

**Impact of Cogongrass (*Imperata cylindrica*) Presence and Management Strategies on
Arthropod Natural Enemy Populations in Longleaf Pine Stands**

by

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Abstract

Cogongrass (*Imperata cylindrica* Beav.) is an aggressive, invasive weed that is threatening the integrity of longleaf pine (*Pinus palustris* Mill.) ecosystems in southeast Alabama. Various management strategies are implemented to suppress cogongrass. In Objective 1, populations of natural enemies and bark beetles (*Hylastes* spp.) were monitored over 2 years in response to four different management strategies; herbicide, seeding with native species, seeding plus herbicide, and control (no treatment) in burned and unburned longleaf pine plots. In Objective 2, the impacts of cogongrass on the abundance of arthropods and soil dwelling microfauna in longleaf pine were investigated. Insects were collected via pitfall traps and sweep samples, then sorted and counted in the lab. For Objective 1, sentinel species (mainly generalist predators) were monitored. For Objective 2, all arthropods were sorted to family level and counted. In spring and fall 2011, earthworms were sampled using a disclosing solution in longleaf pine with or without cogongrass. Soil cores were taken in May 2011 to determine the relative abundance and diversity of nematodes. Cogongrass management strategies had minimal impacts on natural enemies and no impact on *Hylastes* populations. The seeding plus herbicide treatment had significantly fewer spiders than other treatments but this was the only affected group of natural enemies. Thirty of the >100 families collected were significantly impacted by cogongrass but not all impacts were negative. For example, Curculionidae, Tettigoniidae, Acrididae, and Cercopidae were more abundant in longleaf pine infested with cogongrass. Similarly, earthworm abundance and weight were

significantly greater in longleaf pine with cogongrass present. Plant parasitic nematodes were significantly greater in longleaf pine with cogongrass present, the opposite being true for free-living nematodes. These results suggest that cogongrass management does not significantly affect natural enemy and bark beetle populations, but its presence does. The families that were impacted, particularly Scarabaeidae (and their relatives) or Gryllidae, can be used in future evaluations of ecosystem impacts of cogongrass invasion. Also, families of herbivores that increased in the longleaf pine stands where cogongrass was present may provide insight for future studies seeking herbivores that may be able to utilize this invasive grass.

Dedication

I would like to dedicate my thesis and its work to my ever encouraging parents, Herb and Marynm, and my brothers, Sam and Wade. Also, to my fiancée Josh, who stood by my side through every bit of this process and never let me quit.

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Table of Contents

| | |
|---|------|
| Abstract..... | ii |
| Dedication..... | iv |
| Acknowledgments..... | v |
| List of Tables | viii |
| List of Figures..... | ix |
| Chapter 1 | 1 |
| Longleaf Pine Ecosystems | 1 |
| Insects in the Pine Ecosystems | 1 |
| Earthworms and Nematodes | 5 |
| Cogongrass Morphology, Biology, and Distribution..... | 6 |
| Ecosystem Impacts of Cogongrass | 7 |
| Cogongrass Management..... | 9 |
| Interactions..... | 11 |
| Chapter 2 | 15 |
| Introduction..... | 16 |
| Materials and Methods..... | 18 |
| Site description..... | 18 |
| Experimental Design and Treatments..... | 18 |
| Arthropod Sampling..... | 21 |
| Analyses..... | 23 |

| | |
|----------------------------|----|
| Results | 23 |
| Discussion | 25 |
| Chapter 3 | 40 |
| Introduction..... | 41 |
| Materials and Methods..... | 44 |
| Site description..... | 44 |
| Experimental Design..... | 44 |
| Arthropod Sampling..... | 44 |
| Earthworm Sampling | 45 |
| Nematode Sampling..... | 46 |
| Analyses | 46 |
| Results | 48 |
| Discussion | 49 |
| References | 62 |

List of Tables

| | |
|--|----|
| Table 2.1 Seed Mix for Seeding and Seeding + Herbicide in 2010..... | 31 |
| Table 2.2 Seed Mix for Seeding and Seeding + Herbicide in 2011..... | 31 |
| Table 2.3 Average soil characteristics by site..... | 32 |
| Table 3.1 Average soil characteristics for cogon(C) and no cogon (NC) plots for objective 2. ^a | 54 |
| Table 3.2 Comparison of arthropods collected over entire season in longleaf with and without cogongrass present by Wilcoxon In cogongrass present plots, 18,968 arthropods were collected and counted. In cogongrass absent plots, 20,182 arthropods were collected and counted. Both were collected over an 8 month period in 2011..... | 55 |
| Table 3.3 Comparison of populations of selected arthropods using all observations from each sample collected in pitfalls and sweep samples in longleaf pine with cogongrass present and cogongrass absent, Bay Minette and Grand Bay, AL, 2011, using repeated measures analyses | 58 |
| Table 3.4 Mean abundance of different nematode taxa. Total nematodes, plant parasitic (PP), and free-living (FL) are represented. If P <0.05, difference is noted between cogongrass present (C) and cogongrass absent (N). Repeated measures test used. | 59 |

List of Figures

| | |
|--|----|
| Figure 1.1 Longleaf pine stand with shrub and forb understory | 14 |
| Figure 1.2 Map of cogongrass distribution in Alabama in 2009 | 14 |
| Figure 2.1. Map of plot locations in Mobile County (Rainwater) and Baldwin County (Beebe)..... | 33 |
| Figure 2.2 Pitfall traps were arranged in an X pattern 3 meters apart on each 10 x 10 m plot | 33 |
| Figure 2.3 Composition of the natural enemy complex in the cogongrass treatment plots from pitfall traps and sweep samples. Samples contained mainly these three groups with parasitic hymenoptera and syrphids comprising the “other” natural enemy group. Percentages reflect total number of individuals collected in 31 sampling periods, 2010 and 2011..... | 34 |
| Figure 2.4 Natural enemies collected in each treatment per month over 31 collection trips in 2010 and 2011. Biweekly samples pitfall traps and sweep samples were pooled into monthly total. ANOVA revealed: year (ANOVA, $F= 6.72$, $df=1,63$, $P=0.0119$) and treatment ($F=2.06$, $df=3,59$, $P=0.1152$). Significant interaction was: year by site (ANOVA, $F=43.41$, $df=1,63$, $p <0.0001$)..... | 34 |
| Figure 2.5 Abundance of total natural enemies sampled from May 2010-Oct 2011 from pitfall traps and sweep samples. Main effect of site (ANOVA, $F=10.36$, $df=1,59$, $P=0.0021$), burn treatment (ANOVA, $F=14.69$, $df=1,59$, $P=0.0003$), and site by burn treatment (ANOVA, $F=28.67$, $df=1,59$, $P=<0.0001$) were significant..... | 35 |
| Figure 2.6 Abundance of predatory beetles sampled from May 2010-Oct 2011 from pitfall traps and sweep samples. Main effect of site, burn (ANOVA, $F =29.46$, $df=1,59$, $P= <0.0001$), and site by burn interaction (ANOVA, $F =26.20$, $df=1,59$, $P =<0.0001$) were significant..... | 35 |
| Figure 2.7 Abundance of ants sampled from May 2010-Oct 2011 from pitfall traps and sweep samples. Main effect of site (ANOVA, $F=0.06$, $df=1, 59$, $P=0.8093$), burn treatment (ANOVA, $F =10.89$, $df=1,59$, $P = 0.0016$), and site by burn treatment (ANOVA, $F =10.51$, $df=1,59$, $P = 0.0020$)..... | 36 |

| | |
|--|----|
| Figure 2.8 Abundance of spiders sampled from May 2010-Oct 2011 from pitfall traps and sweep samples. Main effect of site (ANOVA, $F = 11.45$, $df = 1, 59$, $P = 0.0013$), burn treatment (ANOVA, $F = 0.11$, $df = 1, 59$, $P = 0.7398$), and site by burn treatment (ANOVA, $F = 28.84$, $df = 1, 59$, $P = < 0.0001$). | 36 |
| Figure 2.9 Predatory beetles collected in each treatment over 31 collection trips in 2010 and 2011. ANOVA revealed: year (ANOVA, $F = 37.96$, $df = 1, 63$, $P = < 0.0001$), and treatment (ANOVA, $F = 1.75$, $df = 3, 59$, $P = 0.1670$). Significant interaction was: site by treatment (ANOVA, $F \text{ value} = 4.25$, $df = 3, 59$, $P = 0.0087$). | 37 |
| Figure 2.10 Ants collected in each treatment over 31 collection trips in 2010 and 2011. ANOVA revealed: year (ANOVA, $F = 6.39$, $df = 1, 63$, $P = 0.0140$), and treatment (ANOVA, $F = 1.94$, $df = 3, 59$, $P = 0.1334$). Significant interaction was: year by site (ANOVA, $F = 24.52$, $df = 1, 63$, $P = < 0.0001$). | 37 |
| Figure 2.11. Spiders collected in each treatment over 31 collection trips in 2010 and 2011. ANOVA revealed: year (ANOVA, $F = 1.21$, $df = 1, 63$, $P = 0.2758$), and treatment (ANOVA; $F = 4.11$; $df = 1, 63$; $P = 0.0102$). Significant interaction was: year by site (ANOVA, $F = 46.01$, $df = 1, 63$, $P = < 0.0001$). | 38 |
| Figure 2.12. Total <i>Hylastes</i> spp. collected in each treatment over 31 collection trips in 2010 and 2011. Biweekly samples from baited pitfall traps were pooled into monthly totals. ANOVA revealed: year (ANOVA, $F = 6.38$, $df = 1, 63$, $P = 0.0141$), site (ANOVA, $F = 0.81$, $df = 1, 59$, $P = 0.3730$), burn treatment (ANOVA, $F = 0.14$, $df = 1, 59$, $P = 0.7072$), and treatment (ANOVA, $F = 0.43$, $df = 3, 59$, $P = 0.7317$). Significant interaction was seen in year by site (ANOVA, $F = 4.18$, $df = 1, 63$, $P = 0.0452$). | 38 |
| Figure 2.13. Mean monthly temperatures in 2010 and 2011 for Baldwin County (Beebe) and Mobile County (Rainwater). (AWIS 2008) | 39 |
| Figure 2.14. Mean monthly precipitation in 2010 and 2011 for Baldwin County (Beebe) and Mobile County (Rainwater). (AWIS 2008) | 39 |
| Figure 3.1 Average soil characteristics for cogon(C) and no cogon (NC) plots for objective 2. ^a | 60 |
| Figure 3.2 Comparison of arthropods collected over entire season in longleaf with and without cogongrass present by Wilcoxon In cogongrass present plots, 18,968 arthropods were collected and counted. In cogongrass absent plots, 20,182 arthropods were collected and counted. Both were collected over an 8 month period in 2011 | 60 |
| Figure 3.3 Earthworm abundance in 2011 on cogongrass present and cogongrass absent plots by abundance. Year was not significant, therefore samples were combined. ANOVA: $F = 8.00$, $df = 1, 37$, $P = 0.0075$ | 61 |

Figure 3.4 Earthworms collected in 2011 on cogongrass present and cogongrass absent plots by weight (mg). Year was not significant, therefore samples were combined. ANOVA $F=7.57$, $df=1, 37$, $P=0.0091$ 61

Chapter 1

Introduction

Longleaf Pine Ecosystems

In the southeastern United States, longleaf pine (*Pinus palustris* Mill.) savannahs are unique ecologically (Bartholomew et al. 2006) because they are composed of many threatened and endangered endemic plant and animal species, and host an extremely species rich plant community (Brewer and Cralle 2003). They are characterized by an open canopy, lack of mid-story trees or shrubs, and well-developed, highly diverse herbaceous understory which is dominated by bunch grasses, graminoids, shrubs, vines, and legumes and forbs (Figure 1.1) (Boyer 1990, Daneshgar and Jose 2009). Without frequent, low intensity fires, this ecosystem quickly succeeds to a closed canopy hardwood mixed pine or evergreen shrub habitat (Boyer 1990). Historically, the range of longleaf pine stretched from southeastern Virginia to central Florida and west into Texas (Haywood 2009). Within Alabama, the majority of remaining longleaf sites are located in the southern portion of the state. Little of the original vast longleaf forests remain, and conservation and restoration of longleaf ecosystems is a high priority in the Southeast (Outcault and Sheffield 1996).

Insects in the Pine Ecosystem

Bark beetles and weevils (Curculionidae: Scolytinae) are the most destructive forest pests in the United States (Moeck and Safranyik 1984, Massey 1974). These include *Dendroctonus*, *Ips*, and *Hylastes* spp. Root-feeding *Hylastes* spp. are currently considered non-aggressive and not a threat. They typically attack unhealthy, declining, wounded or even dead pines (Eckhardt et al. 2004). Stress-altered allelochemical profiles of pine roots attract the scolytids and they aggressively feed on the roots releasing these chemicals (Eckhardt et al. 2008). Some other insects associated to Scolytid pheromones are: Rhizophagidae, Trogossitidae, Staphylinidae,

Histeridae, Nitidulidae, Tenebrionidae (Coleoptera), Dolichopodidae, Lonchaeidae, Asilidae, Pallopteridae (Diptera), Anthocoridae (Hemiptera), and Raphidiidae (Neuroptera). Beetles in the Carabidae and Silvanidae families are loosely associated with bark beetles (Kenis et al. 2004).

Certain arthropod taxa (i.e., ants, beetles, spiders) are known to be bioindicators in forest management. Ants (Formicidae) are effective bioindicators because of their sensitivity to ecosystem disruption caused by forest fires, species invasion, and other forms of disturbance (Maleque et al. 2009). They are associated with important ecosystem processes such as soil aeration, soil nitrogen cycling, seed dispersal and seed predation (Hölldobler and Wilson 1990). In Alabama, there are approximately 166 species of ants. Of those, twenty-eight species are exotic (MacGown and Forster 2005). Red imported fire ants (*Solenopsis invicta*, or RIFA) play a role in simplification of ecosystems by out-competing other ants (Reagan 1982). Red imported fire ants were introduced near Mobile, AL around 1930 (Lofgren 1986). Within 10 years of introduction, *S. invicta* had spread to 10 states in the southeast. High reproductive capacity, propensity for soil invasion, and efficient foraging behaviors undoubtedly contributed to their rapid spread (Lofgren 1986). Ecologically, *S. invicta* can be considered a weed species. Weeds are plant or animal species adapted for opportunistic exploitation of ecologically disturbed habitats (Tschinkel 1986). Disturbed habitats can occur from natural forces such as wind and storms as well as human disturbances like agriculture or homestead establishment. Human mediated activities are the greatest encourager of *S. invicta* range expansion (Tschinkel 1986). Fire ants flourish on early and secondary succession communities but are absent or rare in late succession and climax communities. They are often found mixed with other early succession weed species, both plants and animals. Overall, the red imported fire ant in Alabama is fundamentally an opportunistic weed species dependent on habitat disturbance for its continued success and existence (Tschinkel 1986).

Although *S. invicta* is notoriously detrimental to human activities, it is beneficial in some situations (Reagan 1986). In the crop fields of southern Louisiana, *S. invicta* was the most

important predator relative to other common predator groups (spiders, carabids, earwigs, and staphylinids; Charpentier et al. 1967). In forest systems, ants are important predators of eggs and larvae, especially of Lepidoptera. Red imported fire ants, specifically, are known predators of forest pests including Nantucket pine tip moth (*Rhyacionia frustrana*) (Reagan 1986) and spruce budworm (*Choristoneura hebenstreitella*) (Youngs 1983).

Like ants, predatory beetles in the families Carabidae, Staphylinidae, Cicindelidae, Histeridae and Cleridae are also generalist predators of forest pests (Kenis et al. 2004, Gandhi et al. 2001). Specifically, ground beetles (Carabidae) are used to gauge local-scale disturbance intensity because they are poorly dispersing, large-bodied insects that typically decrease in abundance with interference (Maleque et al. 2009, Rainio and Niemela 2002). Carabids decline quickly in response to disturbance, forest fragmentation, fertilization, and grazing and are sensitive to temperature, humidity, vegetation, and patch size. The most important factors impacting ground beetle abundance are soil water content, fragmentation and vegetation. They are also among the first to respond to forestry disturbances (Rainio and Niemela 2002). They can also function as indicators of arthropod groups on which they feed. Carabid species richness tends to be lower in closed canopy situations compared to open habitats (Humphrey et al. 1999). Larvae and adults occur in the leaf litter or soil and are omnivorous or predaceous usually feeding on dead or dying insects (White 1983). Adult carabids are ground active, sometimes in large numbers. Both larvae and adults are active at night and hide during the day. Ground beetles in the Carabini tribe will climb trees in their search for prey and the adults consume a much greater amount of food than the larvae (White 1983). Pedunculate ground beetles (tribe Scaritini) have the largest mandibles in the family and are some of the most rapacious predators. They are most commonly found in open fields, pastures and woodlands feeding on caterpillars and other larvae. False bombardier beetles (tribe Galeritini) are important for reducing cankerworm populations (White 1983). Carabids in the genus *Dromius* (tribe Harpilini) are associated with Scolytids (Kenis et al. 2004).

Similar to the ground beetles, tiger beetles (Cicindelidae) are large beetles and both the larvae and adults are predaceous, with the adults being particularly voracious. Usually, adult tiger beetles are found in open sandy areas or paths and seek out their prey. Larvae create vertical burrows in compact soil and are sit-and-wait predators (White 1983).

Another predaceous beetle family is the hister beetles (Histeridae). The diet of adults and larvae consists almost completely of insects and other small animals. Usually these beetles are found in dead or dying tree bark or in galleries under the bark surface (White 1983). Staphylinid, or rove beetle, adults and larvae are also predominantly predaceous insects. A few of these beetles are known to be parasitic (Borror and White 1970). Rove beetles are one of the largest families of beetles and about half are found in leaf litter. They comprise one of the most ecologically important and common insect constituents of soil fauna (Bohac 1999). The majority of these beetles are non-specific predators that feed on soil arthropods such as nematodes, mites, collembolans, and small insect larvae or immatures. Staphylinids serve as bioindicators of environmental status, especially when measuring human influence on ecosystems (Maleque et al. 2009). These families comprise the predatory beetle group that will be later discussed.

Natural enemies exist aerially as well. Species richness and diversity of hover flies (Diptera: Syrphidae) are positively correlated with stand structural complexity and ground layer vegetation (Maleque et al. 2009). Syrphids are good indicators because they serve varying functional roles in an ecosystem and have diverse habitat requirements. Much like carabids, hover fly diversity is reduced in areas with a closed canopy (Humphrey et al. 1999). Parasitic wasps are complex and specialized because of their high trophic position, intricate biology, narrow host ranges, and habitat requirements. These wasps are found to be more abundant in species rich mixed woodland habitats than in coniferous woodlands (Maleque et al. 2009).

Spiders respond to natural and anthropogenic disturbances and are indicators of ecosystem changes caused by forest fires, vegetation development and changes in stand complexity (Maleque et al. 2009). Spiders and other arthropods play a large role in ecological

features of soil and detritus of forest floors. Spiders are among the most essential entomophagous predators in an ecosystem and a reduction in abundance can interfere with ecosystem function (Moulder and Reichle 1972).

Earthworms and Nematodes

Earthworms (Oligochaeta) are well known as burrowers in soil systems and are important bioindicators. Their burrowing and feeding activities add inorganic residues to the soil, as well as enhance decomposition, humus formation and nutrient cycling. They also play a critical role in soil structure development (Werner 1990). Distribution of earthworm species is strongly influenced by the physiographic region. In Alabama, 43% of earthworm species are exclusively found in the coastal plain (Reynolds 1994).

Nematodes are roundworms (Nematoda) and second only to arthropods in number of species and individuals present. There are two main groups of nematodes: free-living and parasitic (Wharton 1986). Free-living nematodes live in the soil or water. Nematodes can be plant feeding, animal feeding (parasitic guilds) or bacterial/fungal feeders. About 48 % of nematode genera are parasites of plants and animals (Wharton 1986). In the pine system, nematodes primarily affect seedlings. The stunt nematode, *Tylenchorhynchus claytoni*, and a stubby-root nematode, *Paratrichodorus minor* invade southern forest nurseries (Cram and Fraedrich 2007). Ruehle (1973) investigated the influence of plant-parasitic nematodes on longleaf pine and found several nematodes that decreased weight of seedlings. These include *Holoplolaimus galeatus*, *Tylenchorhynchus claytoni*, *Meloidodera floridensis* and *Pratylenchus brachyurus* (Ruehle 1973). Overall, nematodes are not considered a problem in nurseries because their populations are controlled by soil fumigation with methyl bromide (Cram and Fraedrich 2007).

Nematodes are one of the major biotic factors impacting bark beetle populations. Members of the *Contortylenchus* genus are probably the most familiar parasitic nematode on bark beetles. Most nematodes associated with bark beetles are true parasites; only a few will kill the

host. The most pronounced effect seen on bark beetles from internal parasites is brood reduction. This is because, in most cases of mortality, it is the female that perishes (Massey 1974).

Cogongrass Morphology, Biology, and Distribution

Cogongrass, Japanese bloodgrass, or alang-alang (*Imperata cylindrica* (L.) Beauv) is a long-lived, invasive C₄ grass. Morphologically, leaves are a lime green color (0.3–1.5 m long, 1.2–2.5 cm wide) with a frequently off set midrib and minutely serrated leaf margins. Except for the flowering stalk, cogongrass is virtually stemless aboveground (Dozier et al. 1998, MacDonald 2004). The root and rhizome system can, and often does, compose 60% or more of the total plant biomass and is found in the upper 15–40 cm of soil (MacDonald 2004, Daneshgar and Jose 2009). Rhizomes are long, white, tough and scaly with very short internodes. The branched rhizomes create a dense mat that excludes most other vegetation. Apical ends of rhizomes are sharp and may grow through roots of other plants, thus enhancing its ability to debilitate other plant species (Dozier et al. 1998, MacDonald 2004). Unlike seeds, rhizomes can invade undisturbed pine savannas (Lippincott 2000). Site to site variation can occur, but a single ramet can have over 350 descendant shoots in just 6 weeks under optimal conditions. After 11 weeks, clones can of a ramet cover at least 4 m² (Brewer and Cralle 2003). Soil contaminated with rhizome fragments as small in size as 0.1 g can be an important mechanism of cogongrass spread (Dozier et al. 1998, Daneshgar et al. 2008, Daneshgar and Jose 2009). Cogongrass rhizomes can also penetrate rocky soils and even asphalt pavement (Bryson and Carter 2004).

Plants typically flower in the early spring, but flowering may occur year round in Florida (Van Loan et al. 2002). Flowers are a terminal, cylindrical panicle, typically 10–20 cm long, comprised of spikelets (MacDonald 2004). As many as 460 white spikelets, approximately 3–6 cm in length are found on each panicle (Dozier et al. 1998). Panicles are silky and become fluffy with maturity. Seeds are viable for approximately one year and can germinate immediately after harvest (MacDonald 2004). There are contrasting reports on how critical seed production is for

reproduction (Sajise 1972, MacDonald et al. 2004). Human mediated activity, water, and animals disperse seeds, or they may be carried by wind within 15m of the plant or up to 25 km in open areas (Bryson and Carter 2004, Daneshgar et al. 2008). Anthropogenic means of spread are much worse than natural factors (Capo-chichi et al. 2008) and soil disturbance (i.e., discing, tilling, road creation) encourages seeding establishment (Brewer and Cralle 2003). Cogongrass spread in Alabama can be linked to the northeastern winds from the gulf as well as seed spread along roadways (Dozier et al. 1998).

Globally, cogongrass is ranked number seven among the ten worst weeds (Coile and Schilling 1993, MacDonald 2004, Brewer 2008) because of severe impacts on agriculture and forestry. It is a major hindrance to reforestation efforts in Southeast Asia and the number one weed in agriculture production in many parts of Africa (Brewer and Cralle 2003). Cogongrass was accidentally and intentionally introduced into North America. Populations introduced in the early 1900's to Grand Bay, Alabama, have had significant impacts on native habitat in the southeastern United States (Jose et al. 2002, Dozier et al. 2008). At first, its distribution was slow, occupying 1235 ha by 1950 (Dickens 1974) but current estimates surpass 200,000 ha of land primarily in southwest Alabama (Capo-chichi et al. 2004, Brewer 2008; Figure 1.2).

Ecosystem Impacts of Cogongrass

Cogongrass invasiveness can be attributed to its being adaptable to poor soils, having high drought tolerance, wind-disseminated seed production, fire adaptability and high genetic plasticity (MacDonald 2004, Dozier et al. 1998). Cogongrass also possesses allelopathic characters (Bryson and Carter 2004, Coile and Shilling 1993, Daneshgar et al. 2008) that characters inhibit growth, germination, and survival of other plants (Daneshgar et al. 2008) and aid in the creation of a monotypic expanse (Coile and Shilling 1993). Cogongrass has not become a threat to agricultural production in the US because tillage and herbicides relegate it to field edges. However, as its frequency increases worldwide, ecologists and conservationists fear

cogongrass will displace native animal and plant species as well as alter the fire regime in some forest ecosystems (Brewer and Cralle 2003).

Its fire tolerant capabilities provide it with an advantage over non-tolerant, usually native, species. Cogongrass fires burn about 15-20% hotter than other groundcover. However, this temperature effect is not seen on the surface, but at 1.2 - 1.8 m off the ground (Duever 2008). Rhizomes persist below the soil surface and produce new shoots quickly following a burn (Bryson and Carter 2004). Cogongrass' dense rhizome mat creates a high root to shoot ratio allowing it to thrive after fire or mowing. Burns in grass-invaded areas tend to be more frequent and have a greater spatial expanse (Daneshgar et al. 2008).

Cogongrass invasion can cause significant declines in herbaceous plants in longleaf pine savannas and reduce the species richness of native flora. Brewer and Cralle (2003) found that the older the infestation, the lower the richness of native plant species. Cogongrass stands create deep shade, which can displace lower growing native plant species. Cogon-tolerant native species therefore are mostly taller and woodier plants. For example, yaupon (*Ilex vomitoria*), wax myrtle (*Morella cerifera*), and persimmon (*Diospyros virginiana*) all possess the potential to displace smaller native species, especially after long periods of suppressed fire (Glitzenstein et al. 2003). Unlike cogongrass, these species lack aggressive rhizomes and do not quickly recover from fire (Brewer 2008). In the longleaf pine ecosystems, already in danger, cogongrass presence could push endemic plant species to threatened and even endangered listings (Brewer 2008). Brewer (2008) concluded that even though most suggest cogongrass poses a greater threat because of the altered fire regime, reduced plant diversity might be a more pressing conservation concern.

Despite the rate at which cogongrass can and will overtake a community, little is known about its affect on invertebrate populations. Native plant and animal species tend to be most susceptible to invasion by exotic species because they are usually specialists and inferior competitors or they lack defense mechanisms and high reproductive outputs (Brewer 2008).

Cogongrass Management

A variety of tools have been used for cogongrass management including biological, physical, chemical and cultural methods. The key to success in cogongrass management lies in the ability of a given treatment to successfully kill it or suppress its recovery from the extensive rhizome network that cogongrass develops (Dozier et al. 2008). Repeated tillage, or discing, which fragments the rhizome layer, has been useful and is primarily why cogongrass is not a greater problem to agriculture production in the southeastern US. Repeated mowing is only helpful for seed head suppression because shoot recovery is rapid. Both tilling and mowing provide temporary suppression of cogongrass, but are not effective in providing long-term control and are often impractical and unrealistic for large highly infested areas (Dozier et al. 1998). Burning is useful for removing dense cogon thatch but provides no long-term control when used alone. Burning and mowing remove old growth and dead biomass (thatch) and weaken rhizomes through allocation of starch resources to make new shoots. Also, removal of the dense thatch layer makes secondary management strategies more successful.

Glyphosate and imazapyr are the only herbicides that significantly impact cogongrass and are approved for use in pine ecosystems. Imazapyr is an acetolactate synthase (ALS) inhibitor. The ALS enzyme is a key enzyme in production of three amino acids (valine, leucine, and isoleucine). Blocking ALS activity prevents the conversion of pyruvate to acetolactate (eventually to valine and isoleucine) and conversion to 2-ketobutyrate to 2-acetohydroxybutyrate and eventually isoleucine (Senseman 2007). Imazapyr is a non-selective, foliar, and soil active herbicide that is generally applied post emergence. Symptoms of imazapyr toxicity appear after 3–10 d and after 17–24 d plant will show a deep red color and symptoms of chlorosis, leading to necrosis and plant death (Senseman 2007). However, symptoms may not be expressed on cogongrass for up to 60 days. Imazapyr has considerable soil residual activity that may interfere with seedling establishment and injure or kill other species where roots are in the treated area.

Glyphosate is a 5-enolpyruvyl shikimate 3-phosphate (EPSP) synthase inhibitor. It inhibits the activity of EPSP synthase, which converts shikimate-3-phosphate plus phosphoenolpyruvate to 5-enolpyruvyl shikimate-3-phosphate; this inhibits production of aromatic amino acids: tryptophan, tyrosine and phenylalanine. Like imazapyr, glyphosate is non-selective with symptoms of growth cessation beginning within 3–10 d. After 17–24 d, plants will show dehydration and desiccation and eventually necrosis and plant death (Senseman 2007). High application rates of imazapyr and glyphosate cause rapid foliar kill, but intermediate levels have proven to be more effective (Dozier et al. 1998). In fact, the most effective method of herbicide treatment involves multiple applications. Most effective multiple treatment management strategies work best under the earliest developmental stages. Older developmental stages show a susceptible foliar reaction, but rhizomes are slightly resistant to glyphosate. Also, basal area of the leaf proved to be the most likely area to absorb glyphosate. Seasonal differences can affect the impact herbicides have on cogongrass management strategies (Dozier et al. 1998). It is critical to use the correct rate during the appropriate time. Herbicide applications can be expensive, up to \$400 ha/year, and multiple applications per year may be required (Capo-chichi et al. 2008).

Integrating burning, mowing, discing, herbicides, and cultural control will increase the prospect of cogongrass suppression. First discing in the fall, then following with an herbicide application has the potential of providing over 90% control and suppression. The seedlings' under developed tissues are a perfect target and offer maximized absorption and increased efficacy. Following fall mowing and herbicide treatments, it is essential that desirable plant species be planted. In order to remove cogongrass more effectively, it must be replaced or it will regain dominance in the empty niche. Careful consideration must be taken when deciding which species to include. If the planted species cannot withstand cogongrass, they will be easily taken over and a monotypic expanse will eventually be recreated. Some successful characters of the new species include herbicide tolerance, as well as tolerance of soil type, canopy cover and other factors

(Dozier et al. 1998). Revegetation is very useful to suppress cogongrass recovery following control (MacDonald 2004) and is an integral component of ecologically based weed management.

Long term, highly intensive management strategies have not worked so far. The only known survey of insects within cogongrass in the US focused solely on herbaceous feeding insects as potential biological control candidates (Minno and Minno 1999). From this work, 51 insect species were observed in cogongrass, but no suitable insects for the purpose of biological control were found. In Indonesia, Maeto et al. (2009) found reduced braconid abundance and species richness in *Imperata* grasslands compared to *Acacia* plantations and old secondary forests. Few promising candidates have been found for biological control and none are currently available (Van Loan et al. 2002).

Interactions

Root-feeding bark beetles (*Hylastes* spp.) spread ophiostomatoid fungi that are linked to pine decline (Eckhardt et al. 2004). Pine decline in the southeastern United States is another growing problem that also severely impacts both the environment and the livelihood of the Southeast (Eckhardt et al. 2008). Loblolly pine decline (LPD) is an example of a complicated syndrome involving the host, insect, pathogen and site interactions (Eckhardt et al. 2007). Symptoms of LPD are seen in trees on well-drained sites as sparse crowns, short chlorotic needles, and gradual mortality. Older trees begin to decline slowly and die prematurely. Eventually, LPD spread by root-feeding bark beetles to younger trees and the onset of symptoms associated with fungal and insect species are seen (Eckhardt et al. 2004, Eckhardt et al. 2007). Trees over 40 years old are most frequently weakened by LPD. These declined trees are more prone to southern pine beetle (SPB) attack (Otrosina et al. 1997).

Current studies of pine stands in the southeastern United States containing trees expressing symptoms of decline show significantly higher numbers of root-feeding bark beetles and the presence of ophiostomatoid fungi (Eckhardt et al. 2007). Species of *Leptographium* and

Ophiostoma are most commonly reported as causes of blue stains in pine, spruce and other conifers. These stain fungi rarely kill healthy trees, but they do reduce the value of timber and may be associated with mortality in stressed trees (Wingfield et al. 1998). *Leptographium* species are commonly associated with various species of root-feeding bark beetles (primarily *Hylastes* spp.) and root weevils, which attack healthy and stressed trees (Eckhardt et al. 2007). These insects serve as vectors of these fungi and as wounding agents creating courts, which permit fungal infection. In a study done on loblolly pine in Alabama, Eckhardt et al. (2007) found that symptomatic sites were associated with increased numbers of insects. Root and lower stem feeding beetles were significantly more abundant in symptomatic sites when compared to asymptomatic ones. The root-feeding beetles considered for this study were: *Hylastes salebrosus*, *H. tenuis*, *Hylobius pales* and *Pachylobius picivorus*. Beetles collected were associated with *Leptographium procerum*, *L. terebrantis* and *Grosmannia. serpens* and could be serving as vectors (Eckhardt et al. 2004).

Hylastes spp. are found in greater numbers within pine trees that are symptomatic for pine decline when compared to asymptomatic pines (Eckhardt et al. 2007). Although the beetles are responsible for vectoring fungal spores, the fungus alone does more damage because it interacts with the vascular tissue of the trees. While not completely understood, pine decline has been attributed to a combination of tree stress, native and exotic insects, and native and exotic fungi that can severely stress or kill species such as longleaf, shortleaf, and loblolly pine (Eckhardt et al. 2008). While these arthropods, disease, and weeds commonly co-occur, little is known concerning how these pests interact or if cogongrass promotes cascading negative impacts within pine ecosystems (Daneshgar et al. 2008).

Similarly, burning and herbicide strategies used for managing cogongrass can also impact the abundance of key arthropods in the pine system (Burke et al. 2008). Loblolly pine managed with burning and imazapyr actually had greater arthropod diversity relative to control plots, however, impacts among taxonomic groups varied widely (Burke et al. 2008). Abundances of

spiders and Orthoptera were impacted by management strategy but there was no treatment effect among Coleoptera, Lepidoptera, Diptera, Hemiptera, or Hymenoptera collected in pitfall or sweep net samples (Burke et al. 2008). Glyphosate applications can negatively affect survivorship and behavior of wolf spiders (Evans et al. 2010). These effects can be direct (Evans et al. 2010) or indirect impacts related to reductions in vegetation height and increases in dead vegetation resulting from treatment (Bell et al. 2002). Weed management can also alter the prey taken and ant abundance. In weed-free areas, smaller ant populations were recorded and a greater number of nests were abandoned (Reagan 1986). Similar management strategies for cogongrass in longleaf pine have not been assessed for their potential impacts on arthropods. Reductions in generalist predators and other natural enemies would potentially reduce predation on root-feeding bark beetles and possibly increase the incidence of pine decline in cogongrass-infested pines.



Figure 1.1. Longleaf pine stand with shrub and forb understory

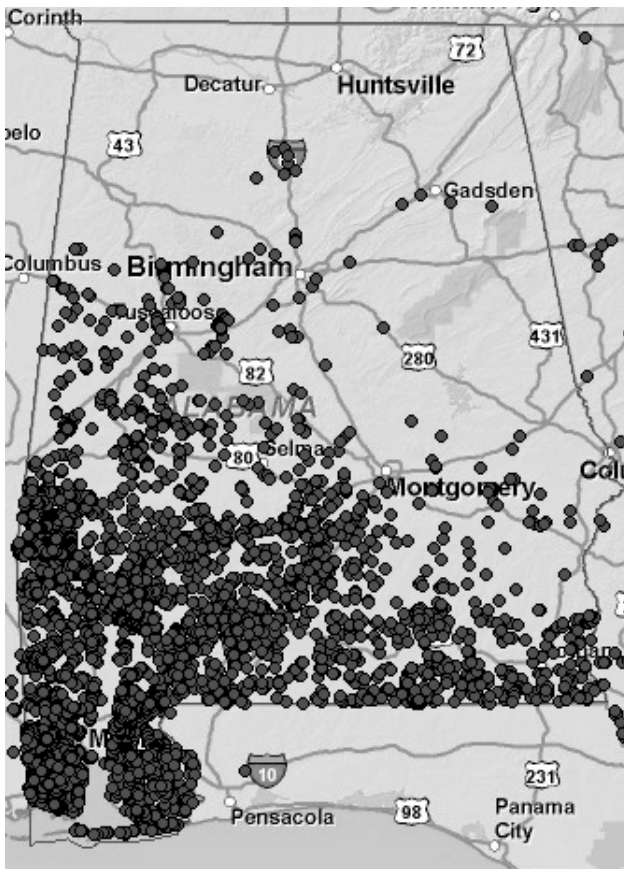


Figure 1.2. Map of Cogongrass distribution in Alabama in 2009

Chapter 2

Impact of Cogongrass Management Strategies on Generalist Predators of *Hylastes* spp. in Longleaf Pine

Abstract

Cogongrass (*Imperata cylindrica* Beav.), an aggressive, invasive weed, is threatening the integrity of longleaf pine (*Pinus palustris* Mill.) ecosystems. Various management strategies are used to suppress cogongrass such as fire, herbicide, and seeding with native plants. To understand the effect management strategies have on arthropod natural enemies and root-feeding bark beetles a split plot design to test fire (whole plot) and 4 sub plot treatments: control (no additional treatment), herbicide, seeding, and herbicide plus seeding were used. Arthropods were sampled using pitfall traps and sweep samples. After two years of sampling, counts of all natural enemies were not significantly affected subplot by treatment but were affected by burn treatment. Upon subdividing natural enemies into groups, only spiders were significantly affected by subplot treatment, but predatory beetles and ants were significantly affected by burn treatment. Root-feeding bark beetle (*Hylastes* spp.) abundance was not significantly affected by treatment or burn treatment. Cogongrass management strategies overall do not affect natural enemy populations. Undisturbed cogongrass provides the best habitat for natural enemy assemblages.

Introduction

Cogongrass (*Imperata cylindrica* Beav.) is an aggressive, invasive, perennial, C₄ grass originating from south East Asia that was introduced to south Alabama around 1910 and again in the mid 1940s (Dozier et al. 1998). Since its introduction, it has spread into all states bordering Alabama, as well as Louisiana, Texas, and South Carolina. Cogongrass changes the fire regime and inhibits commercial forestry and reforestation efforts (Lippincott 2000). Invasion of cogongrass into longleaf pine (*Pinus palustris* Mill.) stand results in a monotypic understory (Lippincott 2000) and established pines may begin to decline (Daneshgar et al. 2008). Longleaf pine ecosystems are typically composed a diverse understory of graminoids, shrubs, forbs, vines and legumes (Daneshgar and Jose 2009). Burning is an important regulator of longleaf pine stands. In the presence of cogongrass, prescribed or natural fires are higher and hotter, which causes more damage to trees and understory plants than a normal, cogongrass-free burn (Bryson and Carter 2004).

Control of cogongrass requires an integrated approach of cultural and chemical controls (MacDonald 2004). Natural or prescribed burning, although not recommended for control, removes the thatch layer and forces the plant to allocate carbohydrates to shoot growth, temporarily weakening the rhizome (Brewer 2008). Mowing is also not a recommended method of control but is used for seedhead suppression, especially on roadsides. Because 60% of cogongrass biomass is underground in the extensive rhizome mat, it can also quickly recover after a fire or mowing. Discing alone does not provide control, but can reduce biomass (Brewer 2008). Discing followed by late fall herbicide

application, however, can yield greater than 90% control (Dozier et al. 1998). Native plants generally do not recover as quickly as cogongrass, and plant diversity is reduced after disturbances due to displacement (Brewer 2008).

Herbicides, mainly imazapyr and glyphosate (MacDonald 2004) are effective, widely used controls used for cogongrass. However, multiple applications for several years are required (Capo-chichi et al. 2008). High application rates of herbicides will result in rapid foliar death, but to successfully kill cogongrass, rhizomes must receive a lethal dose of herbicide (Dozier et al. 1998). Herbicides alone can be expensive (\$400 ha/yr) (Capo-chichi et al. 2008). For long-term control, establishing desired plant species to ultimately replace cogongrass is essential after suppression or removal with herbicides (Dozier et al. 1998). These revegetation treatments are done by introducing native plants that produce heavy shade and dense rhizomes that can compete with cogongrass and prevent recolonization (Yager et al. 2011, Dozier et al. 1998).

Although not very well studied, all of these aforementioned control tactics for cogongrass may have measureable effects on the arthropod community. Longleaf pine invaded by cogongrass would produce hotter fires subjecting trees to increased stress (Lippincott 2000). Beetles in the genus *Hylastes* (Curculionidae, Scolytinae) are root-feeding bark beetles and cannot attack and kill healthy trees, unlike the southern pine beetles (*Dendroctonus frontalis* Zimmerman). *Hylastes* are attracted to stressed, already weakened trees. Environmental disturbance like hotter, more frequent fire can weaken trees, stressing them causing them to be and more susceptible to beetles and root infections. Changes in the structure and cover of vegetation resulting from burning or herbicide application may impact generalist predators such as ants (Castano-Meneses and

Palazzo-Vargas 2003, Matsuda et al. 2011), spiders (Bell et al. 2002, Burke et al. 2008) or predatory beetles (Maleque et al. 2009, Holland and Luff 2000). Populations of parasitic Hymenoptera are negatively impacted by cogongrass invasion into forest systems, but populations can recover through replanting and increased plant diversity (Maeto et al. 2009).

The objective for this study was to understand how cogongrass management strategies used in longleaf pine communities would influence root-feeding bark beetles and diversity and abundance of invertebrate natural enemies. Treatments such as burning locally increase bark beetles (Bauman 2003) whereas the recovery of plant diversity following removal of cogongrass should increase natural enemies.

Materials and Methods

Site Description. Two sites in southwest Alabama, located within open longleaf pine stands approximately 10 years old, were selected for study. Site one was located in Mobile County, AL near the Mississippi State line on Airport Road and will hereafter be referred to as the “Rainwater” site. Site two was located in Baldwin County, AL near Bay Minette on White House Fork Road and will hereafter be referred to as the “Beebe” site (Figure 2.1). Each site was heavily infested with established swards of cogongrass, which completely dominated the understory. Both sites had similar soil characteristics including texture, pH, CEC, and water holding capacity. However, soil organic matter was slightly higher at the Rainwater site than the Beebe Farm site (Table 2.1). Longleaf trees at each site ranged from 3.4 to 12.2 m in height and 11.4 to 25.4 cm diameter at breast height (DBH)

Experimental Design and Treatments. At each site, we utilized a split plot experimental design to allow for a consolidated prescribed burn. The whole plot treatments were prescribed burning or no burning. Fire breaks were established around the perimeter of the burned portion of each site to prevent fire escape. The prescribed burn was conducted by a private contractor in early March of 2010. Since cogongrass is a pyrogenic species that is not well suited for safe prescribed burns, a backing fire was used to minimize fire severity and prevent more severe conditions that typically occur when cogongrass is burned.

Following the burning treatment, four cogongrass management treatments were imposed on 10 by 10 meter subplots in the burned and unburned main plots: 1) an untreated control; 2) disking and seeding with native herbaceous species; 3) repeated glyphosate treatment, and 4) an integrated treatment consisting of glyphosate treatment followed by disking and seeding with native herbaceous species and subsequent spot treatments of glyphosate as necessary. Subplot treatments were randomly assigned and replicated five times within each main plot at each site except for the seeding only treatment, which was replicated three times within each main plot. Plots were surrounded by a 1.5 m buffer that was treated with glyphosate as needed to prevent cogongrass spread between plots.

In 2010, plots assigned to the glyphosate herbicide treatment received the herbicide glyphosate (Accord XRT 2; 646 g ai per liter; Dow AgroSciences, Indianapolis, IL) at 2.5% and 4% v/v solution in the spring and fall respectively. The spring treatments were broadcast applied to each plot on April 14, 2010 and fall treatments on October 14, 2010. In 2011, glyphosate was applied as a spot treatment only to cogongrass within plots on November 11. For all applications, glyphosate treatments were applied at 514 l/ha (55 gpa) with a single nozzle Solo CO₂ pressurized backpack sprayer.

Plots receiving the disking and seeding treatment also received glyphosate treatments as previously described. A mixture of commercially available, competitive, native species (Table 2.2) was selected to create a desirable herbaceous understory. This species mix is a functionally diverse mix of species that are drought tolerant, create heavy shade, grow densely, are N-fixers, and are rhizomatous or deeply rooted. The species in the mix also provide numerous benefits including habitat and food sources for insects and wildlife. The plots were seeded at a total seeding rate of 40 kg per hectare (36 lb/a) with varying percentages of each species in the mix (Table 2.2). After year one, the species mix was altered due to an extremely low germination and establishment rate due to drought. Four species were removed, and six were added (Table 2.3). The seed mix was sown by hand in April of 2010 and February of 2011. Each plot was divided into 1.5 m strips and an equal portion of seed was then hand broadcasted across each strip. Before

seeding, plots were lightly disked (5-8 cm deep) to break up cogongrass thatch and provide bare soil for seed contact.

Arthropod Sampling. Arthropods were sampled using two methods: pitfall traps for ground active arthropods, and sweep samples primarily targeting arthropods associated with vegetation. Five pitfall traps (15 cm dia funnel type traps, Multi-pher, Bio-Controle, Quebec City, Quebec), were installed in each plot, arranged in an X pattern 3 m apart along each diagonal with one in the center (Figure 2.2). Inside each trap, a 453 ml plastic cup (16K, Dart, Mason, MI) was filled with 50 ml of a 1:1 (v/v) mixture of water and propylene glycol (Peak, Old World Industries, Northbrook, IL) to serve as a preserving solution. Every 2 wk, contents from all five pitfall traps were pooled into a labeled plastic container. Empty traps were then recharged with preserving solution. Pitfall traps were active from May 1 through Oct 28, 2010. Pitfall traps remained empty in the field overwinter. In 2011, traps were activated on Feb 15 and serviced biweekly through Oct 25. Soil displaced from pitfall trap installation was collected and brought back to the AU soil dynamics lab for analysis.

Sweep samples were taken biweekly, concurrent with servicing pitfall traps. One sample was made per plot, following each diagonal making approximately 30–40 sweeps per plot. Insects collected in the sweep net were put into a 3.8 liter plastic zipper bags and placed into a cooler for transport back to the laboratory.

Inside the laboratory, arthropods from pitfall traps contents were filtered to remove dirt and other debris. The remaining arthropods were stored in 50 ml plastic vials with 90% ethanol in a freezer until they were sorted. Arthropods were sorted into taxonomic groups and stored in 22 mL glass vials with 90% ethanol. The taxonomic

groups of interest were: predatory beetles (Staphylindiae, Carabidae, Cleridae, Histeridae, Cicindelidae, Coccinellidae, and predatory larvae), ants (Formicidae), spiders (Aranae), all of which are bioindicators in forest systems (Maeto et al. 2009, Maleque et al. 2009) and important in ecosystem services (Isaacs et al. 2009).

In the lab, sweep samples were placed in the freezer for ≥ 1 d. Arthropods were then removed from the bags and placed into 15 mL vials by plot in 90% ethanol. From these samples, Formicidae, Aranae, hover flies (Syrphidae), and parasitic Hymenoptera were identified and counted.

In addition to monitoring beneficial arthropods, root-feeding bark beetles (Scolytinae: *Hylastes* spp.) were also collected in baited pitfall traps commonly used for root-feeding or butt feeding beetles in the pine system (adapted from Eckhardt et al. 2007). Pitfall traps were located 1 m from the center in six plots per treatment on both sites. Each trap contained an insert with two freshly cut pine stems and two glass vials, one with 8 ml of 95% ethanol, and the other with 8 ml of turpentine. The interior of each trap was coated with liquid Teflon™ (Northern Products, Woonsocket, RI) to discourage insect escape. Eight evenly spaced entrance holes were drilled around the pipe, as well as two smaller holes in the bottom for drainage. Traps were inserted so that entrance holes were at ground level. Traps were baited and serviced biweekly from May-Oct in 2010 and Mar-Oct in 2011. The number and species of beetles collected were sorted to species and counted.

Weather data were obtained for the 2010 and 2011 sample seasons from Alabama Mesonet Weather Data (AWIS Weather Services Inc) stations in Grand Bay and

Fairhope. Mean monthly temperatures and average monthly rainfall were summarized for each site (AWIS 2008).

Analyses. Counts of insects for each taxonomic group of natural enemies, total natural enemies (all groups collectively), and lower bole and root-feeding bark beetles and weevils were analyzed by fitting each insect count using a Poisson generalized linear mixed model (GLMM) with log-link function (PROC GLIMMIX, SAS Institute 2002). GLMM allows for a Poisson regression (insect counts) with the inclusion of random effects in the model. A Poisson GLMM with repeated measures was fitted to the count data. The random effects were sampling error and plot error. Compound symmetry (CS) was used for the within-subject covariance structure. If significant, pairwise mean comparisons among main effects and interactions were made using the least-squares means (LSMEANS) method.

Results

Total arthropod natural enemies (>58,000 individuals) collected over the 2 years of sampling were dominated by ants, spiders, and predatory beetles, with parasitic Hymenoptera and hover flies accounting for <1% of the total (Figure 2.3). Cogongrass management treatments, including the revegetation treatment, did not significantly affect total counts of natural enemies, however, there were significantly ($P < 0.0003$) fewer natural enemies in burned plots (Figure 2.4) (Eckhardt 2003). Although not all groups revealed a significant burn response, total natural enemies, predatory beetles, ants, and spiders all significantly differed in site by burn interactions (Figure 2.5, 2.6, 2.7, 2.8). The most total natural enemies and predatory beetles were found in not burned Beebe plots. Ants and spiders were more abundant on not burned Rainwater plots. There was no

significant treatment effect on predatory beetles or ants among cogongrass management treatments (Figures 2.9 and 2.10), but there were significantly more spiders in the untreated control plots (Figure 2.11). Differences were also apparent at the whole plot. For all taxa except spiders, 2011 had significantly more arthropods collected than 2010.

Predatory beetles were represented by: Staphylinidae, Carabidae, Histeridae, Cicindelidae, and Coccinellidae. Staphylinidae and Carabidae were the most abundant (about 58% and 39% respectively) of the predatory beetles, followed in abundance by Histeridae, Cicindelidae, and then Coccinellidae, which accounted for about <2%. No Clerids were collected. Predatory beetle larvae (Elateridae, Cantharidae) were also included but accounted for <1% of the total predatory beetles. Predatory beetle populations had peak collections in June–July 2010, March 2011, and October 2011 (Figure 2.9).

Ant captures were greatest in summer (June–Sept) of each year (Figure 2.6) with *Solenopsis invicta* Buren accounting for > 98% of ants collected. Spiders had very distinctive peaks in Sept 2010 and June 2011. Almost consistently through the 2011 sample period, fewer spiders were found in the seeding plus herbicide, or eco-based, plots (Figure 2.11). Spiders were mostly represented by Lycosidae, Gnaphosidae, Liocranidae, and Ctenidae families with lycosids and gnaphosids accounting for the largest portion. In 31 samples across 2 years, only 164 parasitic Hymenoptera and 21 syrphid flies were collected: too few collected to run statistical tests for differences by treatment. Over two years, 381 *Hylastes* spp. were collected and their abundance showed no significant effect by treatment (Figure 2.12) or by burn treatment.

Beebe site (Baldwin County) also had significantly more insects for each taxonomic group than the Rainwater site (Mobile County). Of all of the natural enemy groups, predatory beetles differed the most between the two sites; more beetles were found on Beebe. Beebe was also host to a larger carabid morphotype (*Pasimachus* sp) not collected on the Rainwater sites. Mean temperatures were relatively similar throughout both collecting seasons (Figure 2.13). Rainwater had greater levels of precipitation in 2010 than 2011 (Figure 2.14).

Discussion

In this 2-year study, we determined how management strategies for cogongrass impacted all arthropod natural enemies and impacts on ants, spiders, and predatory beetles, which were the dominant predators in our samples. This is the first study investigating the effects of management strategies for cogongrass on arthropods. Overall, all arthropod natural enemies were not significantly affected by treatment (subplot), and only spiders were significantly greater in the control treatment. When compared to other treatments, seeding + herbicide had significantly fewer spiders. The greatest number of spiders were collected on control plots, which were the only ones left undisturbed. Seeding + herbicide plots, disturbed twice, yielded the fewest spiders and the greatest differences between other treatments. Complete recolonization was required on the seeding + herbicide plots, but on the control plots spiders present remained undisturbed. These findings reiterate spiders' sensitivity to disturbance (Maleque et al. 2009). Given that most of the spiders collected were lycosids and gnaphosids, ground dwelling instead of plant residing spiders, these differences may not be due directly to vegetation changes.

More than half of the total collected natural enemies were fire ants. Fire ants have the ability to establish rapidly and take advantage of disturbed areas (Lofgren 1986). Since fire ants are more aggressive and have the ability to take over an ecosystem, this could explain the lack of significant differences between treatments. More fire ant mounds were observed on plots with a reduced presence of cogongrass. Since fire ants forage up to 37 m from their mound (Martin et al. 1998), it is plausible that ants would venture into nearby plots and return to the mound. If this is in fact the case, fire ant foraging distance would explain the lack of significance by treatment.

The lack of change in natural enemies, predatory beetles and ants contradicts literature that implicates changes in arthropod communities are seen due to various management strategies (Evans et al. 2010, Maleque et al. 2009, Burke et al. 2008, Guiseppe et al. 2006, Ulyshen et al. 2004, Buckelew et al. 2000). Changes in arthropod communities can be linked to factors such as habitat fragmentation, vegetation change, herbicides and mowing (Maleque et al. 2009, Guiseppe et al. 2006). Cogongrass is reported to drastically change ecosystems (Brewer 2008) and impact animal life (Simberloff et al. 1997), but arthropods seem to follow a different trend. Direct impacts of cogongrass on arthropods and soil invertebrates are being further investigated (Chapter Three).

Few parasitic Hymenoptera, even fewer syrphids, and no checker beetles were collected during this two-year study. These groups are found at the top of the food chain and are easily disrupted if their prey become less abundant or absent (Maleque et al. 2009).

Whether ant populations increase or decrease after fire is dependent on species. (Hanula and Wade 2003, Castano-Meneses and Palacois-Vargas 2003, Jackson and Fox 2006). *Solenopsis* spp. usually survive high intensity burns and species density increases after burning in disturbed areas (Castano-Meneses and Palacois-Vargas 2003). Although spiders were not impacted by the burn treatment, abundances of predatory beetles and ants were greater in the unburned plots. Previous studies on predatory beetles suggest the impact may be family-specific. For example, fire regimes that do not affect carabid populations and diversity may negatively impact staphylinids. Furthermore, these impacts on staphylinids may last more than 30 years post-fire (Gandi et al. 2001). Staphylinidae compose about 60% of the predatory beetles collected and could explain the significant difference among predatory beetles from the burning treatment. Further analysis is being conducted to determine if there is a significant difference by treatment between the two families. Ground and rove beetles (Carabidae and Staphylinidae) live predominantly in the litter layer of the soil. Cogongrass creates a dense but open thatch allowing arthropods and vertebrates to easily navigate. Since cogongrass quickly recovers after a burn, ground cover is only absent for a short period of time (Daneshgar et al. 2008).

Site by burn interactions were inconsistent across natural enemy groups. Sites differences, such as more evergreen vegetation on Rainwater plots, could explain the inconsistencies. Burned rainwater plots consistently had a lower amount of natural enemies across all groups; presence of evergreen woody plants persisting after the burn may have influenced the natural enemy populations (S. Enloe and N. Loewenstein, unpublished data). Since longleaf communities are fire regulated (Brockway et al. 2005), the suppression of fire could allow less fire tolerant species to inhabit the community.

After invasion of cogongrass and an altered fire regime, arthropod communities may respond differently.

Hylastes spp., or root-feeding bark beetles, in this study were trapped at very low numbers. However, they were greater in burned plots but not significantly. Although not significant, the greatest numbers of beetles were captured on the herbicide only plots. Campbell et al. (2008) found that *Hylastes* were most abundant on herbicide plus burn plots in longleaf pine. Longleaf pines are typically less susceptible to decline and environmental as well as forest pest related stressors (Eckhardt and Menard 2009). Longleaf are actually planted as a substitute for loblolly pine (*Pinus taeda* L.) and are better adapted to anthropogenic disturbances (Eckhardt and Menard 2009). The low numbers of *Hylastes* spp. collected could be a reflection of longleaf pine's ability to withstand attack and tolerate LPD.

Total natural enemies and all taxonomic groups except spiders were more abundant in 2011. Yearly differences can be linked to temperature, precipitation or the change in vegetation between 2010 and 2011. Temperatures varied more in 2011 than in 2010 (Figure 2.6). Rainwater (Site 1) had a greater mean precipitation in 2010 compared to Beebe (Site 2) (Figure 2.7). More total natural enemies were collected at Beebe.

We found the management strategy most effective for removing cogongrass to be the seeding + herbicide treatment. Herbicide only plots had thinned cogongrass density, but cogongrass was still present. Seeded only plots revealed quick cogongrass recovery with limited native plant growth (S. Enloe and N. Loewenstein, unpublished data). Between year one and year two, the species mix was changed to the US Forest Service's recommended seed mix for longleaf pine forests. After the cogongrass recovered from

the burn, it overwintered and created a thatch of the prior year's grass. At the start of the 2011 sampling season, control plots looked the same (S. Enloe and N. Loewenstein, unpublished data).

Most cogongrass research focuses on its aggressive behavior (MacDonald 2004, Jose et al. 2002, Dozier et al. 1998), and its impact on forest systems (Daneshgar et al. 2008, Ramsey et al. 2003, Lippincott 2000), and fire regimes (King and Grace 2000, Lippincott 2000). After two years of sampling natural enemies under four different treatments, only one arthropod group, spiders, exhibited a significant effect. Burning had a greater affect on arthropods, with total natural enemies, predatory beetles and ants all found in greater numbers in unburned plots. Natural enemies were not only chosen to serve as indicators of the community, but to monitor the root-feeding bark beetle populations as well. Root-feeding bark beetles, namely *Hylastes*, do not have known specialist predators but a variety of associated generalist predators (Kenis et al. 2004, Moeck and Safranyik 1984). Pine decline is spread by beetles being attracted to stressed and weakened trees (Eckhardt 2004), but we did not find a significant increase in root-feeding bark beetles due to treatments or burn treatments. Natural enemy numbers were high and root-feeding bark beetles numbers were low consistently through all treatments. Cogongrass management strategies did not significantly impact natural enemies. Since Mobile and Baldwin Counties were in a state of drought the past two years, the impact of management strategies could be completely different in a year with average amounts of rainfall. As cogongrass was removed, there was a lack of understory replacement by native species. Eventually, cogongrass with overtake the niche and once again create a monotypic expanse. Given more time and better weather conditions, natural enemies may

begin to resemble longleaf pine without cogongrass present on plots where it was removed.

Table 2.1. Average soil characteristics^a by site

| Site | Burned | pH | Soil type | H ₂ O availability | CEC | % OM |
|--------------------|----------|------|------------|-------------------------------|------|------|
| Rainwater 1 | Burned | 5.12 | Sandy Loam | 0.13 | 0.36 | 3.83 |
| Rainwater 2 | Unburned | 5.08 | Sandy Loam | 0.11 | 0.3 | 4.17 |
| Beebe 1 | Unburned | 5.38 | Sandy Loam | 0.09 | 0.35 | 1.94 |
| Beebe 2 | Burned | 5.26 | Sandy Loam | 0.09 | 0.29 | 1.68 |

^aSoil tests conducted by AU Soil Testing Laboratory.

Table 2.2. Seed Mix for seeding and seeding + herbicide plots in 2010

| Common Name | Scientific Name | Family | Life History | Growth Form | Height (ft) | Min root depth (in) | % of mix |
|---------------------------------|---------------------------------|------------|------------------|--------------------------|-------------|---------------------|----------|
| Common sunflower | <i>Helianthus annuus</i> | Asteraceae | annual | Single stem | 1.5-8' | 8" | 35 |
| Switchgrass | <i>Panicum virgatum</i> | Poaceae | perennial | Bunch grass, rhizomatous | 4-6' | 12" | 32 |
| Partidge Pea | <i>Chamaecrista fasciculata</i> | Fabaceae | annual | Bunch-type | 3' | 14" | 15 |
| Purpletop Tridens | <i>Tridens flavus</i> | Poaceae | perennial | Bunch grass, rhizomatous | 2-5' | 10" | 10 |
| Dixie ticktrefoil | <i>Desmodium tortuosum</i> | Fabaceae | perennial | Single crown | 2-9' | 6" | 2 |
| Coreopsis Greyheaded coneflower | <i>Coreopsis grandiflora</i> | Asteraceae | perennial | Single crown | 1-2' | - | 2 |
| Black-eyed susan | <i>Ratibida pinnata</i> | Asteraceae | perennial | Single crown | 2-4' | 14" | 2 |
| | <i>Rubeckia hirta</i> | Asteraceae | annual/perennial | Single stem | 1-3' | 10" | 2 |

Table 2.3. Seed Mix for seeding and seeding + herbicide plots 2011

| Common Name | Scientific Name | Family | Life History | Growth Form | Height (ft) | Min root depth (in) | % of mix |
|------------------------|---------------------------------|------------|-------------------|-----------------------------|-------------|---------------------|----------|
| Partridge pea | <i>Chamaecrista fasciculata</i> | Fabaceae | Annual | Bunch-type | 2.4' | 14" | 34 |
| Indian grass | <i>Sorghastrum nutans</i> | Poaceae | Perennial | Graminoid | 6' | 24" | 17 |
| Virginia wild rye | <i>Elymus virginicus</i> | Poaceae | Perennial | Graminoid | 2.5' | 16" | 17 |
| Purple top tridens | <i>Tridens flavus</i> | Poaceae | Perennial | Bunch grass, rhizomatous | 2.5' | 10" | 12 |
| Broomsedge | <i>Andropogon virginicus</i> | Poaceae | Perennial | Bunch grass, rhizomatous | 3-5' | 14" | 19 |
| White wingstem | <i>Verbesina virginica</i> | Asteraceae | Perennial | Forb/herb | 4-6' | - | 2 |
| Grey headed coneflower | <i>Ratibida pinnata</i> | Asteraceae | Perennial | Single crown | 4' | 18" | 2 |
| Black-eyed susan | <i>Rudbeckia hirta</i> | Asteraceae | Annual, Perennial | Single stem | 1' | 10" | 2 |
| Perplexed tick trefoil | <i>Desmodium perplexum</i> | Fabaceae | Perennial | Forb/herb | 4' | 12" | 2 |
| Lance-leaved coreopsis | <i>Coreopsis lanceolata</i> | Asteraceae | Perennial | Single crown | 1.5' | - | 2 |
| Spurred butterfly pea | <i>Centrosema virginianum</i> | Fabaceae | Perennial | Vine, Forb/herb | 6-12' | - | 1 |



Figure 2.1. Map of plot locations in Mobile County (Rainwater) and Baldwin County (Beebe).



Figure 2.2. Pitfall traps were arranged in an X pattern 3 meters apart on each 10 x 10 m plot

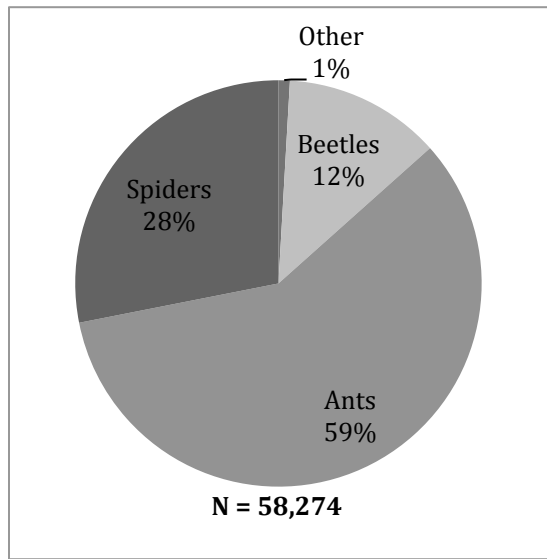


Figure 2.3. Composition of the natural enemy complex in the cogongrass treatment plots from pitfall traps and sweep samples. Samples contained mainly these three groups with parasitic hymenoptera and syrphids comprising the “other” natural enemy group. Percentages reflect total number of individuals collected in 31 sampling periods, 2010 and 2011.

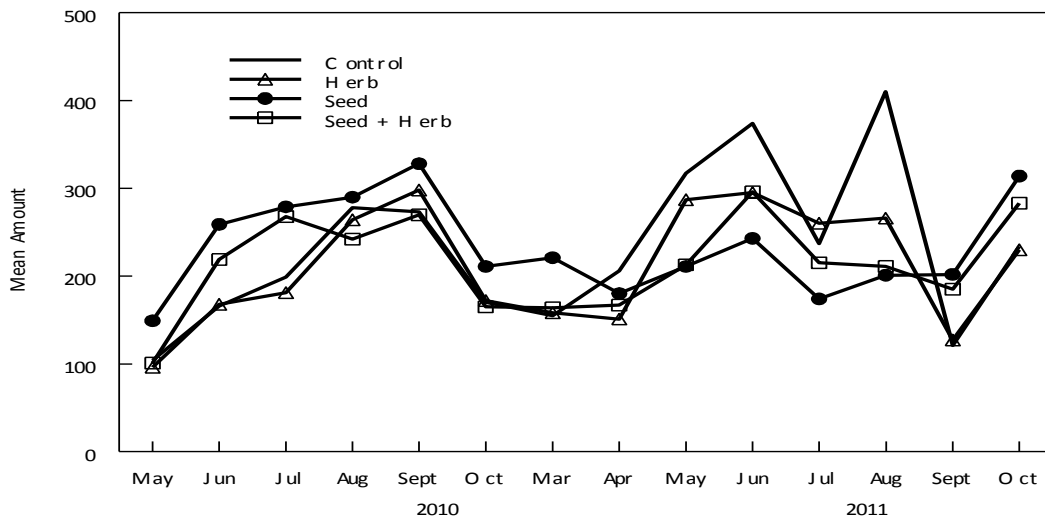


Figure 2.4. Natural enemies collected in each treatment per month over 31 collection trips in 2010 and 2011. Biweekly samples pitfall traps and sweep samples were pooled into monthly total. ANOVA revealed: year (ANOVA, $F=6.72$, $df=1,63$, $P=0.0119$) and treatment ($F=2.06$, $df=3,59$, $P=0.1152$). Significant interaction was: year by site (ANOVA, $F=43.41$, $df=1,63$, $p < 0.0001$).

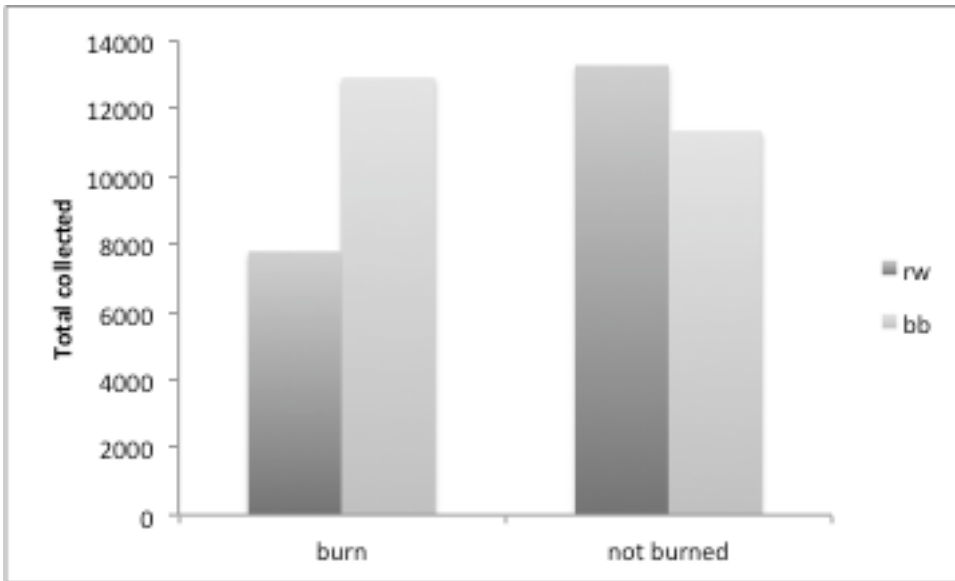


Figure 2.5. Abundance of total natural enemies sampled from May 2010-Oct 2011 from pitfall traps and sweep samples. Main effect of site (ANOVA, $F=10.36$, $df=1,59$, $P=0.0021$), burn treatment (ANOVA, $F=14.69$, $df=1,59$, $P=0.0003$), and site by burn treatment (ANOVA, $F=28.67$, $df=1,59$, $P=<0.0001$) were significant.

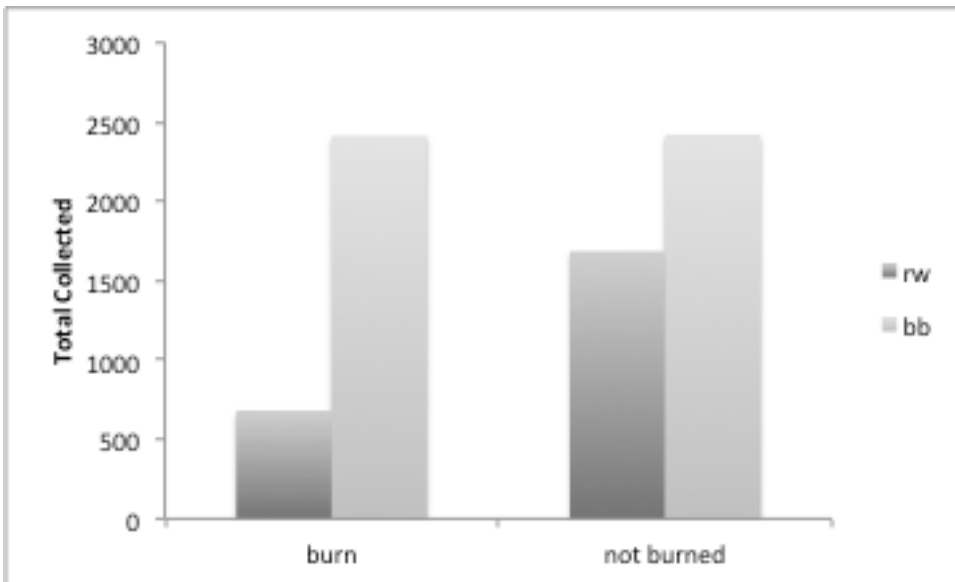


Figure 2.6. Abundance of predatory beetles sampled from May 2010-Oct 2011 from pitfall traps and sweep samples. Main effect of site, burn (ANOVA, $F=29.46$, $df=1,59$, $P=<0.0001$), and site by burn interaction (ANOVA, $F=26.20$, $df=1,59$, $P=<0.0001$) were significant.

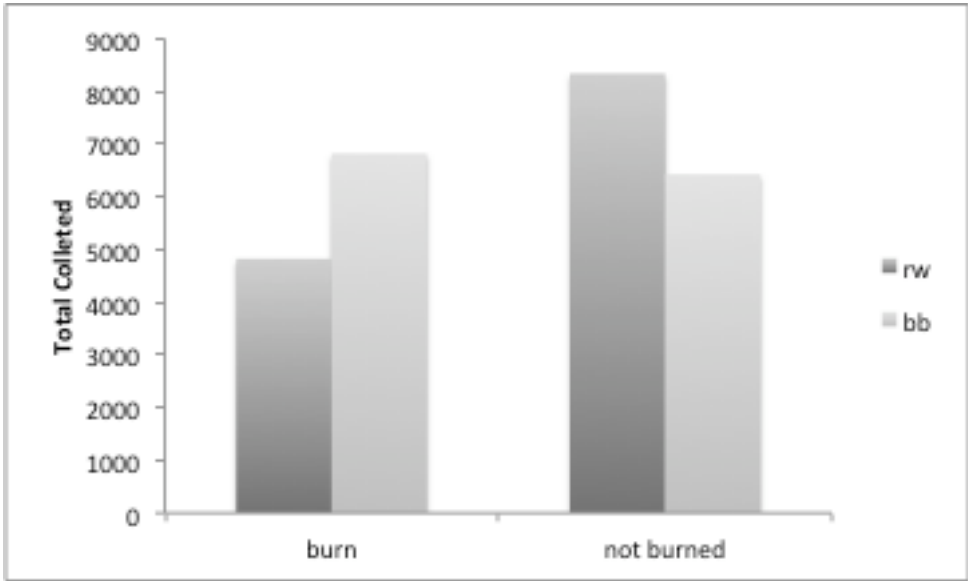


Figure 2.7. Abundance of ants sampled from May 2010-Oct 2011 from pitfall traps and sweep samples. Main effect of site (ANOVA, $F=0.06$, $df=1, 59$, $P=0.8093$), burn treatment (ANOVA, $F=10.89$, $df=1, 59$, $P=0.0016$), and site by burn treatment (ANOVA, $F=10.51$, $df=1, 59$, $P=0.0020$).

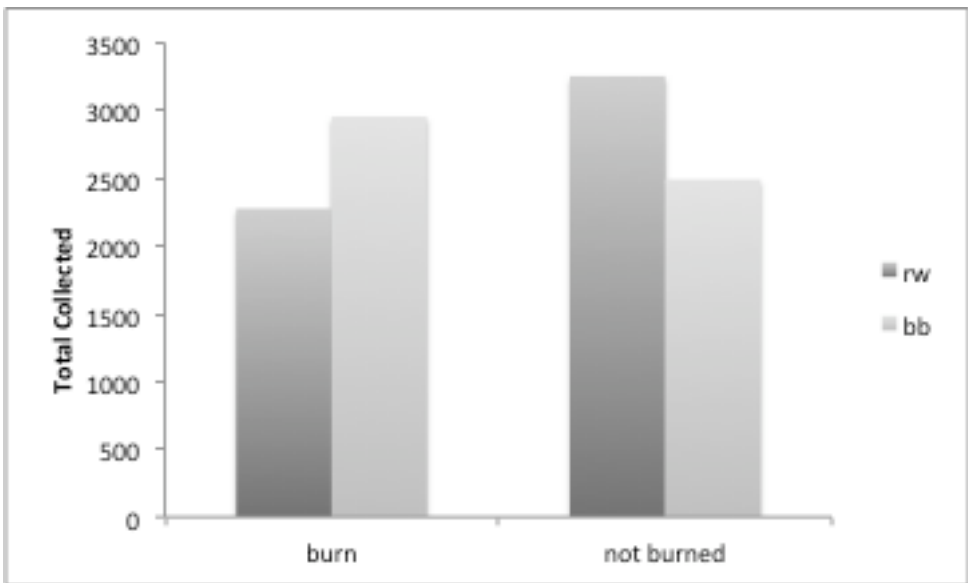


Figure 2.8. Abundance of spiders sampled from May 2010-Oct 2011 from pitfall traps and sweep samples. Main effect of site (ANOVA, $F=11.45$, $df=1, 59$, $P=0.0013$), burn treatment (ANOVA, $F=0.11$, $df=1, 59$, $P=0.7398$), and site by burn treatment (ANOVA, $F=28.84$, $df=1, 59$, $P<0.0001$).

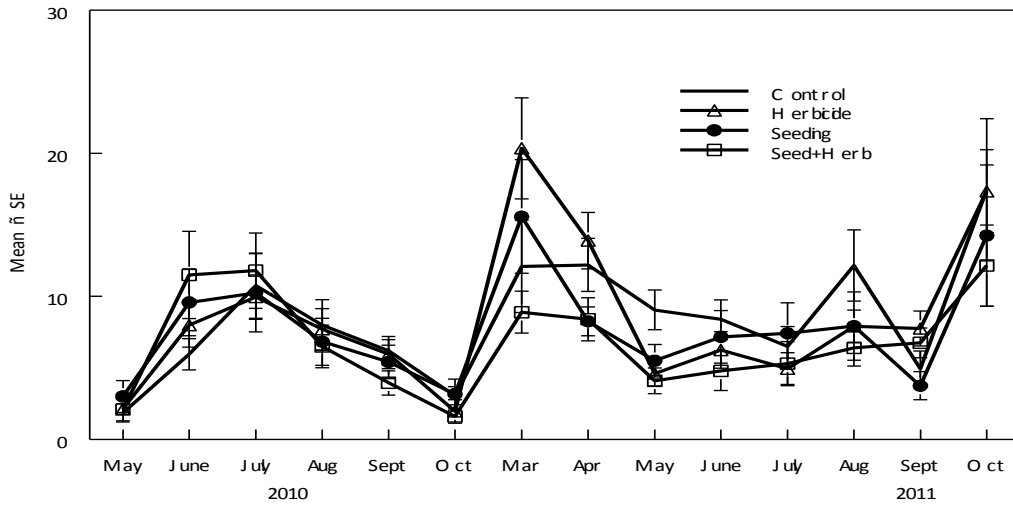


Figure 2.9. Predatory beetles collected in each treatment over 31 collection trips in 2010 and 2011. ANOVA revealed: year (ANOVA, $F=37.96$, $df=1,63$, $P<0.0001$), and treatment (ANOVA, $F=1.75$, $df=3,59$, $P=0.1670$). Significant interaction was: site by treatment (ANOVA, $F\text{ value}=4.25$, $df=3,59$, $P=0.0087$).

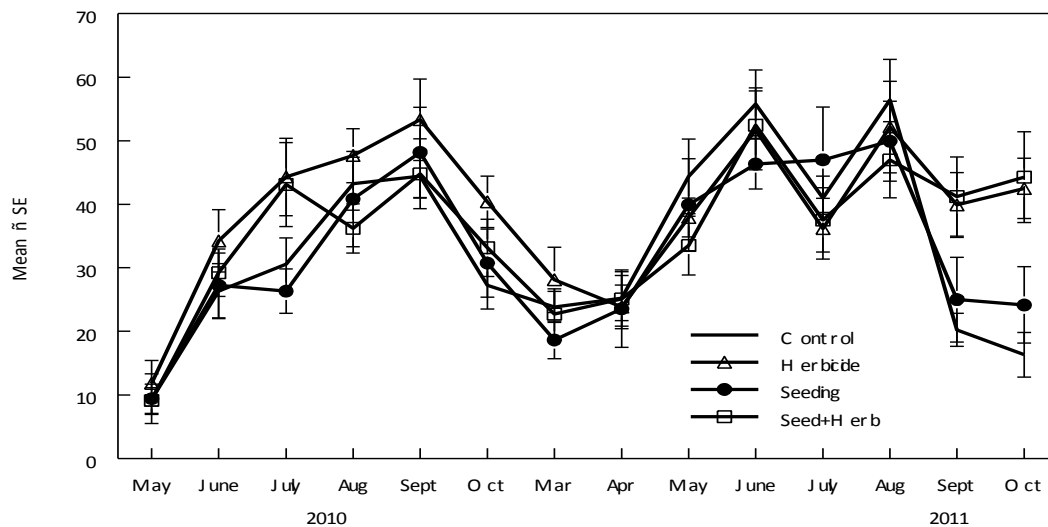


Figure 2.10. Ants collected in each treatment over 31 collection trips in 2010 and 2011. ANOVA revealed: year (ANOVA, $F=6.39$, $df=1,63$, $P=0.0140$), and treatment (ANOVA, $F=1.94$, $df=3,59$, $P=0.1334$). Significant interaction was: year by site (ANOVA, $F=24.52$, $df=1,63$, $P<0.0001$).

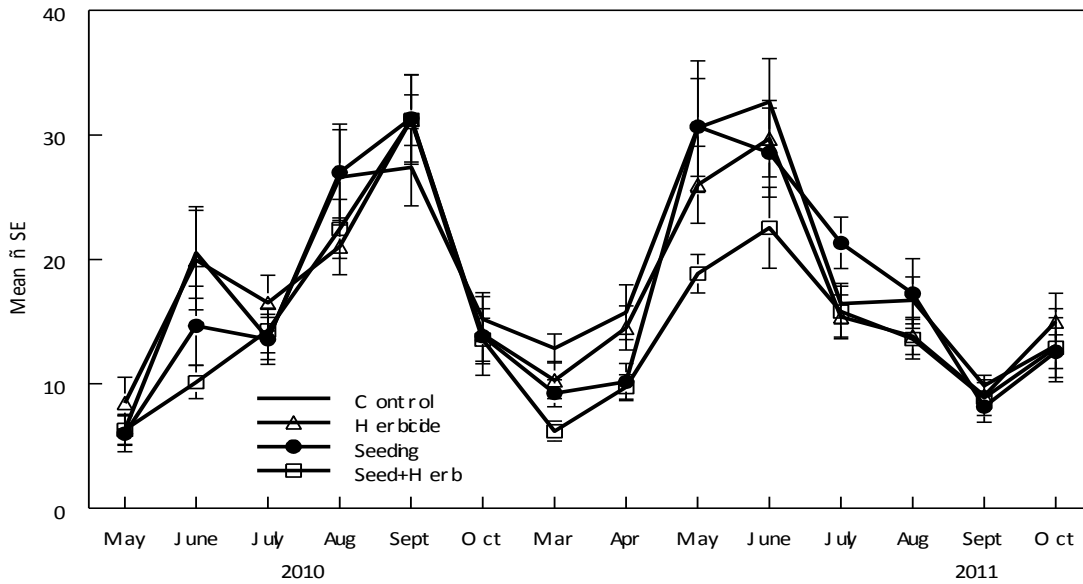


Figure 2.11. Spiders collected in each treatment over 31 collection trips in 2010 and 2011. ANOVA revealed: year (ANOVA, $F=1.21$, $df=1,63$, $P=0.2758$), and treatment (ANOVA; $F=4.11$; $df=1,63$; $P=0.0102$). Significant interaction was: year by site (ANOVA, $F=46.01$, $df=1,63$, $P=<0.0001$).

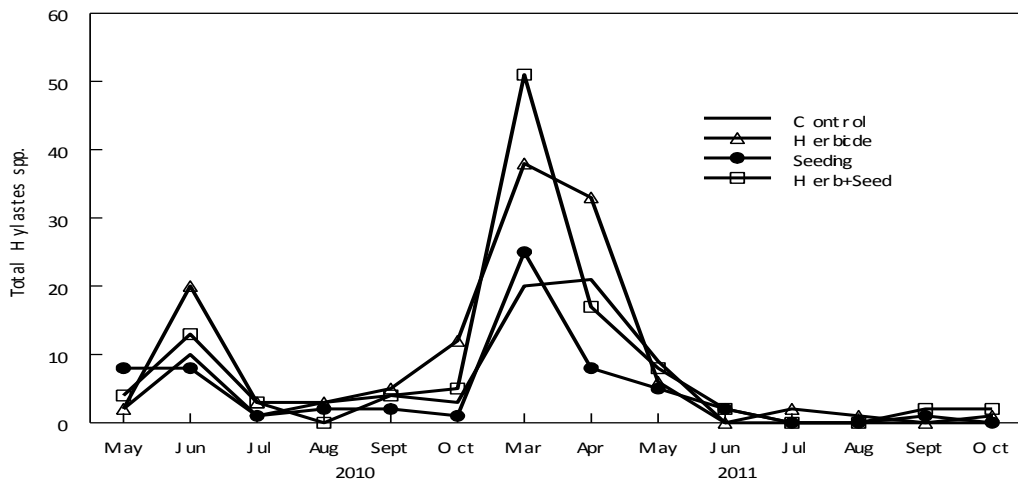


Figure 2.12. Total *Hylastes* spp. collected in each treatment over 31 collection trips in 2010 and 2011. Biweekly samples from baited pitfall traps were pooled into monthly totals. ANOVA revealed: year (ANOVA, $F=6.38$, $df=1,63$, $P=0.0141$), site (ANOVA, $F=0.81$, $df=1,59$, $P=0.3730$), burn treatment (ANOVA, $F=0.14$, $df=1,59$, $P=0.7072$), and treatment (ANOVA, $F=0.43$, $df=3,59$, $P=0.7317$). Significant interaction was seen in year by site (ANOVA, $F=4.18$, $df=1,63$, $P=0.0452$).

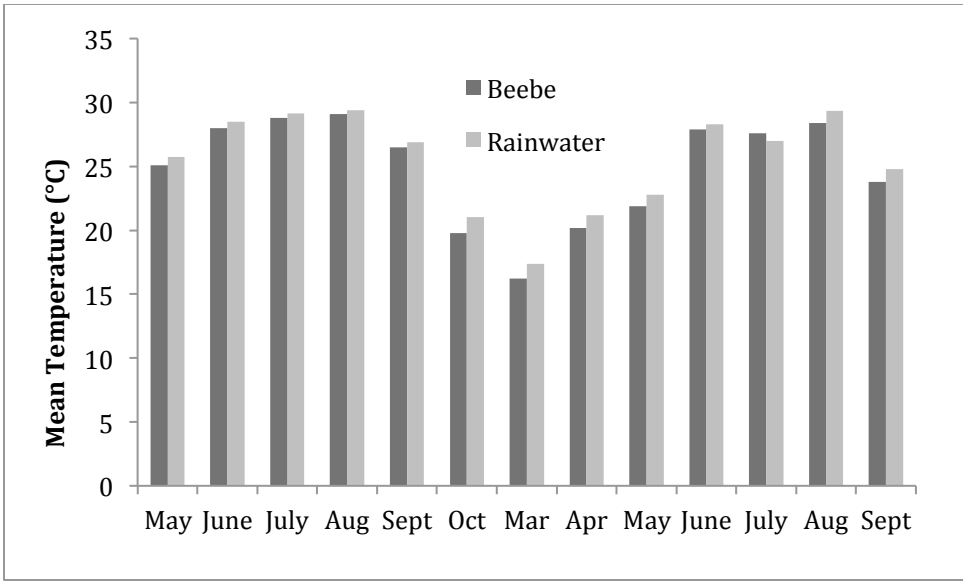


Figure 2.13. Mean monthly temperatures in 2010 and 2011 for Baldwin County (Beebe) and Mobile County (Rainwater). (AWIS 2008)

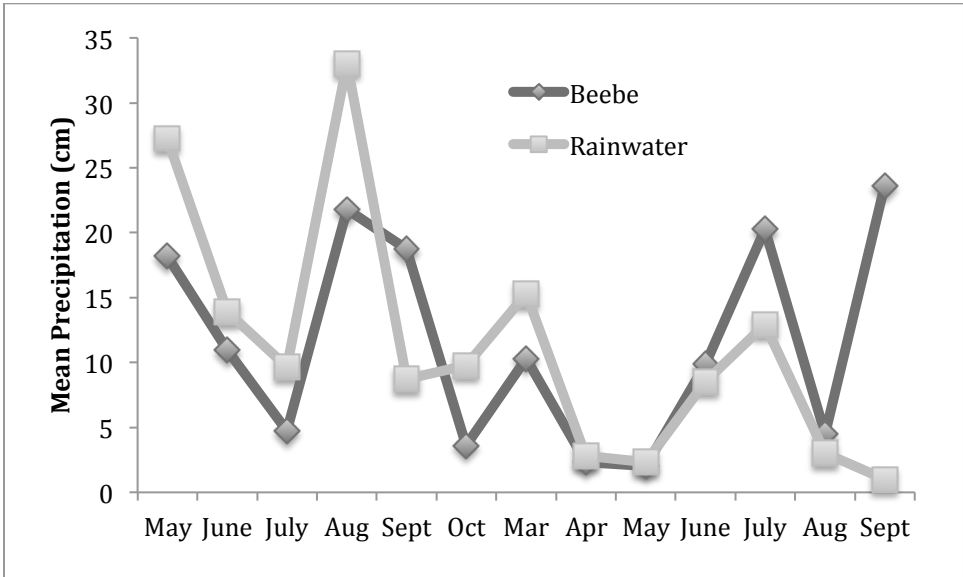


Figure 2.14. Mean monthly precipitation in 2010 and 2011 for Baldwin County (Beebe) and Mobile County (Rainwater). (AWIS 2008)

Chapter 3

Impact of cogongrass on diversity and abundance on arthropods and soil dwelling microfauna in longleaf pine

Abstract

Cogongrass (*Imperata cylindrica* Beav.) is an invasive C₄ grass currently invading the southeastern coastal plains of Alabama and its surrounding states. To assess the impact cogongrass has on arthropod and soil dwelling invertebrates, we used pitfall traps, sweep samples, soil purging and soil samples. Although cogongrass does impact arthropod diversity, it is necessary to evaluate each individual taxa to understand the impact it has on arthropod communities. Seasonally, Gryllidae, Curculionidae, Scarabaeidae and Araneae were all significantly impacted by cogongrass presence. Most herbivore groups were significantly more abundant in longleaf pine with cogongrass present. Dung beetles and their relatives arose as excellent bioindicators of cogongrass presence in longleaf pine. Earthworms yielded significantly greater numbers in cogongrass present plots by both abundance and weight. Nematodes reacted differently by functional group: plant parasitic nematodes were significantly more abundant in cogongrass present plots while free-living nematodes revealed the opposite.

Introduction

The introduction of aggressive, non-indigenous species poses one of the greatest threats to ecosystems worldwide (Capo-chichi et al. 2008, Mack et al. 2000). Cogongrass (*Imperata cylindrica*), an aggressive, rhizomatous grass indigenous to Southeast Asia, was introduced to South Alabama around 1910 (MacDonald 2004, Dozier et al. 1998). Cogongrass' ability to hinder nutrient uptake and retention, interfere with respiration, and create monotypic expanses makes it especially threatening to ecosystems (MacDonald 2004). It is ranked as one of the ten worst weeds worldwide (Holm et al. 1977), fundamentally because of its impacts on agriculture, forestry, and fire regimes (Lippincott 2000, Dozier et al. 1998). It is a threat to forest systems because even after burning, cogongrass remains dominant, prevents forest succession, and displaces native plants (Lippincott 2000). It possesses several strategies that promote its survival such as drought tolerance, fire adaptability, adaption to poor soils, a widespread rhizome network, and prolific seed production (MacDonald 2004, Dozier et al. 1998).

Longleaf pine savannas are particularly vulnerable to cogongrass invasion. These fire dependent savannas previously stretched from Virginia to Texas but due to anthropogenic activity are now isolated to a few areas (Brockway et al. 2005). They are home to a variety of rare and endangered plant and animal species sensitive to disturbance (Brewer 2008). Game animals such as white-tailed deer, turkey and northern bobwhite as well as rare, non-game animals including red-cockaded woodpecker, eastern indigo snake, gopher tortoise, and fox squirrel all take advantage of longleaf pines for a stable habitat. Longleaf ecosystems are among the most species-rich plant communities

outside the tropics due to the rich diversity of understory plants found there (Brockway et al. 2005).

Measuring biodiversity in changing ecosystems is critical for determining health conservation needs. Forests support the greatest global biodiversity in terrestrial ecosystems, therefore conserving forest biodiversity should be an important objective for forest management (Maleque et al. 2009). Entire inventories of forest biodiversity, even if possible, are very time-consuming and expensive (Maleque et al. 2009). As an alternative to complete inventories, bioindicators are used to monitor ecosystems and biodiversity because they are typically functionally important to the ecosystem and are determined by disturbance regimes (Maleque et al. 2009, Langor and Spence 2006). Arthropods are frequently used as bioindicators because they have short generation times, have high species richness, and are sensitive to environmental changes and habitat disturbance. They also play critical roles in an ecosystem as herbivores, parasites, predators, decomposers, pollinators and seed dispersers (Maleque et al. 2009, Maleque et al. 2006).

Gerber et al. (2007) proposed a link between replacement of native plant species by exotic species and the overall reduction of abundance and richness of arthropods. Invaded areas supported lower numbers of native plants as well as a lowered abundance and richness of invertebrates when compared to uninvaded areas (Gerber et al. 2007). In the grasslands of Indonesia, braconid (Hymenoptera: Braconidae) diversity declined in cogongrass dominated areas. As grasslands returned to *Acacia* forests, parasitoid abundance and species richness increased (Maeto et al. 2009).

Soil dwelling organisms provide a series of ecosystem services, including decomposition of organic matter, nutrient cycling, and bioturbation (Brussaard et al. 1997, Syers and Springett 1984). Earthworm diversity is determined by soil type, nutrient content, organic matter, and disturbance more than by plant diversity (Brussaard et al. 1997). Their body tissues and cast materials are enriched with nutrients and their ability to ingest organic material increases rate of nutrient cycling (Syers and Springett 1984). Earthworm abundance can be influenced by fertilizers, physical disruption to soils, and changes of soil nutrients (Kladivko 2001). In some forests, earthworm communities have high species richness and diversity and a relatively high biomass (Blanchart and Julka 1997). Although earthworms can serve as beneficial species, many earthworms are invasive in forests. They can have significant impacts on plant communities and soil processes in native ecosystems (Hendrix and Bohlen 2002).

Nematode species composition is effected by soil disturbance, erosion and water quality (Brussaard et al. 1997). Nematodes have a very limited range and exist in the top few centimeters of soil. Changes in temperature and moisture, as well food supply, pore disruption and moisture conditions after forest disturbance all influence nematode richness and diversity. After soil disturbance, nematodes must cope with many of these changes while the soil profile is reassembled (Ettema 1998). Forest nematodes are not frequently studied because of the volume of unknown species and amount of resources required (Bloemers et al. 1997). The objective of this study was to better understand the impact of cogongrass invasion in longleaf pine on arthropods and soil dwelling microfauna.

Materials and Methods

Site Description. Site one, Rainwater, is in Mobile County, and site two, Beebe, is in Baldwin County, each with 10 plots of well-established cogongrass in longleaf pine per site. On original Rainwater and Beebe sites, cogongrass was too prevalent for cogongrass absent plots to be established. Additional plots were established nearby on land with similar geographic traits. Both sites consisted of sandy loam soils, which is typical for this region (Table 3.1).

Experimental Design. Plots of cogongrass present and cogongrass absent sites were established in longleaf pine stands in south Alabama to understand the impacts of cogongrass invasion on arthropod communities of Southeastern pine forests. On each of the four sites, ten 10 x 10 m plots were established containing 5-8 trees each with a DBH ranging between of 11.4 and 54.6 cm, and a height ranging from 6.22 to 25.27 m.

Arthropod Sampling. Arthropods were sampled using two methods: pitfall traps for ground active arthropods, and sweep samples primarily targeting arthropods associated with vegetation. Five pitfall traps (15 cm dia funnel type traps, Multi-pher, Bio-Controle, Quebec City, Quebec), were installed in each plot, arranged in an X pattern 3 m apart along each diagonal with one in the center (Figure 2.2). Inside each trap, a 453 ml plastic cup (16K, Dart, Mason, MI) was filled with 50 ml of a 1:1 (v/v) mixture of water and propylene glycol (Peak, Old World Industries, Northbrook, IL) to serve as a preserving

solution. Every 2 wk, contents from all five pitfall traps were pooled into a labeled plastic container. Empty traps were then recharged with preserving solution. Pitfall traps were active from May 1 through Oct 28, 2010. Pitfall traps remained empty in the field overwinter. In 2011, traps were activated on Feb 15 and serviced biweekly through Oct 25. Soil displaced from pitfall trap installation was collected and brought back to the AU soil dynamics lab for analysis.

Sweep samples were taken biweekly, concurrent with servicing pitfall traps. One sample was made per plot, following each diagonal making approximately 30–40 sweeps per plot. Insects collected in the sweep net were put into a 3.8 liter plastic zipper bags and placed into a cooler for transport back to the laboratory.

Inside the laboratory, arthropods from pitfall traps contents were filtered to remove dirt and other debris. The remaining arthropods were stored in 50 ml plastic vials with 90% ethanol in cooler until they were sorted. Arthropods were sorted into taxonomic groups (insects and other arthropods) and stored in 22 ml vials with 90% ethanol. From these samples, specimens were sorted to family. Sweep samples were placed in the freezer for ≥ 1 d. Insects were then removed from the bags and placed into 15 ml vials by plot in 90% ethanol. Like the arthropods collected from pitfall traps, all specimens that could be identified to family were, and those that could not be were given to Dr. Charles Ray, Insect Diagnostic Lab, Auburn University, to be classified.

Earthworm Sampling. Earthworm samples were collected twice (spring and fall 2011) from two, 0.5 m² samples in each plot. Vegetation or leaf litter was manually cleared from each area and a plastic frame (40.6 cm wide, 30.5 cm tall) defining the area placed

and inserted slightly (about 2.5 cm) into the soil surface (Figure 3.1). An expellant solution of mustard seed powder (allyl isothiocyanate) in water (100 mg per liter) was prepared (Zaborski 2003), and 2 liters of that solution were applied to each frame in successive pourings (Lawrence and Bowers 2002). All earthworms that emerged within 10 minutes of pouring were collected into small plastic bottles with moist paper towels for transport to the lab. In the lab, the number and mass of worms from each plot were determined (Zaborski 2003, Lawrence and Bowers 2002).

Nematode Sampling. Soil samples of nematodes were collected in late spring 2011. Each plot was sampled in a systematic pattern using a cone soil probe with a 1.9 cm opening. Five uniform soil samples were taken, composited into one sample per plot and stored in separate one-quart plastic ziploc bags. In the AU Nematology Lab, individual soil samples were mixed thoroughly, and 100 cm³ of soil was extracted from each sample. Nematodes were extracted using gravity screening and centrifugal flotation methods. Numbers of nematodes present in each plot were identified to genus level and nematodes will be identified to trophic level (plant parasitic or free-living). All nematode samples were quantified under an inverted microscope (Nikon T-100).

Analyses. Counts of arthropods exceeding 900 within each family were analyzed by fitting each insect count using a Poisson distribution generalized linear mixed model (GLMM) with log-link function (PROC GLIMMIX, SAS Institute 2002). GLMM allows for a Poisson regression (insect counts) with the inclusion of random effects in the model. A Poisson GLMM with repeated measures was fitted to the count data. The random effects were sampling error and plot error. Compound symmetry (CS) was used for the

within-subject covariance structure. If significant, pairwise mean comparisons among main effects and interactions were made using the least-squares means (LSMEANS) method. Non-parametric analysis (Wilcoxon Rank Sum Test) was conducted to compare insect families between cogon and no cogon treatments (SAS 2005).

Earthworm weight was analyzed using a split-plot experimental design, where site (RW and BB) and treatment (cogon and no cogon) was sub-plot factor. Earthworm count data were analyzed by fitting each counts using Poisson generalized linear mixed model (GLMM) with log-link function. The random effects were site (RW and BB) and sampling error. Data analysis was conducted with SAS software using generalized linear mixed model procedure (PROC GLIMMIX, SAS Institute 2002). If significant, pairwise mean comparisons among main effects and interactions were made using the least-squares means (LSMEANS) method.

Diversity of nematode genera and orders in cogon and no cogon fields in different sites (RW and BB) were examined by the following metrics: Shannon Diversity, species richness, species number, species evenness and ratio of plant parasitic/free-living nematodes. All statistical tests for diversity were performed in R using (R Development Core Team 2011). Counts of nematodes were analyzed by fitting each nematode count using Poisson generalized linear mixed model (GLMM) with log-link function (PROC GLIMMIX, SAS Institute 2002). GLMM allows for a Poisson regression (nematode counts) with the inclusion of random effects in the model. A Poisson GLMM with repeated measures was fitted to the count data.

Results

Over 40,000 arthropods in over 100 families were recorded from eight months of sampling in longleaf pine. Coenagrionidae (Odonata), Nabidae (Hemiptera), Hippoboscidae, Tachinidae, Ulididae (all Diptera), Sphecidae (Hymenoptera) were only found in longleaf pine with cogongrass present. Gryllotalpidae (Orthoptera), Achilidae, Aradidae, Caliscelidae, Tingidae (all Hemiptera), Attelibidae, Bostrichidae, Buprestidae, Cicindellidae (all Coleoptera), Ceratopogonidae, Cloropidae, Sarcophagidae, Simuliidae (all Diptera), Scoliidae and Trichogrammatidae (both Hymenoptera) were only recorded in longleaf pine with cogongrass absent (Table 3.2). After identifying and counting all arthropods to the family level, it was found that only 8 groups revealed differences across the season (Table 3.3). Only Gryllidae (crickets), Scarabaeidae (scarabs), and Aranae (spiders) were significantly more abundant in longleaf pine without cogongrass while Curculionidae (weevils) were significantly more abundant in longleaf pine with cogongrass.

The Wilcoxon rank-sum test revealed 30 taxa to be significantly impacted by cogongrass presence (Table 3.2). First, parasitic hymenopterans (Ichneumonidae and Braconidae) and chalcids (Chalcoidea) were more abundant in longleaf pine with cogongrass present. Herbivores such as katydids (Tettigoniidae), grasshoppers (Acrididae), walking sticks (Phasmatodea), seed bugs (Alydidae), spittle bugs (Cercopidae), jumping plant lice (Psyllidae), leaf beetles (Chrysomelidae), gall midges (Cecidomyiidae), and caterpillars (Lepidoptera) were generally twice as abundant in longleaf pine with cogongrass present. A few other herbivores, leaf hoppers

(Cicadellidae), plant hoppers (Issidae), and tree hoppers (Membracidae) revealed the opposite. Three members of the Scarabaeoidea superfamily, scarabs (Scarabaeidae), earth-boring scarab beetles (Geotrupidae), and hide beetles (Trogidae), were all found to be at least seven times more abundant in longleaf pine with cogongrass absent than those with cogongrass present. Lastly, springtails (Collembola) were three times more abundant in longleaf pine with cogongrass present.

Earthworm weight (Figure 3.3) and abundance (Figure 3.4) were significantly greater in longleaf with cogongrass present. Similarly, for total nematodes, total plant parasitic and *Mesocriconema* spp, nematodes were significantly greater in longleaf pine with cogongrass present (Table 3.4). In contrast, free-living nematodes were significantly more abundant in longleaf pine without cogongrass. Diversity and evenness are fairly low in longleaf pine with and without cogongrass. More species were recorded in longleaf pine with cogongrass present than absent. Also, longleaf pine with cogongrass present had a greater ratio of plant parasitic nematodes than cogongrass absent where the ratio was equal.

Discussion

In this 8 month study, we compared abundance and diversity of arthropod and soil microfauna in longleaf pine. The only group of natural enemies to show a significant seasonal impact due to cogongrass presence was spiders (Table 3.3). Spiders are extremely sensitive to disturbance (Maleque et al. 2009), especially soil disturbance and litter type (Pearce and Venier 2006). Other families included Gryllidae, Scarabaeidae and

Curculionidae. A study conducted on wood crickets (*Nemobius* spp) suggested open, wooded edges were their preferred habitat (Brouwers and Newton 2009). Conservation of forests and reducing edge effect is critical for habitat preservation.

Those beetles in the Scaraboidea super family are important nutrient recyclers, soil aerators, seed dispersers and indicators in the forest ecosystem (Maleque et al. 2009, Vulinec 2000). They feed on dung, carrion, fungi and decaying leaves (Maleque et al. 2009). They are especially susceptible to ecosystem change with respect to soil characteristics. Degraded soils are not conducive to large scarab populations (Vulinec 2000) and beetle size and diversity increase in intact forests (Scheffler 2005). Scarab distribution between longleaf pine with cogongrass present compared to longleaf pine with cogongrass absent reveals the greatest difference in all the families surveyed (Table 3.2). With almost 3,000 scarabs collected in longleaf pine with cogongrass absent, evidence of community change due to cogongrass is overwhelming. Scarabs were collected abundantly throughout the season in longleaf pine with cogongrass absent. At least eight different morphotypes were observed consistently in longleaf pine with cogongrass absent, reducing of possibility of one species population exploding and skewing abundance results. Also, there were no carcasses of vertebrates detected in the vicinity of pitfall traps, which could have biased captures in adjacent plots.

Parasitic Hymenoptera were found significantly more abundant in longleaf with cogongrass present. These findings contradict research (Maeto et al. 2009) where cogongrass presence in forest systems reduced braconid richness and diversity. Braconid larvae are parasitoids on larvae of beetles, flies, butterflies and moths, as well as aphids

and Heteroptera (Matthews 1974). Larvae of click beetles, weevils, leaf beetles, flies, Lepidopterans, and a few Heteropteran families were more abundant in longleaf pine with cogongrass present (Table 3.3)

Generalist herbivores may be in greater abundance in longleaf pine with cogongrass because of the greater amount of biomass available compared to longleaf pine with cogongrass absent. Chewing marks, like those left from grasshoppers or katydids, were observed on cogongrass leaf blades. Spittle bugs (Cercopidae), a family of grass-feeding Hemipterans (Borror and White 1970), were also significantly more abundant in longleaf pine with cogongrass. Spittle bug immatures were observed on leaf bases of cogongrass on multiple occasions. Jumping plant lice (Psocoptera) were twenty times more abundant in longleaf pine with cogongrass present. Since they are monophagous or oligophagous plant feeders, it could be suggested that they are feeding on cogongrass, but further research would be needed to confirm since other plants were present (Enloe and Loewenstein unpublished data). Weevils were the only family significantly greater in longleaf pine with cogongrass present over the collecting season and total (Table 3.2 and 3.3). Most of the weevils collected were billbugs (*Sphenophorous* spp.). Billbugs are obligate grass feeders with adults feeding and larvae developing as a stem borer. It is possible that billbugs are using the dense root mass of cogongrass as a nest although this behavior was not observed.

Earthworm abundance and weight were more significant in longleaf pine with cogongrass present. Because earthworms are active decomposers of the litter layer, a link between cogongrass thatch and earthworm presence is possible. Cogongrass' ability to

alter the nutrient availability of the soil (Jose et al. 2002) could explain the vast difference between the cogongrass presence and absence. If the presence of cogongrass seriously alters the earthworm community, the soil and soil dwelling arthropod community could experience a cascading effect. Increased abundance of earthworms could lead to a spike in small mammals or ground-active birds that prey on earthworms. In fact, multiple small mice were accidental victims to pitfall traps, but only in longleaf pine with cogongrass present.

Nematodes are a major component of all soil food webs and soil disturbance affects species composition (Brussaard et al. 1997). Samples revealed seven different species of free-living nematodes and 3 orders of free-living nematodes (Table 3.4). Plant parasitic nematodes were significantly more abundant in longleaf pine with cogongrass present and only *Mesocriconema* spp. were significantly impacted by cogongrass presence. Free-living nematodes had an inverse response, having greater numbers in longleaf pine with cogongrass absent. This leads us to conclude free-living nematodes are more sensitive to cogongrass invasion than plant parasitic one. The ratio indicates plant parasitic nematodes composed 70% of collected species in longleaf pine with cogongrass present. In longleaf pine without cogongrass present, the ratio was approximately 1:1, leading us to believe the absence of cogongrass provides a more suitable habitat for both types of nematodes. Overall, diversity and evenness were fairly low in longleaf pine with and without cogongrass even though they are significantly different. Without a pristine habitat for comparison, it is hard to compare

Cogongrass presence significantly impacted arthropod abundance in approximately 30 taxa. Scarabs, the most diverse family collected, present a hard to ignore indication of disturbance due to cogongrass. Significant differences in these arthropod groups highlight the necessity for more research. In cogongrass systems, especially those within longleaf pine, scarabs, geotrupid beetles, trogid beetles, carabids, and crickets could all serve as indicators of disturbance by invasion. These taxa should be of interest in other systems where cogongrass invades. Herbivores such as grasshoppers, katydids, seed bugs, spittle bugs, psyllids, and leaf beetles present the potential for a biological control that might have been overlooked in previous studies. Earthworms seem to take advantage of the altered soil regime found in longleaf pine with cogongrass present. Nematodes, however, are more sensitive to cogongrass based on functional group, with plant parasitic nematodes significantly more abundant in longleaf pine with cogongrass present. Cogongrass does impact arthropod diversity and for future studies it is necessary to investigate each individual taxa to understand the overall impact cogongrass has on an entire arthropod community. These taxa present opportunities for further investigation. Although families and subfamilies share similar life histories, perhaps species within these larger taxonomic groups maybe found as specific indicators of disturbance for cogongrass.

Table 3.1. Average soil characteristics for cogon(C) and no cogon (NC) plots for objective 2.^a

| Site | pH | Soil Type | CEC |
|-------------|------|-----------|--|
| RainwaterC | 4.97 | Sandy | < 4.6 cmol _c kg ⁻² |
| RainwaterNC | 4.83 | Sandy | < 4.6 cmol _c kg ⁻² |
| BeebeC | 5.39 | Sandy | < 4.6 cmol _c kg ⁻² |
| BeebeNC | 5.41 | Sandy | < 4.6 cmol _c kg ⁻² |

^aSoil tests conducted by AU Soil Testing Laboratory.

Table 3.2. Comparison of arthropods collected over entire season in longleaf with and without cogongrass present by Wilcoxon In cogongrass present plots, 18,968 arthropods were collected and counted. In cogongrass absent plots, 20,182 arthropods were collected and counted. Both were collected over an 8 month period in 2011.

| Arthropod Group | Total | | P |
|-------------------|--------------|----------|---------|
| | Cogon | No cogon | |
| | Odonata | | |
| Coenagrionidae | 6 | 0 | 0.020 |
| | Orthopteroid | | |
| Rhaphidiophoridae | 509 | 560 | 0.457 |
| Gryllidae | 338 | 1022 | <0.0001 |
| Gryllotalpidae | 0 | 2 | 0.342 |
| Tetrigidae | 4 | 7 | 0.624 |
| Tettigoniidae | 212 | 41 | <0.0001 |
| Acrididae | 123 | 32 | <0.0001 |
| Mantidae | 1 | 4 | 0.164 |
| Phasmatidae | 15 | 6 | 0.026 |
| Blattodea | 111 | 86 | 0.108 |
| | Hemiptera | | |
| Achillidae | 0 | 1 | 0.342 |
| Acanaloniidae | 3 | 6 | 0.270 |
| Aphididae | 39 | 57 | 0.835 |
| Alydidae | 117 | 69 | 0.055 |
| Aradidae | 0 | 6 | 0.040 |
| Berytidae | 6 | 12 | 0.325 |
| Caliscelidae | 0 | 1 | 0.342 |
| Cercopidae | 26 | 16 | 0.012 |
| Cicadellidae | 115 | 207 | 0.044 |
| Cixiidae | 15 | 22 | 0.244 |
| Coreidae | 5 | 3 | 0.706 |
| Cydnidae | 30 | 29 | 0.505 |
| Delphacidae | 4 | 9 | 0.426 |
| Dictyopharidae | 7 | 12 | 0.945 |
| Flattidae | 1 | 3 | 0.554 |
| Geocoridae | 1 | 0 | 0.342 |
| Issidae | 2 | 19 | 0.009 |
| Lygaeidae | 5 | 7 | 0.508 |
| Membracidae | 9 | 35 | 0.005 |
| Miridae | 2 | 8 | 0.211 |
| Nabidae | 3 | 0 | 0.163 |
| Pentatomidae | 36 | 23 | 0.589 |
| Psyllidae | 435 | 21 | <0.0001 |
| Reduviidae | 53 | 32 | 0.036 |
| Tingidae | 0 | 1 | 0.342 |

| Coleoptera | | | |
|-------------------------------|------|------|---------|
| Attelibidae | 0 | 1 | 0.342 |
| Bostrichidae | 3 | 2 | 0.979 |
| Buprestidae | 0 | 1 | 0.342 |
| Cantheridae | 7 | 5 | 0.307 |
| Carabidae | 768 | 1416 | 0.017 |
| Cerambycidae | 2 | 6 | 0.573 |
| Chrysomelidae | 238 | 68 | <0.0001 |
| Cicindelidae | 0 | 1 | 0.342 |
| Coccinellidae | 2 | 3 | 0.654 |
| Curculionidae | 1425 | 141 | <0.0001 |
| Elateridae | 835 | 338 | 0.001 |
| Erotylidae | 2 | 0 | 0.162 |
| Geotrupidae | 18 | 754 | <0.0001 |
| Histeridae | 4 | 6 | 0.758 |
| Lampyridae | 3 | 0 | 0.080 |
| Mordellidae | 8 | 19 | 0.091 |
| Nitidulidae | 460 | 495 | 0.978 |
| Scarabaeidae | 368 | 2898 | <0.0001 |
| Staphylindidae | 982 | 848 | 0.409 |
| Tenebrionidae | 723 | 332 | 0.144 |
| Trogidae | 5 | 179 | <0.0001 |
| Neuroptera | | | |
| Predatory Larvae ^a | 15 | 12 | 0.569 |
| Diptera | | | |
| Asilidae | 26 | 8 | 0.337 |
| Bibionidae | 1 | 1 | 1.000 |
| Calliphoridae | 3 | 5 | 0.652 |
| Cecidomyiidae | 49 | 9 | 0.001 |
| Ceratopogonidae | 0 | 1 | 0.342 |
| Chironomidae | 223 | 139 | 0.015 |
| Cloropidae | 0 | 2 | 0.162 |
| Culicidae | 10 | 10 | 0.716 |
| Dolichopodidae | 58 | 16 | <0.0001 |
| Drosophilidae | 30 | 3 | 0.004 |
| Empididae | 4 | 2 | 0.615 |
| Hippoboscidae | 1 | 0 | 0.342 |
| Muscidae | 228 | 145 | 0.069 |
| Phoridae | 5 | 13 | 0.130 |
| Pipunculidae | 1 | 1 | 1.000 |
| Sarcophagidae | 0 | 1 | 0.342 |
| Sciaridae | 451 | 344 | 0.155 |
| Simuliidae | 0 | 2 | 0.162 |
| Tabanidae | 1 | 3 | 0.310 |
| Tachinidae | 1 | 0 | 0.342 |

| | | | |
|-----------------------|------|------|---------|
| Tephritidae | 41 | 26 | 0.131 |
| Tipulidae | 10 | 8 | 0.904 |
| Ulididae | 1 | 0 | 0.342 |
| Hymenoptera | | | |
| Apidae | 1 | 7 | 0.040 |
| Chalcoidea | 72 | 22 | <0.0001 |
| Evanidae | 3 | 1 | 0.554 |
| Formicidae | 5525 | 5680 | 0.636 |
| Halictidae | 4 | 12 | 0.219 |
| Mutilidae | 84 | 55 | 0.924 |
| Parasitic Hymenoptera | 67 | 44 | 0.052 |
| Pompilidae | 7 | 5 | 0.508 |
| Scoliidae | 0 | 5 | 0.342 |
| Sphecidae | 3 | 0 | 0.163 |
| Tenthredinidae | 16 | 1 | 0.554 |
| Trichogrammatidae | 0 | 1 | 0.342 |
| Vespidae | 3 | 8 | 0.141 |
| Other Orders | | | |
| Acari | 10 | 2 | 0.197 |
| Aranae | 2905 | 3429 | 0.110 |
| Collembola | 913 | 238 | <0.0001 |
| isoptera | 40 | 16 | 0.194 |
| Lepidoptera | 60 | 31 | 0.007 |
| Thysanoptera | 10 | 2 | 0.030 |

^aInsects in the families Ascalaphidae and Myrmeliontidae.

^bInsects in the families Ichneumonidae and Braconidae.

| Table 3.3. Comparison of populations of selected arthropods using all observations from each sample collected in pitfalls and sweep samples in longleaf pine with cogongrass present and cogongrass absent, Bay Minette and Grand Bay, AL, 2011, using repeated measures analyses | | | | |
|---|-----------------|-------|---------|---------|
| Effect | Arthropod group | F | df | P |
| Treatment | Gryllidae | 23.65 | 1, 38 | <0.0001 |
| | Carabidae | 1.72 | 1, 38 | 0.1982 |
| | Curculionidae | 10.48 | 1, 38 | 0.0025 |
| | Nitidulidae | 0.09 | 1, 38 | 0.763 |
| | Scarabaeidae | 18.45 | 1, 38 | <0.0001 |
| | Staphylinidae | 0.88 | 1, 38 | 0.3544 |
| | Formicidae | 1.48 | 1, 38 | 0.2318 |
| | Araneae | 5.14 | 1, 38 | 0.0292 |
| Time | Gryllidae | 3.76 | 17, 646 | <0.0001 |
| | Carabidae | 8.74 | 17, 646 | <.00001 |
| | Curculionidae | 2.65 | 17, 646 | 0.0003 |
| | Nitidulidae | 12.12 | 17, 646 | <0.0001 |
| | Scarabaeidae | 2.54 | 17, 646 | 0.0006 |
| | Staphylinidae | 5.87 | 17, 646 | <0.0001 |
| | Formicidae | 13.73 | 17, 646 | <0.0001 |
| | Araneae | 18.99 | 17, 646 | <0.0001 |
| Interaction | Gryllidae | 1.75 | 17, 646 | 0.031 |
| | Carabidae | 4.01 | 17, 646 | <0.0001 |
| | Curculionidae | 1.53 | 17, 646 | 0.0768 |
| | Nitidulidae | 2.52 | 17, 646 | 0.0007 |
| | Scarabaeidae | 1.68 | 17, 646 | 0.0413 |
| | Staphylinidae | 6.62 | 17, 646 | <0.0001 |
| | Formicidae | 3.74 | 17, 646 | <0.0001 |
| | Araneae | 2.65 | 17, 646 | 0.0003 |

Table 3.4. Mean abundance of different nematode taxa
 Total nematodes, plant parasitic (PP), and free-living (FL) are represented.
 If $P < 0.05$, difference is noted between cogongrass present (C) and
 cogongrass absent (N). Repeated measures test used.

| Taxon | Cogon Present | Cogon Absent | (P < 0.05) Difference |
|------------------------|---------------|--------------|-----------------------|
| Total | 212.4 | 196.3 | C > N |
| Total PP | 159.7 | 107.5 | C > N |
| <i>Helicotylenchus</i> | 5.8 | 28.3 | . |
| <i>Tylenchulus</i> | 79.8 | 54.7 | . |
| <i>Paratylenchulus</i> | 12.2 | 1.3 | . |
| <i>Meloidogyne</i> | 3.9 | 3.9 | . |
| <i>Mesocriconema</i> | 46.4 | 3.9 | C > N |
| <i>Hoplolaimus</i> | 11.6 | 0.0 | . |
| <i>Tylosorus</i> | 0.0 | 15.5 | . |
| Total FL | 52.8 | 88.8 | C < N |
| Rhabditida | 39.3 | 77.3 | . |
| Desmodorida | 13.5 | 3.9 | . |
| Enoplida | 0.0 | 7.7 | . |
| Shannon Diversity | 0.8 | 0.5 | . |
| Species Richness | 1.5 | 1.1 | . |
| Species Number | 2.5 | 1.9 | . |
| Species Evenness | 0.6 | 0.5 | . |
| Ratio: PP/(PP+FL) | 0.7 | 0.5 | . |



Figure 3.1. Plastic frame with mustard solution applied for earthworm extraction.



Figure 3.2. Pitfall traps were arranged in an X pattern 3 meters apart on each 10 x 10 m plot

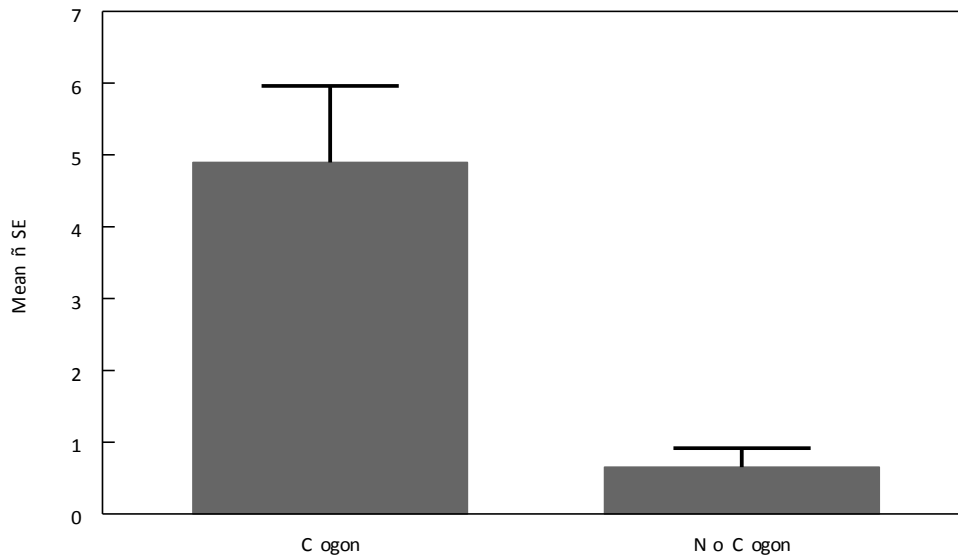


Figure 3.3 Earthworm abundance in 2011 on cogongrass present and cogongrass absent plots by abundance. Year was not significant, therefore samples were combined. ANOVA: $F= 8.00$, $df=1, 37$, $P=0.0075$.

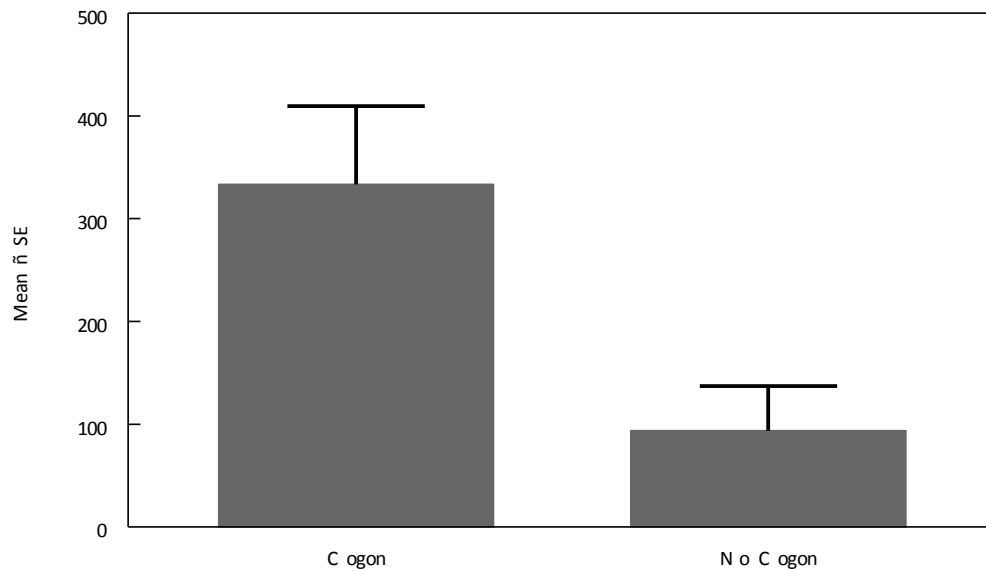


Figure 3.4 Earthworms collected in 2011 on cogongrass present and cogongrass absent plots by weight (mg). Year was not significant, therefore samples were combined. ANOVA $F=7.57$, $df=1, 37$, $P=0.0091$.

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