sweep nets, 2006-2008. (Mixed Linear Model (MLM), * indicated differences at $P < 0.05$).