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Monitoring Abundance of *Ips* Bark Beetles and Determining Related Tree Mortality in  
Arkansas and Texas

Monitoring Abundance of *Ips* Bark Beetles and Determining Related Tree Mortality  
in Arkansas and Texas

A thesis submitted in partial fulfillment  
of the requirements for the degree of  
Master of Science in Entomology

by

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Virginia Polytechnic Institute and State University  
Bachelor of Science in Forest Resource Management, 2011

May 2015  
University of Arkansas

This thesis is approved for recommendation to the Graduate Council

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## ABSTRACT

The abundance of the southern pine engraver beetles, *Ips avulsus* (Eichhoff), *I. grandicollis* (Eichhoff), and *I. calligraphus* (Germar), was monitored with pheromone-baited traps in 2012 and 2013 in Arkansas and eastern Texas. Additionally, vehicle-based and comprehensive ground-based surveys were used to estimate and confirm the amount of dead and fading trees in each trapping site. Pine engraver beetles have historically been recognized as secondary invaders of healthy trees, but observations of pine mortality in Texas and Arkansas appeared to correspond with high population densities of *Ips* spp. Forest sites exhibiting subjectively high or low incidence of *Ips*-associated mortality in Arkansas and Texas were monitored using short trapping periods in the spring, summer, and fall of 2012 and the spring of 2013. Results of trapping indicated that *Ips* abundance steadily declined from the start of the study, and *I. calligraphus* abundance was considerably less than the other two species. Over the course of this study, a total of 127,823 *I. avulsus*, 217,636 *I. grandicollis*, and 26,290 *I. calligraphus* were trapped. The most abundant predators trapped were: 7555 Cleridae (*Thanasimus dubius* F. and *Enoclerus* spp.), 9036 *Temnochila virescens* (F.), 7162 Histeridae (*Platysoma* spp. and *Plegaderus* spp.), and 42,313 *Lasconotus* spp. For the sites used in this study, vehicle-based surveys did provide a good predictive model of actual stand mortality. The existence of a correlation between *Ips* abundance and surveys of tree mortality was investigated. A strong relationship was found between the abundance of *Ips* and the total number of standing dead and fading trees during the spring and summer of 2012; however, this relationship dissolved as *Ips* abundance declined over the course of the study. The number of fading trees alone did not provide a good indication of high *Ips* abundance.

## **DEDICATION**

A special thanks goes to my loving parents, Stefan and Angela Barton, who embraced my life-long passion to learn about entomology and forestry.

I want to express my sincere gratitude for my advisor, Fred Stephen. I thank him for his wisdom and assistance, but equally important, I also thank him for his patience. It was a privilege to work with him. Much like my family, I hope to make him proud with my future accomplishments in the field of forest entomology.

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## INTRODUCTION

The pine engraver beetles, *Ips* spp. (Coleoptera: Curculionidae: Scolytinae), are important pine-infesting insects found throughout North America. Historically, members of the genus *Ips* in North America have been considered secondary pests, and their damage rarely causes economic concern (Wilkinson and Foltz 1982). Pine engravers respond to trees that are stressed by a variety of predisposing agents, and drought is often considered a condition that precedes an outbreak (Clarke *et al.* 2000, Bryant *et al.* 2006, Pase III 2011). Potentially incited by dry conditions in 2011 and 2012, forests in Arkansas and eastern Texas had a recognizable increase of *Ips*-attacked loblolly, *Pinus taeda* (L.), and shortleaf, *P. echinata* (Mill.), pines. The three species of *Ips* responsible for these attacks were *Ips avulsus* (Eichhoff), *I. grandicollis* (Eichhoff), and *I. calligraphus* (Germar), collectively known as the southern pine engravers. This study focused on the monitoring of *Ips* abundance during and after the outbreak caused by recent regional drought.

The first chapter of this master's thesis is a review of the biology, behavior, and impact of the southern pine engravers. Subsequent chapters discuss research that focused on two topics: 1) monitoring *Ips* abundance with pheromone traps and 2) correlating *Ips* abundance with two independent survey methods of tree mortality.

Traps baited with *Ips*-specific pheromones were set up in two regions in Arkansas and one in East Texas and trapping was conducted in spring, summer, and fall of 2012 and in spring of 2013. The relative abundance of each *Ips* species was compared spatially and temporally. Additionally, I compared relative abundance of select predators and other associates attracted to the pheromone traps.

Two surveys of tree mortality were used to quantify damage resulting from an *Ips* outbreak. Both vehicle-based road surveys and comprehensive ground-based tallies were used to document the number of fading and dead trees. *Ips* abundance and the tree survey data was compared to learn if any correlations exist.

The fourth and final chapter is a case study of the eastern pine weevil, *Pissodes nemorensis* (Germar) (Coleoptera: Curculionidae), an often overlooked associate of southern pine engravers. Similar to pine engravers, it is a native beetle that is rarely considered a pest, and few scientific articles consider this insect as a pest in southern pine forests. However, land managers reported tree mortality caused by this beetle in Arkansas and Georgia pine plantations between 2009 and 2013. Information on the biology of *P. nemorensis*, its potential as a mortality factor in intensively managed pine forests, and efforts to monitor its abundance are discussed.

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## CHAPTER 1 – LITERATURE REVIEW

### INTRODUCTION

Recent observations of pine mortality related to bark beetles in the genus *Ips* (De Geer) (Coleoptera: Curculionidae: Scolytinae) are of concern to land managers in the southeastern United States. Stand-wide and regional disturbances that reduce tree vigor permit *Ips* population growth (Bryant 1983, Clarke *et al.* 2000). These pine engravers are generally accepted as ‘secondary invaders’ of dying trees, logging slash, and trees colonized by *Dendroctonus* spp. (Coleoptera: Curculionidae: Scolytinae) (Wilkinson and Foltz 1982). However, labeling them as secondary invaders diminishes the fact that *Ips* pine engravers are capable of causing widespread tree mortality when their populations are high (Wilkinson and Foltz 1982, Connor and Wilkinson 1983, Clarke *et al.* 2000). Historically, southern pine beetle (*Dendroctonus frontalis* Zimmerman) is considered the most destructive pest of southeastern pine forests, but some suggest that southern pine engraver beetles are responsible for more unreported pine mortality (Beal and Massey 1945, Thatcher 1960, Clarke *et al.* 2000). Taking into account the recent absence of *D. frontalis* west of the Mississippi River, pine engraver beetles are the most ubiquitous phloem herbivores in these southern pines.

The primary goal of this chapter is to provide information on the biology of southern pine engravers, to present factors related to their population dynamics, and to describe characteristics of their infestations in the southeastern United States.

## **BIOLOGY OF *IPS* PINE ENGRAVERS**

### **Species and host range**

*Ips* pine engravers are prevalent throughout North America wherever pine trees are found. In his description of the genus *Ips* in North America, Hopping (1963) recognized 32 species in 10 morphological groups. Three native species of *Ips* bark beetles colonize pines in the southeastern United States. These include the four-spined or small southern pine engraver (*Ips avulsus* Eichhoff), the five-spined or southern pine engraver (*I. grandicollis* Eichhoff), and the six-spined or coarse writing engraver (*I. calligraphus calligraphus* Germar). The ranges of all three species extend from Texas and the Gulf States up to southern Pennsylvania. *Ips grandicollis* and *I. calligraphus* extend even further north into Canada, coinciding with the range of pitch pine (*Pinus rigida* Mill.) (Drooz 1985). Southern pine engravers reportedly attack all *Pinus* species native to the eastern United States as well as many introduced pines (Eickwort et al. 2006).

### **Identification**

Pine engraver beetles are most easily recognized by their distinctive pairs of spines around the perimeter of the concave declivity of the elytra (Drooz 1985). The posterior end features a shelf-like structure distinct from the margin of the elytra (Craighead 1950). All species vary in color ranging from a tan callow adult or a reddish-brown to black fully sclerotized adult. The largest of *Ips* pine engravers, *I. calligraphus*, is 3.5 to 6.5 mm long, and it has six spines on each side of the declivity (Drooz 1985). *Ips grandicollis* is 2.8 to 4.7 mm long and has five pairs of spines. *Ips avulsus* is the smallest of *Ips* pine engravers with a body length of 2.3 to 2.8 mm, and it has four pairs of spines. The bases of 2nd and 3rd spines on *I. avulsus* are typically

adjoining. The thorax of *I. avulsus* is usually a darker color than the elytra (Connor and Wilkinson 1983).

### **Host partitioning**

*Ips* pine engravers are regarded as members of the “southern pine bark beetle guild” (Coulson et al. 1986, Smith et al. 1993, Nebeker 2011). The southern pine bark beetle guild includes the southern pine beetle (*D. frontalis*), the black turpentine beetle (*Dendroctonus terebrans* Olivier), and *Ips* pine engravers (Coulson et al. 1986). Members of this guild, with the exception of *D. terebrans*, are capable of colonizing and killing pine trees by mass attack (Schowalter 2012). *Dendroctonus frontalis*, the most aggressive species, will initiate colonization and may later be followed by other members of the guild. A tree may be made more vulnerable to attack by one or more of a spectrum of damaging agents (Smith et al. 1993). Most commonly, several or all species of the southern pine bark beetle guild compete for and utilize the phloem resource together (Thatcher 1960, Paine *et al.* 1981, Hain and McClelland 1979, Smith et al. 1993).

In the case where multiple species of the southern pine bark beetle guild colonize the same tree, they will effectively partition sections of the tree in a predictable manner (Paine et al. 1981, Wagner et al. 1985, Stephen 2011). Southern pine beetle colonizes the mid to lower sections of the bole, with initial attacks and highest densities occurring at about 3.5 m (Coster et al. 1977, Coulson et al. 1979). The crown of an attacked tree is dominated by *I. avulsus* (Paine et al. 1981, Coulson et al. 1986). Mason (1970) stated that *I. avulsus* is the least aggressive of *Ips* pine engravers; however, *Ips avulsus* is capable of top-killing a tree without the other members of the guild. The niche breadth of *I. avulsus* is the largest of *Ips* pine engravers (Paine *et al.* 1981). Initial *I. avulsus* attacks can occur in limbs or uppermost section of the crown (Berisford

and Franklin 1971). *Ips grandicollis* can also colonize large limbs or the bole within the live crown; however, they are more likely to be found in the middle to upper bole of the tree below the crown. *Ips calligraphus* will colonize the thicker phloem found at the basal portion of the tree. Black turpentine beetle will always be found in the lowest part of the tree that has the thickest bark and phloem, with initial attacks in the basal 18 inches (Clark 1970). Most overlap of niche breadth occurs in the mid-bole regions, where interspecific and intraspecific competition plays a large role in developmental success (Paine et al. 1981).

### **Pheromones and gender roles**

Pine engravers, like many other bark beetles, utilize pheromones for intraspecific aggregation to the host tree. Likewise, other members of the southern pine bark beetle guild and various associates are cross-attracted to these aggregation pheromones (Birch *et al.* 1980, Byers 1989, Stephen 2011, Allison et al. 2012a). All three species of *Ips* produce *de novo* compounds such as *cis*-verbenol, ipsenol, ipsdienol, or lanierone (Lanier and Wood 1975, Miller et al. 2005, Birgersson et al. 2012). *Ips* beetles use species-specific pheromone blends to attract both males and females to host trees (Byers 1989, Allison et al. 2012b). Ipsdienol and lanierone are produced by *I. avulsus* and are preferentially attractive to *I. avulsus* (Birgersson et al. 2012). Some interesting cases of interspecific inhibition occur when other combinations are used. For example, *I. grandicollis* is attracted to the synergistic combination of *cis*-verbenol and ipsenol, but this combination inhibits *I. calligraphus* attraction (Birgersson et al. 2012). Alternatively, *I. calligraphus* is attracted to *cis*-verbenol coupled with ipsdienol, but this is inhibitory to *I. grandicollis*. For trapping scenarios, a lure combination of ipsenol, ipsdienol, and *cis*-verbenol is effective for catching all three species (Allison et al. 2012b). However, this combination will capture fewer than if separate traps are designed to target individual species.

The attacking adults of many Scolytinae species have a sex ratio that favors females (Lanier and Oliver 1966). This is especially true for *Ips grandicollis* and *I. calligraphus*, where more females are captured in pheromone traps than males (Renwick and Vité 1971, Dixon and Payne 1979a, Cook *et al.* 1983). However, the use of emergence traps revealed that bark beetle brood sex ratios begin with uniform gender ratio (Cook *et al.* 1983). Emerging brood populations of *I. avulsus* and *I. calligraphus* were not statistically different from the expected 1:1 gender ratio. The gender ratio of attacking *I. calligraphus* and *I. grandicollis* adults appears to vary with different lure types (Renwick and Vité 1971); however, it will typically deviate from a 1:3 (♂:♀) ratio (Cook *et al.* 1983). Unlike *I. grandicollis* and *I. calligraphus*, the sex ratio of attacking *I. avulsus* is nearer to 1:1 (Berisford and Franklin 1971, Cook *et al.* 1983). More research is necessary to determine causes of this differential gender in attacking adults.

Once aggregated to the host and the male entry hole, the female uses a stridulating organ to alert the male of her presence. Wilkinson (1962) reported that *Ips avulsus*, *I. grandicollis*, and *I. calligraphus* have sound-producing structures between the head and pronotum. The stridulating organ is described as the vertex-pronotal type that features a ‘pars stridens,’ or file-like structure, on the head and a ‘plectrum,’ or series of coarse ridges, on the interior side of the pronotum (Barr 1969, Lanier and Cameron 1969). Stridulation is produced when the head is retracted into the pronotum (Wilkinson *et al.* 1967). This head movement can be observed when the live female beetle is held between the fingers or forceps. For the three species of *Ips* pine engravers in the south, only the female has the vertex-pronotal type stridulating organ (Barr 1969).

## Gallery construction

The gallery construction of pine engravers is visually distinctive. Males initiate attack by boring through the outer bark to create a nuptial chamber within the inner bark and phloem (Wilkinson and Foltz 1982). ‘Pitch tubes’ or ‘boring dust’ are the external signs that a beetle has entered the tree. Frass and boring dust, which are pushed outside of the entry hole by the male beetle, contain aggregation pheromone. Pheromones attract females and additional males to the host tree. Females arrive and stridulate outside the entry hole to gain access to the nuptial chamber. Copulation occurs within the nuptial chamber. With *I. grandicollis* and *I. calligraphus*, a single male will mate with 2 to 5 females (Wilkinson and Foltz 1982, Drooz 1985). Once mated, each female creates an egg-laying gallery leading away from the nuptial chamber. Egg-galleries follow the grain of the wood. These adult tunnels give the *Ips* galleries their characteristic “Y” or “H” shape (Wilkinson and Foltz 1982). Females position phloem shavings and frass behind them and use their scoop-shaped posterior to push the waste material out of the egg-galleries. Males then evacuate the excess frass and phloem from the galleries. Males maintain residency for several days after mating and assist in gallery defense and maintenance (Kirkendall et al. 1997). Males will block entry into the nuptial chamber with their bodies, attempting to prevent access of predators and competitors. Vité et al. (1972) noted that male *I. calligraphus* regulate the harem size by ceasing pheromone synthesis and blocking the entrance when having mated with a sufficient number of females (Kirkendall et al. 1997).

Once egg-galleries are created by the females, they will chew evenly spaced notches, or oviposition niches, along the walls of the egg gallery. *Ips calligraphus* will usually create oviposition niches on one side of the gallery, while *I. avulsus* will create their niches on both sides (Wagner et al. 1988). The oviposition niches are spaced apart by an average of 5.9 mm for

*I. avulsus* but are closer together for *I. grandicollis* and *I. calligraphus* (Gouger *et al.* 1975). The female oviposits a single egg in each niche and a ‘phloem plug’ is constructed to seal the egg into place and to protect it from desiccation and foraging predators (Gouger *et al.* 1975). The larvae mine phloem perpendicularly from the egg-laying gallery (Wagner *et al.* 1988). *Ips grandicollis* and *I. calligraphus* create winding larval galleries that only occasionally overlap each other (Drooz 1985). *Ips avulsus* larval galleries are short and terminate in a widened pupal cell where larvae consume mutualistic fungi (Gouger *et al.* 1975, Drooz 1985, Klepzig *et al.* 2001).

### **Development and life history**

The development rate of *Ips* pine engraver beetle brood is dependent on temperature. Their life cycle is very slow below a base temperature of 15°C, and as such, they are not considered a problem in the winter (Wilkinson and Foltz 1982, Connor and Wilkinson 1983). Their development time increases with temperature until an upper threshold limits survival. Wagner *et al.* (1988) reported that the fastest development times for *Ips* pine engravers occurred at temperatures between 34°C and 37°C. Their upper threshold is relatively high compared to other bark beetles of North America, and for this reason, pine engravers develop well in warm climates. At 35°C, *Ips avulsus* has the fastest development time of only 2 weeks (Wagner *et al.* 1988). The optimal development time of *Ips calligraphus* is approximately 18 days at 35°C (Wagner *et al.* 1987). Wagner *et al.* (1987) demonstrated that *Ips* pine engravers have better heat tolerance than *D. frontalis*, while *D. frontalis* has the better cold tolerance. This sensitivity to low temperatures indicates that extreme cold winters may have a negative effect on *Ips* populations. Wagner *et al.* (1988) reported 0% survival of eggs at 10°C and minimal development at 15°C.

The number of generations per year varies due to seasonal temperatures. Also, generations overlap throughout the year, so we recognize a generation as the time it takes for an egg to mature into an adult. With this in mind, *Ips avulsus* could have about 10 generations per year (Drooz 1985). *Ips grandicollis* and *I. calligraphus* mature slower and could have approximately 6 generations per year (Drooz 1985).

## **POPULATION DYNAMICS AND MORTALITY FACTORS**

### **Interspecific and intraspecific competition**

Members of the southern pine beetle guild maintain their reproductive potential by reducing or avoiding competition (Flamm et al. 1987). As attacking populations increase, interspecific and intraspecific competition for the limited phloem resource occurs (Stephen 2011). Variable arrival patterns and host partitioning limit competition among species (Coulson et al. 1986, Wagner et al. 1985, Stephen 2011). Flamm et al. (1987) found that early colonization of *I. avulsus* in the bole will inhibit the colonization of *I. calligraphus* in that same region. Therefore attack strategies of *Ips* pine engravers include a preference for colonizing unattacked sections of the tree. Intraspecific competition causes an exponential decrease in reproductive success as attacking adult populations increase (Robins and Reid 1997). When attacking densities are high, members of the guild reduce competition through ‘reduction’ (i.e. restricting the number of eggs oviposited so that resources are not limited) or avoidance (repositioning attacks and galleries to limit interaction with neighbors) (Coulson et al. 1976b, Flamm et al. 1987). *Ips calligraphus* avoid competition by limiting oviposition in a similar density-dependent manner as that observed with southern pine beetle (Coulson et al. 1976b, Flamm et al. 1987). However, *Ips avulsus* does not exhibit this ‘density-dependent feedback mechanism’ (Flamm et

al. 1987). Also, reemergence will occur sooner when densities are extreme in order to limit overpopulation. An avoidance response exists during gallery construction, and this is demonstrated when adult and larval gallery length decreases to limit competition (Flamm et al. 1987).

The most influential competitors for the phloem resource are large Cerambycidae. *Monochamus* spp., or southern pine sawyers, are the chief interspecific competitor of *Ips* pine engravers because of their ability to consume vast quantities of phloem. Southern pine sawyers have a kairomonal attraction to *Ips* pheromones (Allison et al. 2001, Dodds et al. 2001, Allison et al. 2012a). Dodds et al. (2001) also found that larvae of *Monochamus* are facultative predators of *Ips* larvae. Through intraguild predation, larvae of *Monochamus* acquire a supplemental food source, and the added nutrition may speed development time. Facultative predation, coupled with phloem consumption, can dramatically reduce survival of *Ips* brood (Coulson et al. 1976a, Miller 1985, Stephen 2011, Allison et al. 2012b).

### **Predators and parasitoids**

Pine engraver beetles coevolved with an abundant and specialized complex of natural enemies (Dahlsten 1982, Dahlsten and Whitmore 1989). By a kairomonal response, many of these natural enemies are attracted to *Ips* aggregation pheromones (Allison et al. 2012a). They can negatively influence the populations of *Ips* beetles, and they may play an important role in suppressing outbreaks (Dixon and Payne 1979a, Dahlsten and Whitmore 1989, Reeve 1997). For example, *Thanasimus dubius* (F.) is important in influencing *D. frontalis* populations and may influence *Ips* populations as well (Dixon and Payne 1979b, Reeve 2011).

*Ips* beetles are preyed upon by a diverse assortment of predators and parasitoids (Dixon and Payne 1979a). The families Cleridae and Trogossitidae affect the populations of bark beetles

as both adults and larvae. The clerids, particularly *Thanasimus dubius* (F.), influence the population dynamics of *Ips* bark beetles (Reeve and Turchin 2002, Reeve 2011). From the Trogossitidae, *Temnochila virescens* (F.) and *Tenebroides* spp. are recognized as common bark beetle predators (Linit and Stephen 1983, Stephen 2011). Histeridae are important natural enemies of *Ips* bark beetles (Aukema and Raffa 2002, 2004). Adult Histeridae find the entrance holes of *Ips* and seek the tunneling adults inside, and they also consume eggs and first instar larvae. The most common Histeridae preying on *Ips* are *Platysoma attenuata* Le Conte, *P. cylindrica* (Paykull), *P. parallelum* (Say), and *Plegaderus transversus* (Say) (Shepherd 2004). Aukema and Raffa (2004) found that *P. cylindrica* adults would consume *Ips* adults while the *P. cylindrical* larvae exploited the *Ips* gallery system to reach their soft-bodied prey. Several genera in the subfamily Colydiinae (a subfamily of Zopheridae), including *Lasconotus pusillus* LeConte, *L. referendarius* Zimmerman, and *Aulonium* spp. Erichson, are effective within-bark predators of egg and early instar stages of bark beetles (Rohlf III and Hyche 1981). *Corticeus glaber* (LeConte) and *C. parallelus* (Melsheimer) from Tenebrionidae are described as facultative predators of egg and early instar larvae (Smith and Goyer 1980, Goyer and Smith 1981). Some Hemiptera in the family Anthocoridae are effective subcortical predators due in part to their small size and flat body shape (Lattin 1999). Two such anthocorids, *Scoloposcelis mississippiensis* (Drake and Harris) and *Lyctocoris* Hahn, are known to cause significant mortality on bark beetles (Linit and Stephen 1983, Lattin 1999). Some Diptera species are listed as natural enemies of pine engraver beetles. These include the genera *Lonchaea* Fallen and *Medetera* Fisher van Waldheim, whose larvae dwell in *Ips* larval galleries and prey on *Ips* larvae (McAlpine and Morge 1970, Aukema and Raffa 2004). Numerous Hymenoptera parasitize *Ips* pine engraver beetles (Berisford et al. 1971). These include *Coeloides pissodis* (Ashmead)

(Braconidae), *Eurytoma tomici* Ashmead (Eurytomidae), *Heydenia unica* Cook and Davis (Pteromalidae), *Rhopalicus* spp. (Pteromalidae) and *Roptrocerus xylophagorum* (Ratzeburg) (Torymidae) (Bushing 1965, Berisford et al. 1971).

### **Fungal associates**

*Ips* pine engravers are nonmycangial, but interactions with associated fungi occur. *Ips* beetles are vectors of the antagonistic blue-staining fungi, e.g. *Ophiostoma* (previously *Ceratocystis*) *minus* (Hedgecock) and *O. ips* (Rumbold) Nannfeldt (Yearian *et al.* 1972). The fungal spores are consumed and stored in their gut, and excretion of feces spreads the spores (Yearian *et al.* 1972, Gouger *et al.* 1972, Connor and Wilkinson 1983). Blue-staining fungi can weaken a tree, allowing attacking beetles to more easily overcome host defenses (Yearian *et al.* 1972, Lieutier 2002, Klepzig and Hofstetter 2011). Weakening the tree occurs when blue-staining fungi spread from the phloem into the sapwood, effectively cutting the flow of water and nutrients to the crown (Wilkinson and Foltz 1982). However, *Ophiostoma* spp. causes more harm to *Ips* than benefit as it quickly spreads through the whole tree and limits brood development. The negative relationship of blue-stain on *D. frontalis* is well understood, as it inhibits egg production and decreases larval survival (Barras 1970, Hofstetter et al. 2006). *Ips grandicollis* and *I. calligraphus* larvae do not consume a fungus for nutrition. Therefore, their larval galleries are relatively long. Unlike *I. grandicollis* and *I. calligraphus*, *Ips avulsus* shares a mutualistic relationship with *Entomocorticium* fungi (Klepzig et al. 2001). *Ips avulsus* larvae do not create long larval galleries, but instead their larval gallery foraging patterns are similar to *D. frontalis* (Gouger *et al.* 1975, Klepzig et al. 2001, Stephen 2011). *Ips avulsus* galleries are 4-8 mm long and the distal end is broadened where the larvae feeds on *Entomocorticium* fungus.

## COLONIZATION OF HOST TREES

### Attraction to hosts

*Ips* pine engravers are not normally attracted to healthy trees. Healthy trees produce a less detectable concentration of host semiochemicals (Mattson and Haack 1987). However, stressed or damaged trees release higher rates of semiochemicals, such as monoterpenes and ethanol that have an attractive kairomonal effect to bark beetles (Wood 1982, Byers 1989, Miller and Borden 2000). Without this chemical signature, *Ips* primary attraction does not occur. Interestingly, host monoterpenes are toxic to bark beetles, and this may make initial colonization difficult (Smith 1966, Cook and Hain 1988).

Other factors limiting colonization are the tree's defenses. Success of *Ips* and other bark beetle colonization is dependent on overcoming the constitutive and induced defenses that protect the tree (Berryman 1972, Cook and Hain 1986, Raffa and Berryman 1987). The constitutive or primary line of defense in pine trees is a resin duct system. Resin flows from wound sites and traps invading beetles. The presence of pitch tubes indicates a good supply of moisture (Wilkinson and Foltz 1982). When water availability decreases, resin pressure is reduced and this defense becomes less effective. Pase III (2011) noted that pitch tubes are absent when *Ips* attacks occur in drought stressed trees. The presence of boring dust, or a combination of frass and chips of bark and phloem pushed out by the beetle, will indicate that successful colonization has occurred. Induced defenses, also called hypersensitive reactions, involve walling-off the bark beetles thereby trapping them in necrotic, resinous tissue (Berryman 1972, Cook and Hain 1986, Cook and Hain 1988). This walled-off phloem has lowered nutritive value and a higher amount of toxic or inhibitory compounds that cause bark beetle attacks to be unsuccessful. Also of great importance, the hypersensitive reaction can effectively prevent the

spread of fungal infections associated with bark beetle attack (Berryman 1972, Cook and Hain 1986, 1988). *Ips* pine engravers are attracted to stressed hosts that intrinsically provide a better environment for the survival of attacking beetles, and in doing so, they avoid some of the primary and induced defenses.

### **Factors that enable *Ips* attack**

*Ips* pine engravers are native species to the United States, and disturbance events play an important role in enabling their outbreaks. Climate change, drought, or excessively moist conditions alter plant and insect interactions. As a result, latent populations of native insects can reach outbreak conditions (Ayres and Lombardero 2000, Coyle et al. 2013). Human alteration of forest conditions may also unintentionally cause novel native insect population growth (Clarke *et al.* 2000, Black 2005).

*Ips* engraver beetles preferentially attack dying trees and logging slash, but healthy trees may be attacked when they are damaged or when tree vigor is reduced (Fettig *et al.* 2007, Schowalter 2012). Some abiotic agents that increase susceptibility to attack include drought, fire damage, wind damage, severe storms, and damage to residual trees caused by logging activity (Bryant et al. 2006). In particular, *Ips* outbreaks are frequently linked to extended periods of drought (Wilkinson and Foltz 1982, Clarke *et al.* 2000). Also, lightning can be very important in creating host trees that are susceptible to *Ips* colonization (Coulson et al. 1999). According to Wilkinson and Foltz (1982), an estimated 40 to 60% of *Ips* and black turpentine beetle infestations in Georgia were associated with lightning struck trees.

The biotic agents that predispose trees to an infestation are pathogens and vegetative competition. Fusiform rust (*Cronartium fusiforme* Hedgecock and Hunt ex Cummins) may damage trees and make them vulnerable to an *Ips* attack (Wilkinson and Foltz 1982). Other

pathogens that may weaken pine trees include annosus root disease (*Heterobasidion annosum* [Fries] Brefeld) and littleleaf disease (*Phytophthora cinnamomi* Rands) (Bryant et al. 2006). Additionally, observable stand-wide decline in tree health, potentially incited by root diseases such as *Leptographium* spp., will result in an increase of *Ips*-attacked trees (Eckhardt 2003). Another biotic interaction that can weaken trees is competition. Limited growing space causes forest vegetation to reallocate photosynthates (Oliver and Larson 1996, Fettig et al. 2007). Trees increase their competitive advantage with respiration maintenance, radial growth, and height growth. Consequently, defense mechanisms are compromised and resistance to pests is reduced (Fettig et al. 2007).

#### **Association with *Dendroctonus frontalis* and arrival pattern**

The attack patterns of southern pine beetle are closely tied with *Ips* populations. The successful colonization of southern pine beetle may trigger a secondary aggregation of *Ips* bark beetles to the host tree. *Ips* pine engravers respond to pheromones of other members of their guild (Birch et al. 1980). The ephemeral phloem resource is quickly exploited by a complex of herbivores, most of which are competitors from Scolytinae and Cerambycidae (Dixon and Payne 1979b). The arrival of colonizing insects in response to southern pine beetle attack occurs in a predictable manner (Camors and Payne 1973, Dixon and Payne 1980, Stephen 2011). Peak of arrival for *Ips avulsus* was 12 days after initial *D. frontalis* colonization, and the attack density was more concentrated in those early weeks than the other two *Ips* species. The peak of *I. grandicollis* arrival was at 18 days, but it featured a less concentrated attack pattern. *Ips calligraphus* peak arrival occurred even later, about 3 weeks following initial southern pine beetle attack (Dixon and Payne 1979b, Stephen 2011). Though *Ips* have a close relationship with

*D. frontalis* primary attacks, *Ips* bark beetles are opportunistic colonizers that utilize many other forms of damaged pine that *D. frontalis* does not.

## **CHARACTERISTICS OF OUTBREAKS**

Multiple symptoms can indicate the presence of pine engravers. From a distance, yellow and red discoloration of crown foliage may be evidence of bark beetle colonization. *Ips* gallery construction can girdle the tree's phloem, cutting the flow of nutrients between root and foliage. Additionally, introduction of blue-stain fungi can inhibit the flow of water through the xylem and nutrients through the phloem (Wilkinson and Foltz 1982). The associated needle discoloration and subsequent death of individual limbs, known as 'flagging,' may be the first sign of an *Ips* attack (Pase III 2011). A tree initially attacked by *I. avulsus* will show flagging in the crown and will often decline downwards from the top. Up close, the presence of pitch tubes or boring dust is a clear indication that an attack has occurred (Billings and Pase III 1979). These are general signs of bark beetle attack, and the bark should be removed to confirm the species present. Examination of the distinct galleries under the bark remains the surest way to diagnose an *Ips* attack (Wilkinson and Foltz 1982).

*Ips* pine engraver densities change as a result of multiple factors. When their abundance is low, *Ips* colonize logging slash and dying pines (Hetrick 1942, Wilkinson and Foltz 1982, Clarke *et al.* 2000). However, *Ips* densities can increase with the abundance of lightning struck, windthrown, uprooted, and fire-damaged trees. When high densities are reached, *Ips* bark beetles are then capable of contributing to tree mortality of stressed hosts (Connor and Wilkinson 1983, Hetrick 1942). Hotter summers are conducive to abundance increases as southern pine engraver beetles are well adapted to temperatures between 25 and 37.5°C (Wagner *et al.* 1985, Wagner *et*

al. 1988, Stephen 2011). In contrast, the development rates, oviposition rates, and re-emergence rates of *Dendroctonus frontalis* decline at temperatures higher than 32°C (Wagner et al. 1981, 1984a, 1984b). Southern pine engraver beetles are not well adapted to winter temperatures, so, in theory, unseasonably warm winter temperatures could allow greater survival of overwintering brood and adults (Wagner *et al.* 1985, Bale et al. 2002, Stephen 2011).

Abundant *Ips* populations and the susceptible host trees are necessary conditions for outbreaks. An *Ips* outbreak is characterized by small pockets of tree mortality (Thatcher 1960, Bryant 1983, Clarke *et al.* 2000). Sometimes individual trees may be attacked throughout a stand. The scattered tree mortality makes *Ips* outbreaks difficult to detect with aerial surveys. Investigating individual trees in person is one way to determine presence and densities of *Ips*. Land managers require better monitoring techniques for assessing *Ips* pine engraver beetle abundance and associated tree mortality.

## **RESEARCH GOALS**

The ecological and economic impacts of *Ips* pine engravers are often overlooked (Clarke et al. 2000). Owing to the sporadic and elusive characteristics of their outbreaks, mortality caused by *Ips* is frequently unreported (Clarke et al. 2000). Land managers see the aftermath of *Ips*-associated mortality, but they do not have a means for reporting estimated damage or impact. Additionally, foresters and entomologists know little about the actual populations of *Ips* in outbreak stands or even in stands with low level populations. The relative abundance of predators and competitors in *Ips* outbreaks is also poorly understood. This research was designed to help develop methods for future *Ips* monitoring and tree mortality estimations.

A multifaceted approach to monitoring was used. This combined pheromone trapping with both simple and complete methods of tree mortality surveys. The simple mortality survey was a vehicle-based road survey in which dead and dying trees were tallied, and the complete survey was a ground-based 100% survey of tree mortality in a predefined study area.

Several objectives were addressed in this research: (1) to determine abundance of *Ips* in the stands exhibiting minimal mortality and compare this to stands with high tree mortality, (2) monitor, through pheromone traps, the relative population abundance of the three *Ips* species between regions and seasons, (3) to determine, through pheromone traps, the relative abundance of *Ips* natural enemies and compare between regions and seasons, (4) to evaluate the effectiveness of a vehicle-based road survey and a 100% mortality survey for assessing the impact of southern pine engraver beetles, and (5) to determine if *Ips* abundance, estimated through pheromone traps, correlates with the surveys of tree mortality.

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## CHAPTER 2 – PHEROMONE TRAPPING OF *IPS* AND THEIR NATURAL ENEMIES

### INTRODUCTION

In the southeastern United States, pines are often attacked and colonized by the engraver beetles, *Ips avulsus* (Eichhoff), *Ips grandicollis* (Eichhoff), and *Ips calligraphus* (Germar). *Ips* pine engravers are the most ubiquitous pine-infesting insects of Arkansas, Louisiana, and Texas (Clarke *et al.* 2000, Bryant *et al.* 2006, Pase III 2011). Despite being so common in southern pine forests, our understanding of *Ips* population dynamics is relatively limited.

*Ips* abundance often increases when climate is favorable (hot and dry) and suitable host resources are available. When high densities are reached, *Ips* beetles may become capable of damaging otherwise healthy trees. *Ips* beetles normally sustain a population by propagating in fallen trees, weakened suppressed trees, and the lower limbs of the crown (Wilkinson and Foltz 1982, Bryant *et al.* 2006). However, when their numbers increase, they are also capable of infesting trees that are stressed or weakened by a variety of damaging agents. Harvesting practices that leave residual slash or damaged standing trees provide host material for *Ips* (Fettig *et al.* 2007). Large-scale disturbances (e.g., drought, fire, wind damage, and severe storms) are often associated with *Ips* population growth and outbreaks (Wilkinson and Foltz 1982, Clarke *et al.* 2000, Bryant *et al.* 2006). In particular, droughts frequently precede *Ips* and other bark beetle outbreaks (Mattson and Haack 1987, Christiansen and Bakke 1997, Pase III 2011). Periods of hot, dry weather often results in *Ips* outbreaks, perhaps linked to weakened pine defenses. Pase III (2011) postulated that drought is so frequent in the West Gulf Coastal Plain that every pulpwood stand will be affected by drought-like conditions at least once in its rotation, and sawtimber stands will have been influenced by two or more occurrences of significant drought.

With frequent reoccurrence of drought, *Ips* infestations will likely continue to be an integral part of pine management in the southeastern United States.

In natural forest succession, *Ips* beetles play a pivotal role in forest health processes. They are among the earliest colonizers of weak and dying pines (Fettig *et al.* 2007). In many respects, bark beetles, such as *Ips*, are drivers of healthy forest ecosystems by selectively killing less vigorous trees (Black 2006, Fettig *et al.* 2007). They contribute to gap dynamics and processes of natural forest succession, thereby diversifying and strengthening the forest against future attacks (Amman and Logan 1998, Berryman 1982). However, anthropogenic influences in forests confound their positive role. Outbreaks have a tendency to occur in stands that are changed from their historical successional trends, such as loblolly pine planted in areas traditionally dominated by longleaf pine (Clarke *et al.* 2000). Additionally, even-aged stands are more vulnerable to outbreaks than diverse stand structures which feature higher hardwood densities (Schowalter and Turchin 1993, Black 2006). Burning or thinning in these managed forests may also lead to outbreaks, though they are typically short-lived and selective to damaged residual trees (Fettig *et al.* 2007).

Pheromones have been influential in detection, monitoring, and assessment of bark beetles. Pheromone trapping was used to detect outbreaks and changes in population densities of *Ips typographus* (L.) and *Ips acuminatus* (Gyllenhal) in Europe (Byers 1989, Angst *et al.* 2012, Faccoli *et al.* 2012). Aggregation pheromones have been developed for the *Ips* species of the southeastern United States. These pheromones, deployed on Lindgren® multiple-unit funnel traps that imitate the shape and color of a tree trunk, can lure dispersing *Ips* beetles and aid in monitoring abundance (Lindgren 1983). However, little research has been done in this region using these pheromones to evaluate changes in *Ips* abundance over time. Furthermore, records of

their outbreaks are rarely kept (Clarke *et al.* 2000). The scarcity of research is at least partly due to the short-lived nature of outbreaks that are difficult to actively observe. Infested trees are scattered throughout stands in small patches, and, as a result, direct control measures are not economically profitable (Thatcher 1960, Bryant 1983, Clarke *et al.* 2000). Researchers and land managers can benefit from improved techniques for monitoring *Ips* abundance and understanding how changes in numbers of *Ips* relate to outbreaks.

Through anecdotal evidence, researchers have recognized a close relationship between drought and subsequent *Ips* outbreaks. Understanding this relationship will allow individuals to draw conclusions about how, when, and where *Ips* pine engravers threaten forests. Drought conditions occurred in Arkansas and East Texas in 2011 and early 2012. Coinciding with this drought, pine trees became infested with *Ips* engraver beetles and subsequent mortality was observed. Such a natural event provides an opportunity to investigate monitoring techniques for *Ips* beetles.

The primary objective of this study was to assess, using pheromone traps, the relative abundance of *Ips* pine engraver beetles (total numbers of each species) in different regions and seasons. Additional secondary objectives were to evaluate sex ratios of *Ips* beetles among species and seasons, and to identify and estimate relative abundance of select natural enemies attracted to *Ips* pheromone. The results enhance knowledge of *Ips* population dynamics and provide a basis for improving future monitoring studies.

## **MATERIALS AND METHODS**

Trapping sites were in three geographically distinct regions. Within each of these regions, two sites were chosen: a ‘high incidence’ of tree mortality site and a ‘low incidence’ site. These

designations were assigned subjectively. In a high incidence site, tree mortality was more visibly evident than in the nearby (<50 mile distance apart) low incidence site. It was presumed that a greater number of *Ips*-infested trees existed in high incidence sites. The regions included the Ozark National Forest in northwestern Arkansas, the Ouachita National Forest in west central Arkansas, and two National Forests in eastern Texas, the Sam Houston and Davy Crockett National Forests. Each study site (Table 1) was an even-aged pine stand, which was commercially thinned to a relatively low basal area (10-20 m<sup>2</sup>/hectare), intersected by a Forest Service road. For surveys of tree mortality, a one-kilometer long section of the Forest Service road and 100 meters on both sides of the road were designated. Stands were selected that had similar structures (i.e. pine dominance, sawtimber-sized diameter, and comparable pine basal area per hectare).

Five Lindgren<sup>®</sup> 12-unit funnel traps were erected on one side of the road within each stand. The traps were located 150 meters from the Forest Service road and were inside the boundaries of each pine stand so as to be unaffected by corridor effects of roads or stand boundaries. A predesignated azimuth was established so that the 150 meter distance to the trap was roughly perpendicular to the road. The funnel traps were spaced 250 meters apart. This results in traps aligned on a transect that followed the contour of the Forest Service road, as opposed to a straight line transect within the forest. Traps were suspended ca. 2 meters above the ground using a telescoping ‘shepherds hook’ (Figure 1). The hanging structure, affixed to a steel t-post with wire, was composed of a 1.9 cm dia. (3/4”) electrical metallic tubing (EMT) conduit bent 90 degrees at the top that was inserted into 2.5 cm (1”) EMT conduit. The trap was extended and held in place with a 0.64 cm (1/4”) bolt and nut fastened through the EMT conduit (Figure 2). All trap locations were marked using a Garmin<sup>™</sup> GPSmap 60CSx rated as accurate to +/- 10

meters. Traps remained at the same locations during each season. The seasons of trapping were spring 2012, summer 2012, fall 2012, and spring 2013, and traps were monitored and emptied at 7, 14, and 21 days post set-up (Table 2).

Each trap was baited with aggregation pheromones of *Ips* pine engravers. Host volatiles such as ethanol or monoterpenes (e.g. alpha-pinene) were not used. The decision to exclude host volatiles was intended to keep trap catches to a more manageable level, because attraction to pheromones is synergistically enhanced by host volatiles (Raffa 2001, Allison et al. 2012b). Additionally, the absence of host volatiles allowed traps to capture *Ips* beetles plus natural enemy species that have kairomonal attraction to *Ips* pheromone. The pheromone lures, bubble-cap style provided by Synergy Semiochemicals Corp., included (±)-ipsdienol (racemic ipsdienol with 1:1 mix of the plus and minus enantiomers), (±)-ipsenol (racemic ipsenol with 1:1 mix of the plus and minus enantiomers), and *cis*-verbenol. The *cis*-verbenol, a derivative of alpha-pinene, has a presumed enantiomeric composition between 30:70 and 10:90 (R, S) (David Wakarchuk, Synergy Semiochemicals Corp., personal communication, 2013). The release rates were approximately 200 µg/day for ipsdienol, 400 µg/day for ipsenol, and 600 µg/day for *cis*-verbenol (David Wakarchuk, Synergy Semiochemicals Corp., pers. comm., 2013). The lures were combined into one perforated bag and, on each trap, externally hung from the fifth funnel from the top. The collection reservoir cup at the bottom of the trap was filled with propylene glycol (commercially available as recreational vehicle antifreeze) as a preservative. At collection, the reservoir contents were poured into Whirl-Paks® (Nasco). All trap collections were transported to the University of Arkansas Forest Entomology Lab for sorting and counting specimens.

The *Ips* specimens were identified to the species level, and most natural enemies were identified to genus and species or, in some cases, to family. The natural enemies recorded were those that are documented predators of *Ips* (Dixon and Payne 1979, Dahlsten 1982, Dahlsten and Whitmore 1989, Stephen 2011). Some additional associates, including *Monochamus* spp. and *Acanthocinus* spp. (Cerambycidae), were recorded because they also exhibit kairomonal attraction to *Ips* lures (Allison *et al.* 2012a). Trap catch data are reported for the following families: Curculionidae (Scolytinae), Cerambycidae, Cleridae, Trogossitidae, Histeridae, Zopheridae (Colydiinae), Tenebrionidae, and Buprestidae.

Sex ratios were determined using a 20-specimen random subsample of each of the three *Ips* species in every trap. This subsampling procedure was repeated throughout the course of the study on every trap collection date. The identifying characteristic for gender was the presence of the female stridulating organ located between the posterior portion of the head and the interior of the pronotum. This stridulating organ is visible in all three *Ips* pine engravers of the southeast (Wilkinson 1962). The head was removed in order to see the ‘pars stridens,’ a file-like structure that rakes against the ‘pectrum’ that located in the interior pronotum (Barr 1969). Together, these two structures make the stridulating organ only found in females. The pars stridens becomes easily visible when a directed light source is diffracted off of the structure. Likewise, the plectrum is visible at the anterior undersurface of the pronotum when the head is removed.

### **Statistical analyses**

Statistical analyses were completed with JMP Pro 10 (SAS Institute Inc., Cary, NC). The significance level for all analyses was set at  $\alpha = 0.05$ .

Several graphs were used to illustrate trends and relationships among the *Ips* trap data. In order to demonstrate the change in trap counts with each consecutive trapping period, the mean

trap counts of *Ips* (all species combined) were graphed for each site (Figure 3). Next, mean trap counts of each species were compared, with each trapping period represented by its own graph (Figures 4 through 7). In each of these graphs, the mean counts are an average of insect counts from 15 total collections from that site (five traps, each accumulated for a total of seven days of trapping, and repeated for three weeks).

To discover if the sex ratio of captured adults varied seasonally, the male ratio of each *Ips* species was determined for each trap by identifying the number of male beetles in a sample of 20 specimens (Figure 8). The male ratio of each sample was calculated as the number of male observations divided by the total observations. Traps with less than 10 observed specimens were excluded to eliminate most of the small sample bias. Additionally, subsamples from all three sampling weeks were pooled. As such, the new maximum sample size for each trap was 60 specimens. For each species, a one-way analysis of variance (ANOVA) was used to identify statistical significance between the sex ratios of each trapping season, and Tukey-Kramer Honest Significant Difference (Tukey's HSD) multiple comparison tests were used to further compare the mean sex ratios.

Traps counts of *Ips* pine engravers were compared to trap counts of select natural enemies to determine whether correlations existed (Figures 9 through 13). Predator families that are recognized as natural enemies of *Ips* (i.e. Cleridae, Trogossitidae, Histeridae, and Zopheridae [Colydiinae]) were analyzed in this way. The relationship between the trap count of pine sawyers (genus *Monochamus*) and total *Ips*, was similarly explored.

## RESULTS

### *Ips* trap counts

Over the course of the study, a total of 371,749 *Ips* bark beetles were trapped (Table 3). The most abundant species was *Ips grandicollis* (217,636 specimens), while 127,823 were *Ips avulsus*, and *Ips calligraphus* abundance was considerably lower at 26,290 specimens. *Ips calligraphus* was more abundant in Texas than in the Arkansas sites.

All species combined, the most *Ips* were trapped in spring and summer of 2012 (Figure 3). Few *Ips* were trapped in fall 2012 and spring 2013. During summer of 2012, more *Ips* were trapped in Arkansas sites than in Texas sites.

The results of the spring 2012 trapping season (Figure 4) clearly indicate collectively high *Ips* numbers, especially in the Texas sites, which averaged 2,690 *Ips* per trap. However, a change in regional abundance can be seen between spring 2012 and summer 2012. In an opposite effect from the previous trapping season, the Arkansas sites caught significantly more *Ips* than the Texas sites in summer 2012 (Figure 5). For summer 2012, the mean total *Ips* catches in each region were as follows: 2418 *Ips* in the Ozark National Forest sites, 3108 in the Ouachita National Forest sites, and only 946 in the eastern Texas sites. *Ips grandicollis* was the most abundant species in Arkansas during the summer 2012 trapping season, whereas *I. calligraphus* was the most abundant species in Texas. Interestingly, *I. calligraphus* is rare in the Ozark sites, and although also rare in Ouachita sites, the number of trapped adults was similar to Texas in spring 2013.

Fewer *Ips* were captured during the fall trapping season than in fall or summer trapping seasons (Figure 6). More total *Ips* were captured in Texas with 634 mean total *Ips*, most of which were *I. avulsus*. Negligible amounts of *Ips* were captured in the Arkansas sites; the mean total *Ips*

catches in the Ozark National Forest sites was only 29 and the Ouachita National Forest sites was 57.

The spring 2013 trapping season yielded the most consistent trap catches between regions (Figure 7). Furthermore, trap count variation was the lowest in this trapping period. Like the fall 2012 trapping season, the number of *Ips* was much lower than in the spring and summer 2012 trapping season.

### **Sex ratio of captured *Ips***

The overall average male ratio was 0.55 for *I. avulsus*, 0.29 for *I. grandicollis*, and 0.31 for *I. calligraphus*, and some seasonal variation existed (Figure 8). The male ratio of *I. avulsus* was not statistically different among all trapping seasons. For *I. grandicollis*, the average male ratio was significantly greater in the fall 2012 and spring 2013 trapping seasons than it was in Spring and Summer 2012. *Ips calligraphus* exhibited a significantly higher average male ratio in summer 2012 trapping season, and the ratio in the fall 2012 trapping season was also greater than the spring 2012 and 2013 trapping seasons.

### **Natural enemies of *Ips***

The most abundant predators in the traps were the following: *Thanasimus dubius* (Cleridae), *Enoclerus* spp. (Cleridae), *Temnochila virescens* (Trogossitidae), *Tenebroides* spp. (Trogossitidae), *Platysoma* spp. (Histeridae), *Lasconotus* spp. (Zopheridae: Colydiinae), and *Corticeus* spp. (Tenebrionidae) (Table 2). The most abundant phloem-feeding competitors were *Monochamus* spp. (Cerambycidae), *Acanthocinus obsoletus* (Cerambycidae), and *Buprestis lineata* (Buprestidae).

The correlation between total *Ips* trap catch and total Cleridae trap catch was not statistically significant ( $R^2 = 0.002$ ,  $p = 0.3581$ ) (Figure 9). This was the only natural enemy

group that did not have a significant and positive correlation with *Ips* trap catch. The correlation between total *Ips* trap catch and total Trogossitidae indicated a statistically significant relationship ( $R^2 = 0.18$ ,  $p < 0.0001^*$ ) (Figure 10). Similar to the correlation with Trogossitidae, the relationship between total *Ips* trap catch and total Histeridae trap catch had a significant correlation ( $R^2 = 0.21$ ,  $p < 0.0001^*$ ) (Figure 11). The correlation between total *Ips* trap catch and total Colydiinae trap catch indicated a statistically significant relationship ( $R^2 = 0.44$ ,  $p < 0.0001^*$ ) (Figure 12). This was the strongest correlation of all the natural enemy groups. Finally, the relationship between total *Ips* trap catch and total *Monochamus* spp. trap catch was statistically significant ( $R^2 = 0.04$ ,  $p = 0.0007^*$ ) (Figure 13), although the low  $R^2$  value indicates a weak relationship.

## DISCUSSION

### *Ips* trap counts

The beginning of this study coincided with a visible outbreak of *Ips* pine engravers in some areas of Arkansas and East Texas. Pine mortality, attributed to *Ips*, was observed by foresters and land managers during 2011. Visible tree mortality continued until around May of 2012, after which additional losses appeared minimal. Because little tree mortality occurred after the spring 2012 trapping season, it may be assumed that this study tracked changes in *Ips* abundance in a post-outbreak scenario.

Generally *Ips* abundance declined over the course of the study. In the Arkansas sites however, total *Ips* abundance actually increased between spring 2012 and summer 2012. By the spring 2013 trapping season, all trapping sites had low traps counts of all *Ips* species.

In the spring 2012 trapping season, abundance of *I. avulsus* and *I. grandicollis* was similar within each trapping site. This trend was not as distinct in other trapping periods. In fact, the abundance of *I. avulsus* and *I. grandicollis* in Arkansas during the summer trapping season was very different in that *I. grandicollis* greatly outnumbered *I. avulsus*.

There appear to be strong regional differences between Texas and Arkansas. For instance, *I. grandicollis* was very abundant in Arkansas, whereas the Texas sites caught more *I. calligraphus*. With this study the assumption was made that all stands were similar in structure and were subjected to the same climate conditions. A host of reasons may account for the differences in the abundance of each species. These include, but are not limited to the following: the temperature and weather events varied between regions, the dominant pine species were not the same in the Arkansas and Texas sites (i.e., *Pinus echinata* and *P. taeda* respectively), and the plant hardiness zones are dissimilar.

Fall catches were especially low, particularly in Arkansas. These results suggest that trapping would be more valuable in the spring and summer months. *Ips* pine engravers develop slowly in temperatures under 15°C (Wilkinson and Foltz 1982). Therefore, it may not be beneficial to trap for *Ips* pine engravers in the cooler fall and winter months. Nonetheless, a relatively high number of *I. avulsus* were captured in Texas during the fall season.

### **Sex ratio of captured *Ips***

The mean sex ratios of *I. avulsus* and *I. calligraphus* have been previously reported as 1:1 and 1:3 (♂:♀) respectively (Berisford and Franklin 1971, Cook et al. 1983). Trapping with *Ips* pheromones may bias the sex ratio of captured *Ips*. Dixon and Payne (1979) recorded the arrival patterns of *Ips* and other associates to *D. frontalis* infested trees without the use of artificial pheromones and reported a 1:0.7 (♂:♀) ratio for *I. avulsus* and a female dominated ratio for *I.*

*grandicollis* (1:1.42) and *I. calligraphus* (1:1.5). The reasons for this difference in gender response were hypothesized by Cook et al. (1983) who suggested that males might suffer more mortality than females when searching for a suitable host, or males might avoid competition with other males by preferentially attacking fresh hosts instead of a heavily colonized host tree.

Similar to Cook et al. (1983), results of this study were determined from adults responding to a pheromone combination. The sex ratios were 1:0.82 (♂:♀) for *I. avulsus*, 1:2.44 for *I. grandicollis*, and 1:2.23 for *I. calligraphus*. These results were similar to those reported by Cook et al. (1983), although they reported slightly higher sex ratios for *I. grandicollis* and *I. calligraphus*. Since attacking adults are attracted to the host by aggregation pheromones, it may be that males and females respond differently to certain pheromone combinations. Renwick and Vité (1971) noted that the sex ratio of attacking adults did indeed vary with different lure combinations.

The results of this study reaffirms that the *Ips* pheromone combination favors the capture of female *I. grandicollis* and *I. calligraphus*. *Ips avulsus* captures reflect a sex ratio similar to the expected 1:1 ratio.

### **Natural enemies of *Ips***

Some natural enemy species strongly responded to *Ips* pheromones in this study. The most common genera were *Thanasimus* (*T. dubius*), *Enoclerus* spp., *Temnochila* (*T. virescens*), *Lasconotus* spp., *Corticeus* spp., *Platysoma* spp., *Monochamus* spp., and *Acanthocinus obsoletus* (Table 3). By far, *Lasconotus* spp. was the most abundant predator, with more than 42,000 captured during the study. This genus has been described as a predator/scavenger that will feed on eggs and early instar larvae of *Ips* (Dixon and Payne 1979, Rohlf III and Hyche 1981).

*Corticеus* spp. was caught in great numbers later in the trapping study despite the decline in *Ips* abundance. Members of the genus *Corticеus* are described as facultative predators of bark beetles and it is hypothesized that they may use subcortical fungi after *Ips* emergence as a food source (Goyer and Smith 1981), a behavior that would help to explain their population growth even after *Ips* numbers declined.

Histeridae are often described as predators of *Ips* pine engravers (Aukema and Raffa 2002, Shepherd 2004, Aukema and Raffa 2004). This group exploits the gallery system of *Ips* pine engravers and will consume *Ips* of all life stages. Their effects on *Ips* populations may categorize them as effective biological control agents (Dahlsten 1989). Histeridae were most abundant in the hot summer months in this study.

As native bark beetles, *Ips* abundance is affected by a coevolved group of natural enemies. These insects may provide naturally occurring biological control against bark beetles. The presence of early arriving predators has a significant effect on bark beetle reproductive success (Stephen 2011). Based on spring trapping, the ratio between *D. frontalis* and *T. dubius* populations can be used as a predictive model for *D. frontalis* outbreaks (Billings 1988). In this study, the correlation between total *Ips* caught and Cleridae was not significant (Figure 9); however, the increase in abundance of Cleridae could be expected to lag behind the peak abundance of *Ips*, a fundamental relationship that was not fully explored in this study.

My data suggest a weak correlation between the number of *Ips* pine engravers caught to the number of natural enemies (Figures 10 through 13), and the strongest relationship occurred with *Lasconotus* spp. (Colydiinae). The existence of correlation is interesting, though further research would be necessary to investigate how *Ips* influence natural enemy abundance or,

conversely, how natural enemies affect *Ips* abundance. Research on *Ips* bark beetle population dynamics could benefit from a better understanding of top-down controls, i.e. predation.

## CONCLUSION

The pheromone lure system in this study was effective in catching all three southern pine engraver beetles and their natural enemies. The specificity of the lures had the added benefit of minimizing unwanted by-catch, which made specimen counting and identification simpler. *Ips* were captured in greater abundance during the spring and summer trapping seasons. Trapping during the fall did not yield as many *Ips* captures. More years of data with longer trapping seasons are needed to reaffirm the trends observed during this study.

Future studies should focus efforts on trapping during the spring or summer seasons. One important difference between these seasons is the abundance of natural enemies. In general, spring traps caught lower numbers of natural enemies than summer traps. One exception is that Cleridae were as abundant, or possibly more abundant, in spring as opposed to the other trapping seasons. If an assessment of natural enemy abundance is a goal of future studies, these data suggest that trapping should take place during the warmer, summer months. If a strict assessment of *Ips* abundance is an objective, then the spring months may be more suitable. Low or high *Ips* abundance, particularly *I. avulsus* and *I. grandicollis*, may be easily identified during the spring trapping season.

## TABLES

**Table 1: Stand characteristics of sites. Diameter at breast height (DBH), trees/hectare, and basal area (m<sup>2</sup>/hectare) are estimates based on 10% timber cruise. For the DBH averages, estimate is the mean quadratic diameter  $\pm$  standard deviation.**

Region	Ozark National Forest	Ozark National Forest	Ouachita National Forest	Ouachita National Forest
Site ID	OZH	OZL	OUH	OUL
Subjective Mortality Incidence	High	Low	High	Low
UTM	15S 424439E 3951622N	15S 414601E 3945453N	15S 396169E 3869673N	15S 388121E 3868433 N
Overall Average DBH (cm)	27.2 $\pm$ 9.9	34.5 $\pm$ 11.0	23.9 $\pm$ 9.8	30.8 $\pm$ 10.8
Pine Average DBH (cm)	31.2 $\pm$ 6.9	38.0 $\pm$ 10.4	29.5 $\pm$ 11.5	31.7 $\pm$ 10.0
Pine Trees/Ha.	203	163	152	185
Hardwood Trees/Ha.	161	96	258	27
Pine BA(m <sup>2</sup> /Ha.)	15.5	18.48	10.4	14.56
Hardwood BA(m <sup>2</sup> /Ha.)	5.73	5.75	7.98	1.27
Common Tree Species	<i>Pinus echinata, Quercus alba, Liquidambar styraciflua</i>	<i>P. echinata, Q. alba, Q. stellata</i>	<i>P. echinata, Q. stellata, Carya spp., Ulmus alata</i>	<i>Pinus echinata</i>

**Table 1 (cont.): Stand characteristics of sites. Diameter at breast height (DBH), trees/hectare, and basal area (m<sup>2</sup>/hectare) are estimates based on 10% timber cruise. For the DBH averages, estimate is the mean quadratic diameter  $\pm$  standard deviation.**

<b>Region</b>	<b>East Texas</b>	<b>East Texas</b>
<b>Site ID</b>	TH	TL
<b>Subjective Mortality Incidence</b>	High	Low
<b>UTM</b>	15R 234628E 3387344N	15R 320926E 3453364N
<b>Overall Average DBH (cm)</b>	36.4 $\pm$ 14.7	32.0 $\pm$ 15.3
<b>Pine Average DBH (cm)</b>	44.6 $\pm$ 11.8	38.6 $\pm$ 17.7
<b>Pine Trees/Ha.</b>	172	83
<b>Hardwood Trees/Ha.</b>	167	68
<b>Pine BA(m<sup>2</sup>/Ha.)</b>	20.18	12.98
<b>Hardwood BA(m<sup>2</sup>/Ha.)</b>	7.11	2.71
<b>Common Tree Species</b>	<i>Pinus taeda</i> , <i>Q. stellata</i> , <i>U. alata</i>	<i>P. taeda</i> , <i>L. styraciflua</i> , <i>U. alata</i>

**Table 2: Dates of trapping periods. Between set-up and take down, the trap contents were collected three times (weekly).**

		<b>Spring 2012</b>	<b>Summer 2012</b>	<b>Fall 2012</b>	<b>Spring 2013</b>
<b>Ozark National Forest Sites</b>	<b>Traps Set Up</b>	April 18	June 20	September 27	March 27
	<b>Traps Taken Down</b>	May 9	July 12	October 18	April 19
<b>Ouachita National Forest Sites</b>	<b>Traps Set Up</b>	April 18	June 20	September 27	March 27
	<b>Traps Taken Down</b>	May 9	July 12	October 18	April 19
<b>Eastern Texas Sites</b>	<b>Traps Set Up</b>	May 3	July 12	September 28	March 21
	<b>Traps Taken Down</b>	May 25	August 2	October 19	April 11

**Table 3: Total trap catches of *Ips* species and selected natural enemies (each value is a sum of 30 traps). Cleridae\* are *Thanasimus dubius* (F.), *Enoclerus nigripes* (Say), and *Enoclerus nigrifrons* (Say). Histeridae\*\* are *Platysoma cylindrica* (Paykull), *Platysoma attenuata* (Say), *Platysoma parallelum* Le Conte, and *Plegaderus transversus* (Say). Abbreviations for regions are as follows: OZ is Ozark National Forest, OU is Ouachita National Forest, and ET is eastern Texas (Sam Houston and Davy Crockett National Forests).**

	Spring 2012			Summer 2012			Fall 2012			Spring 2013			Total
	OZ	OU	ET	OZ	OU	ET	OZ	OU	ET	OZ	OU	ET	
<i>Ips avulsus</i>	7187	16965	36750	11945	19310	8188	474	724	15957	1904	4541	3878	127823
<i>I. grandicollis</i>	11387	19431	37128	60402	72754	7195	370	636	801	3130	3267	1135	217636
<i>I. calligraphus</i>	63	205	6822	186	1190	12999	34	242	2270	129	1005	1145	26290
<b>Cleridae*</b>	380	611	621	94	192	324	226	424	1958	901	1347	477	7555
<i>Temnochila virescens</i>	204	251	340	1947	1887	2410	3	4	417	217	717	639	9036
<i>Tenebroides</i> spp.	21	12	8	3	111	19	3	8	7	23	55	21	291
<b>Histeridae**</b>	55	55	1521	597	1489	1968	9	53	126	163	659	407	7162
<i>Lasconotus</i> spp.	266	187	23008	4107	10598	2206	204	288	744	225	288	192	42313
<i>Corticeus</i> spp.	21	12	1444	237	1254	35	35	25	8	744	1653	33	5501
<i>Monochamus</i> spp.	197	363	104	202	150	260	32	68	125	0	0	302	1803
<i>Acanthocinus obsoletus</i>	35	33	353	6	17	1018	0	7	569	1	0	243	2282
<i>Buprestis lineata</i>	0	3	1	107	121	36	0	0	6	0	0	1	275

## FIGURES

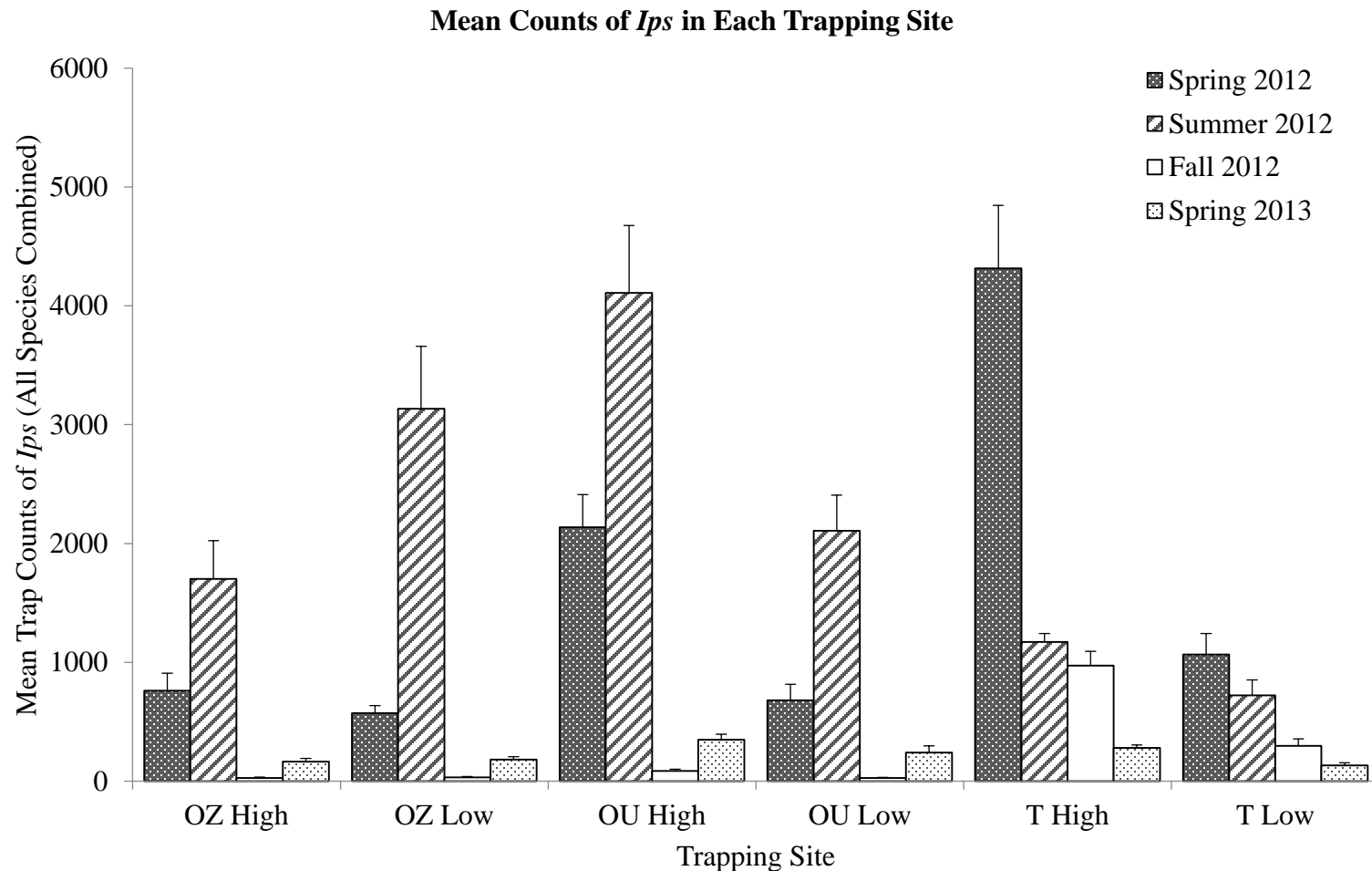
**Figure 1: Lindgren® 12-unit funnel trap set-up. Bottom of trap reservoir is ca. 2m above the ground. Photo credit: Chandler Barton.**



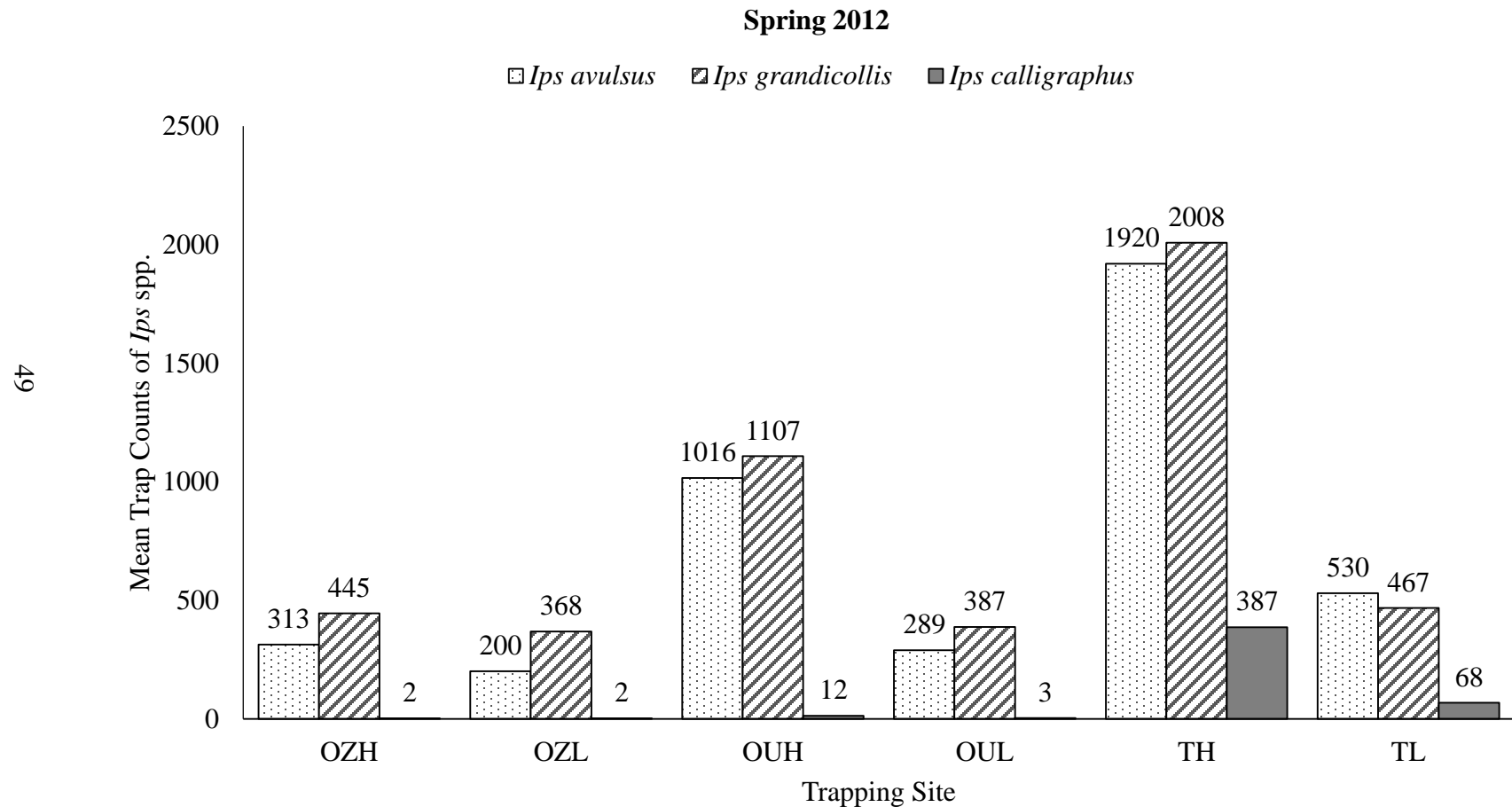
**Figure 2: Close-up of fastener for holding the electrical conduit at a fully extended position.**  
**Photo credit: Chandler Barton.**



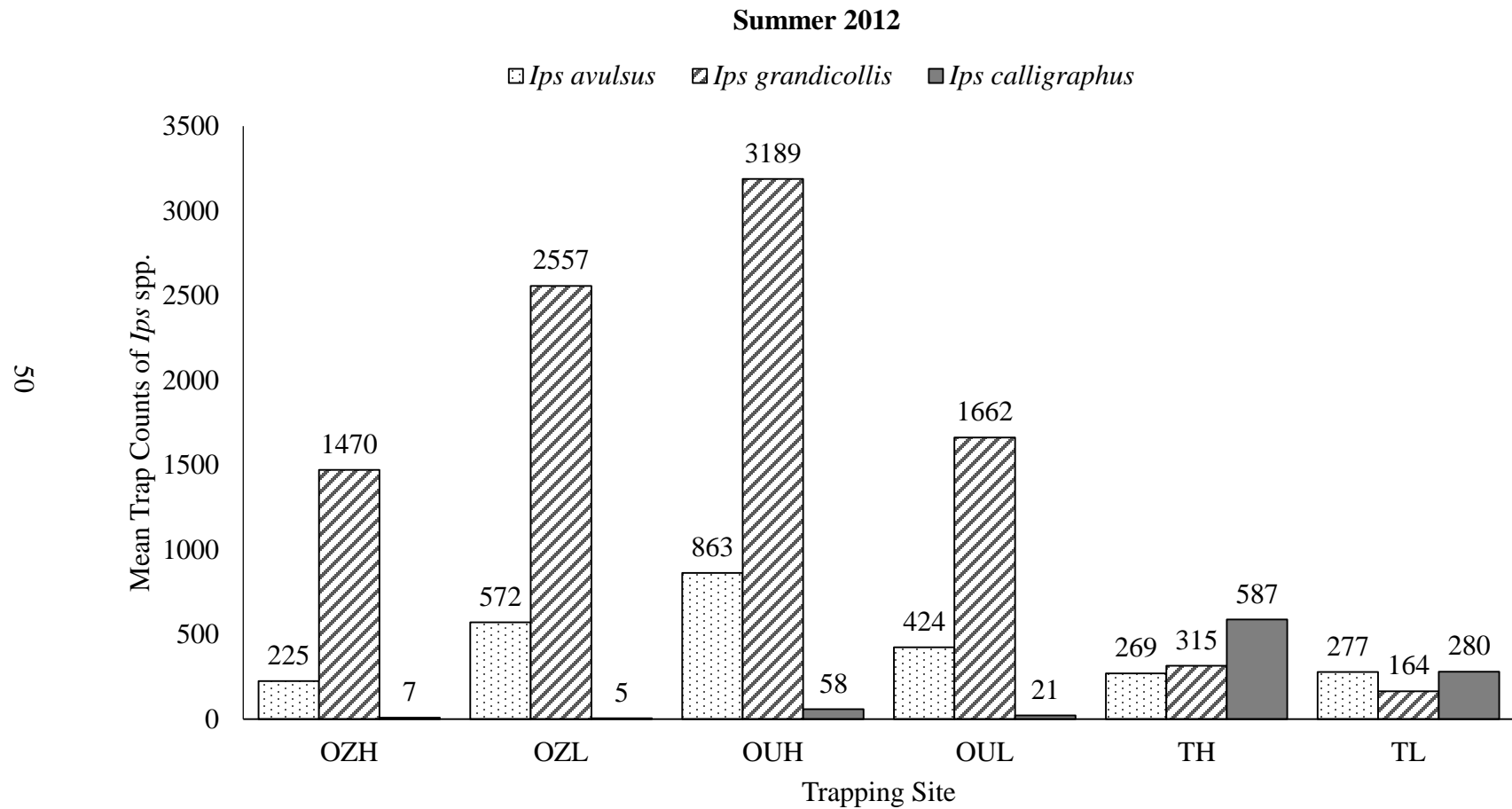
**Figure 3: Mean trap counts (n=15) of *Ips* (all species combined) by trapping site. For each site, the mean trap counts have been separated by trapping season. Trapping sites were located in the Ozark National Forest (OZ High and OZ Low), Ouachita National Forest (OU High and OU Low), and eastern Texas (Sam Houston [T High] and Davy Crockett [T Low] National Forest). High or Low makes the distinction between high incidence of tree mortality and low incidence of tree mortality. Error bars represent standard error of the mean.**



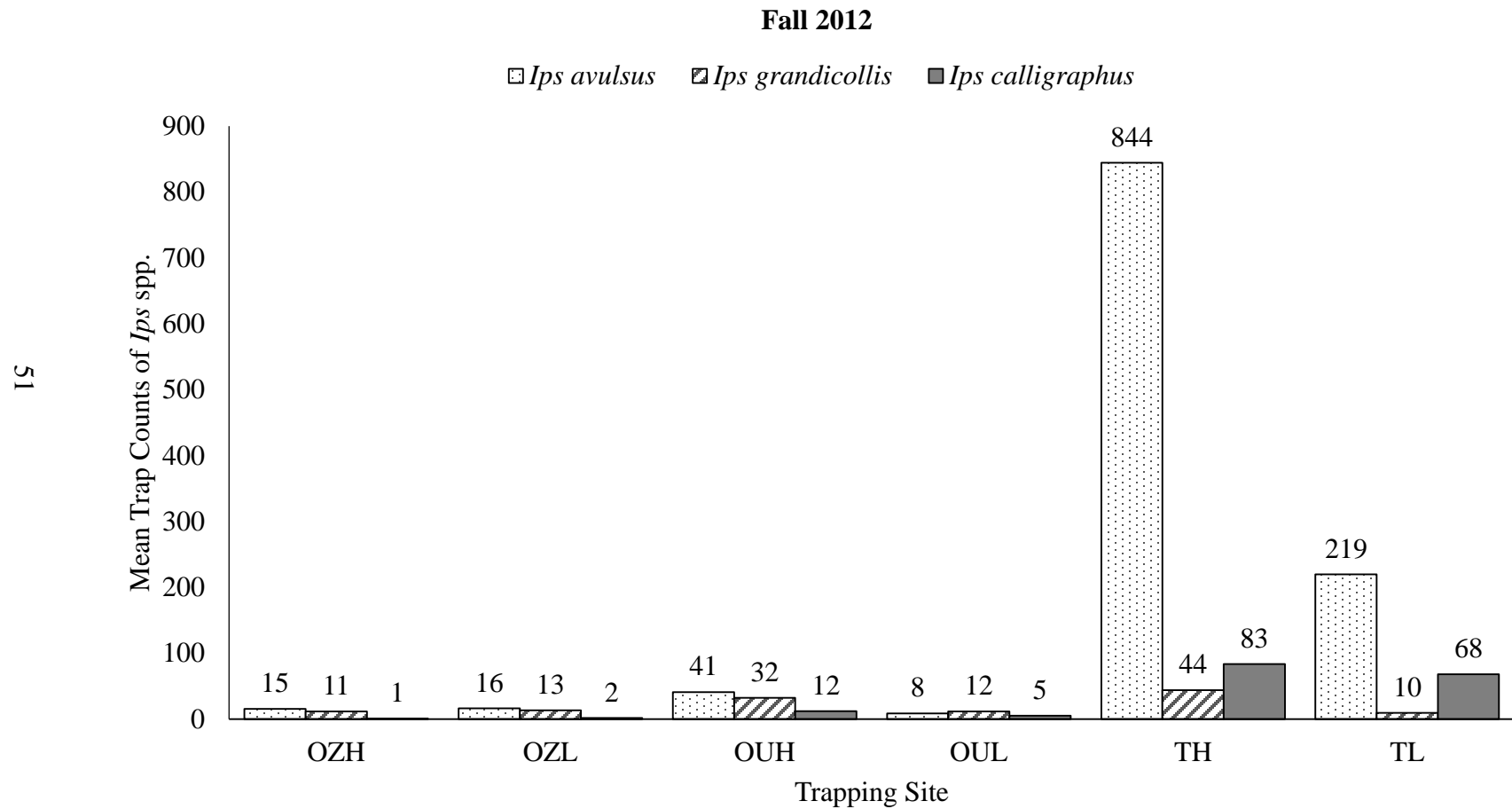
**Figure 4: Mean trap counts (n=15) of *Ips* spp. spring 2012 (Late-April through May) for locations in Ozark National Forest (OZH and OZL), Ouachita National Forest (OUH and OUL), and eastern Texas (Sam Houston [TH] and Davy Crockett [TL] National Forest). The last letter in the location ID makes the distinction between high incidence of tree mortality (H) and low incidence of tree mortality (L).**



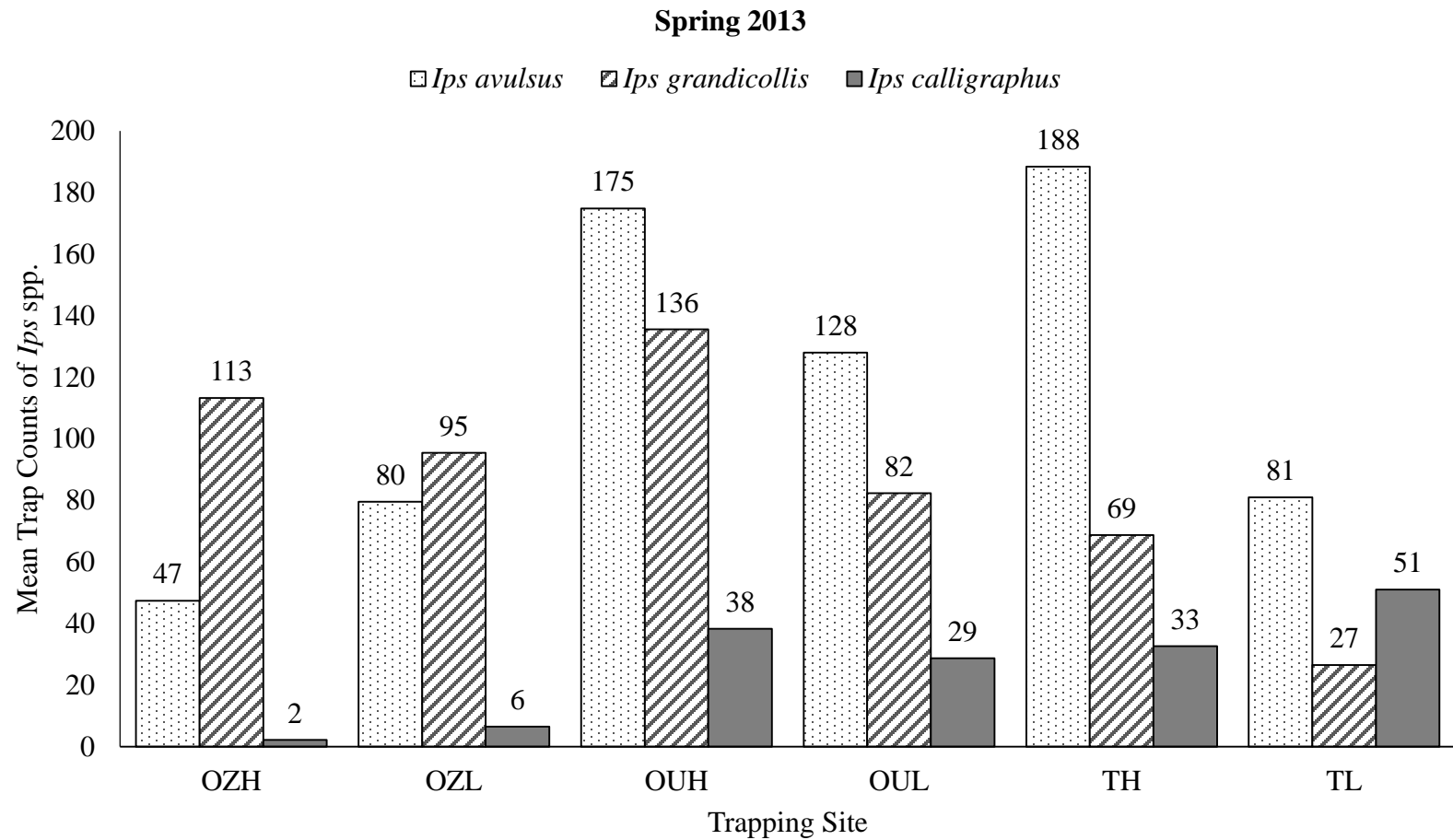
**Figure 5: Mean trap counts (n=15) of *Ips* spp. in summer 2012 (July) for locations in Ozark National Forest (OZH and OZL), Ouachita National Forest (OUH and OUL), and Eastern Texas (Sam Houston [TH] and Davy Crockett [TL] National Forest). The last letter in the location ID makes the distinction between high incidence of tree mortality (H) and low incidence of tree mortality (L).**



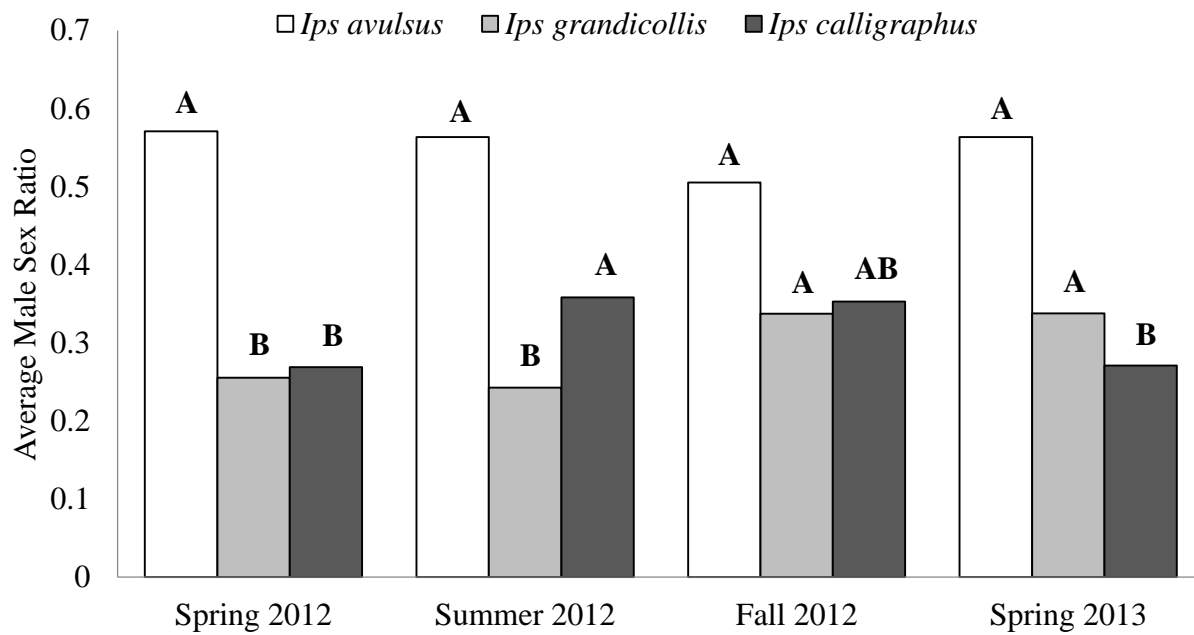
**Figure 6: Mean trap counts (n=15) of *Ips* spp. in fall 2012 (October) for locations in Ouachita National Forest (OUH and OUL), Ozark National Forest (OZH and OZL), and Eastern Texas (Sam Houston [TH] and Davy Crockett [TL] National Forest). The last letter in the location ID makes the distinction between high incidence of tree mortality (H) and low incidence of tree mortality (L).**



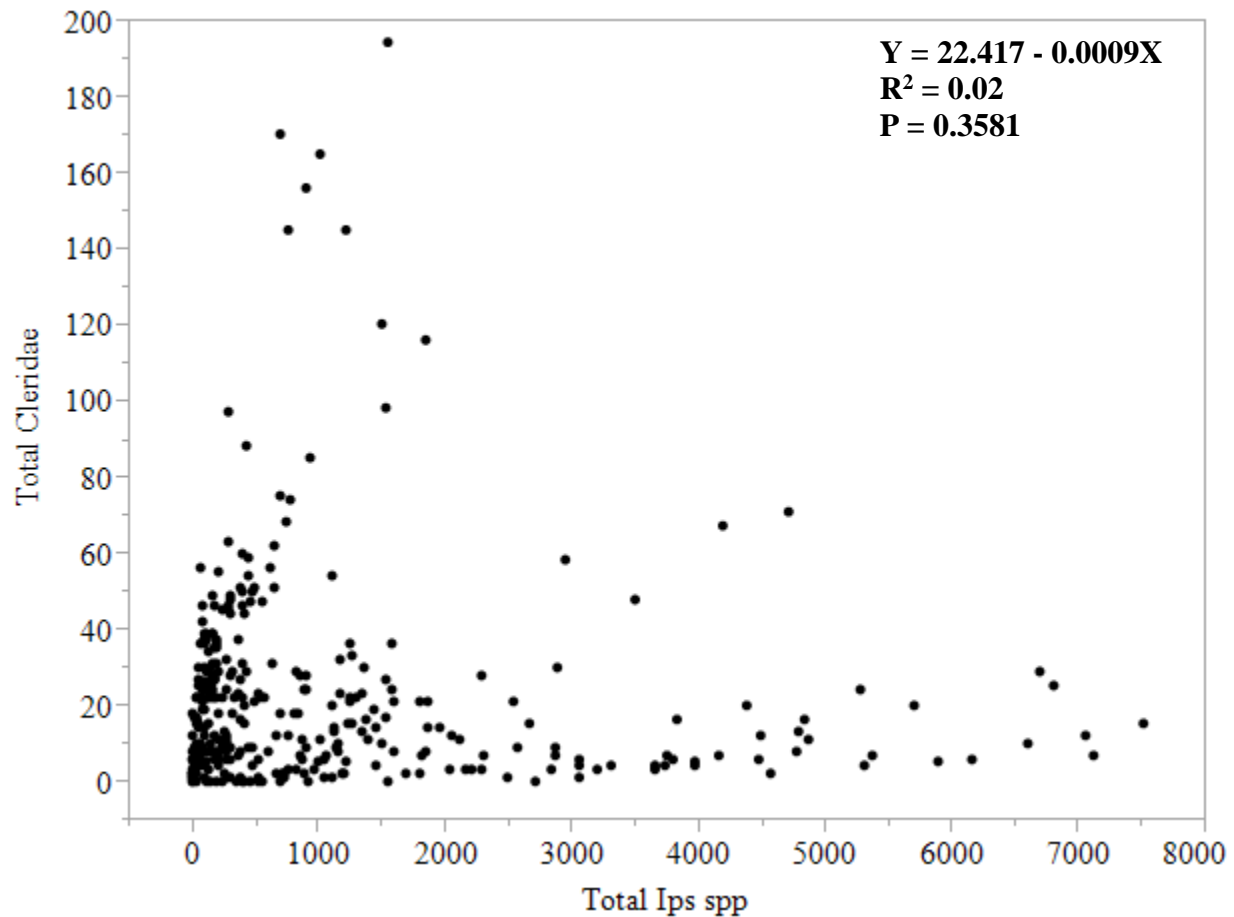
**Figure 7: Mean trap counts (n=15) of *Ips* spp. in spring 2013 (March) for locations in Ozark National Forest (OZH and OZL), Ouachita National Forest (OUH and OUL), and Eastern Texas (Sam Houston [TH] and Davy Crockett [TL] National Forest). The last letter in the location ID distinguishes high incidence of tree mortality (H) and low incidence of tree mortality (L).**



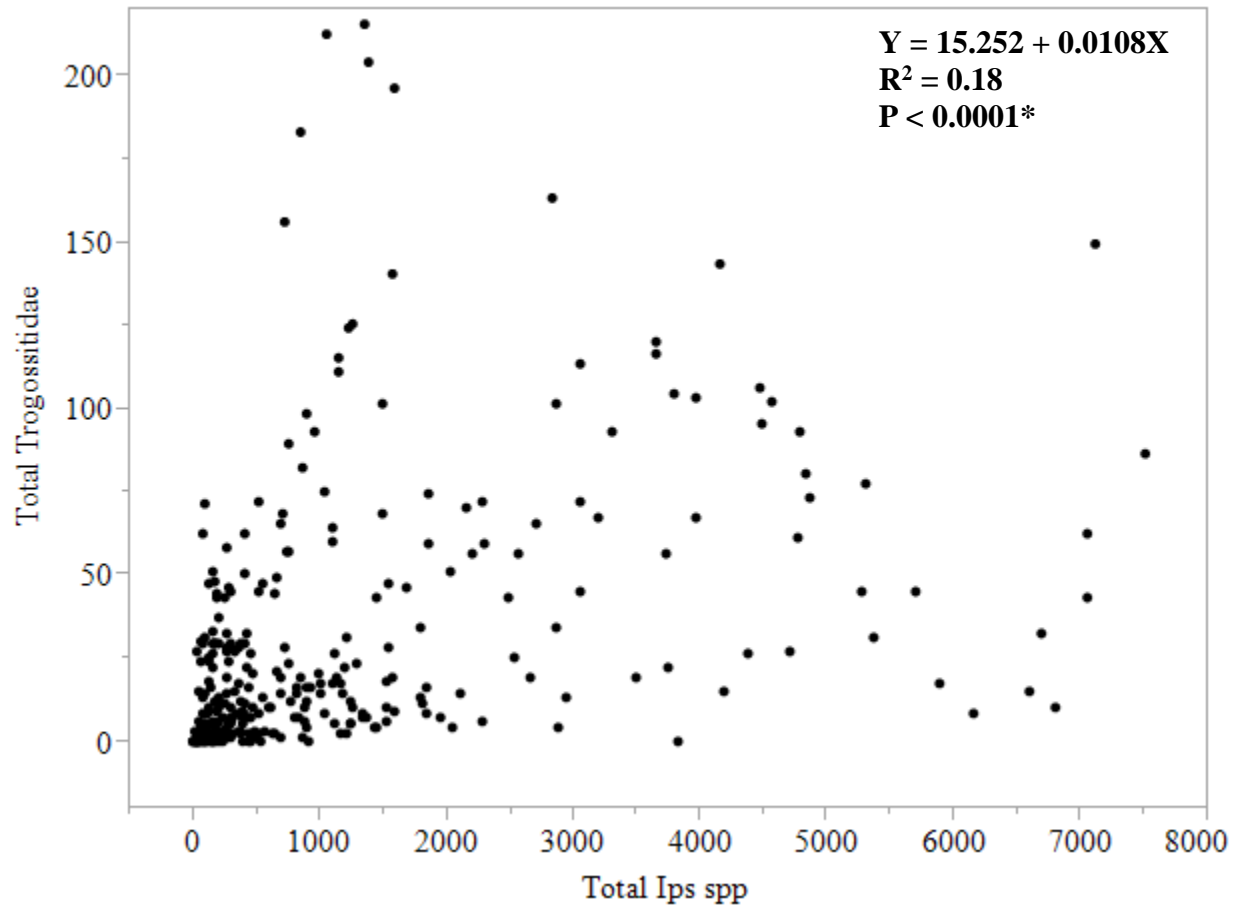
**Figure 8: Average male sex ratio for each species of *Ips* organized by trapping season. Considering each species separately, mean ratios with the same letter are not significantly different ( $P = 0.05$ , Tukey's HSD).**



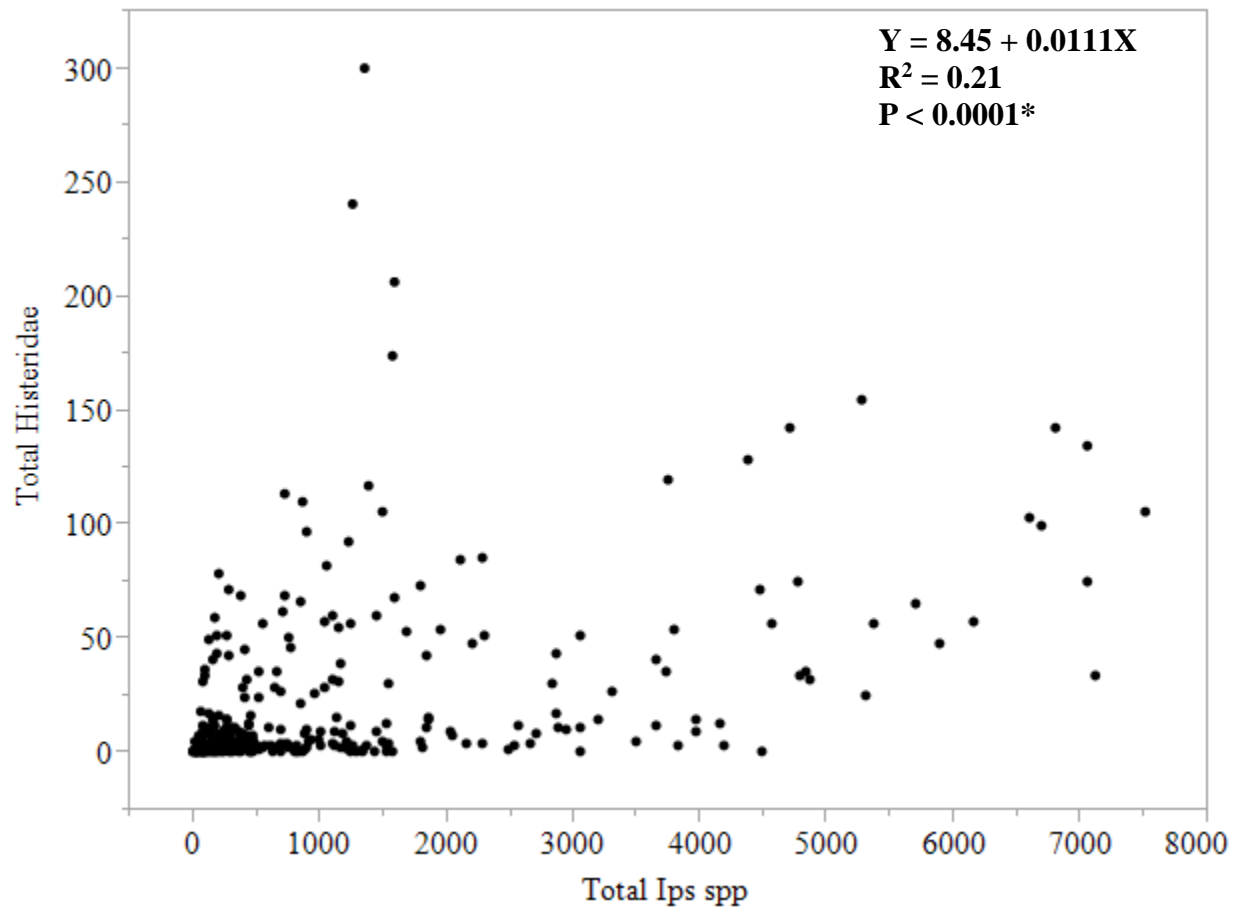
**Figure 9: Correlation between total *Ips* trap catches and Cleridae (n = 352). Each data point represents the sum of all *Ips* adults and all Clerid adults collected in a single Lindgren funnel trap on a single collection date. Trapping spanned four collection periods: spring 2012, summer 2012, fall 2012, and spring 2013.**



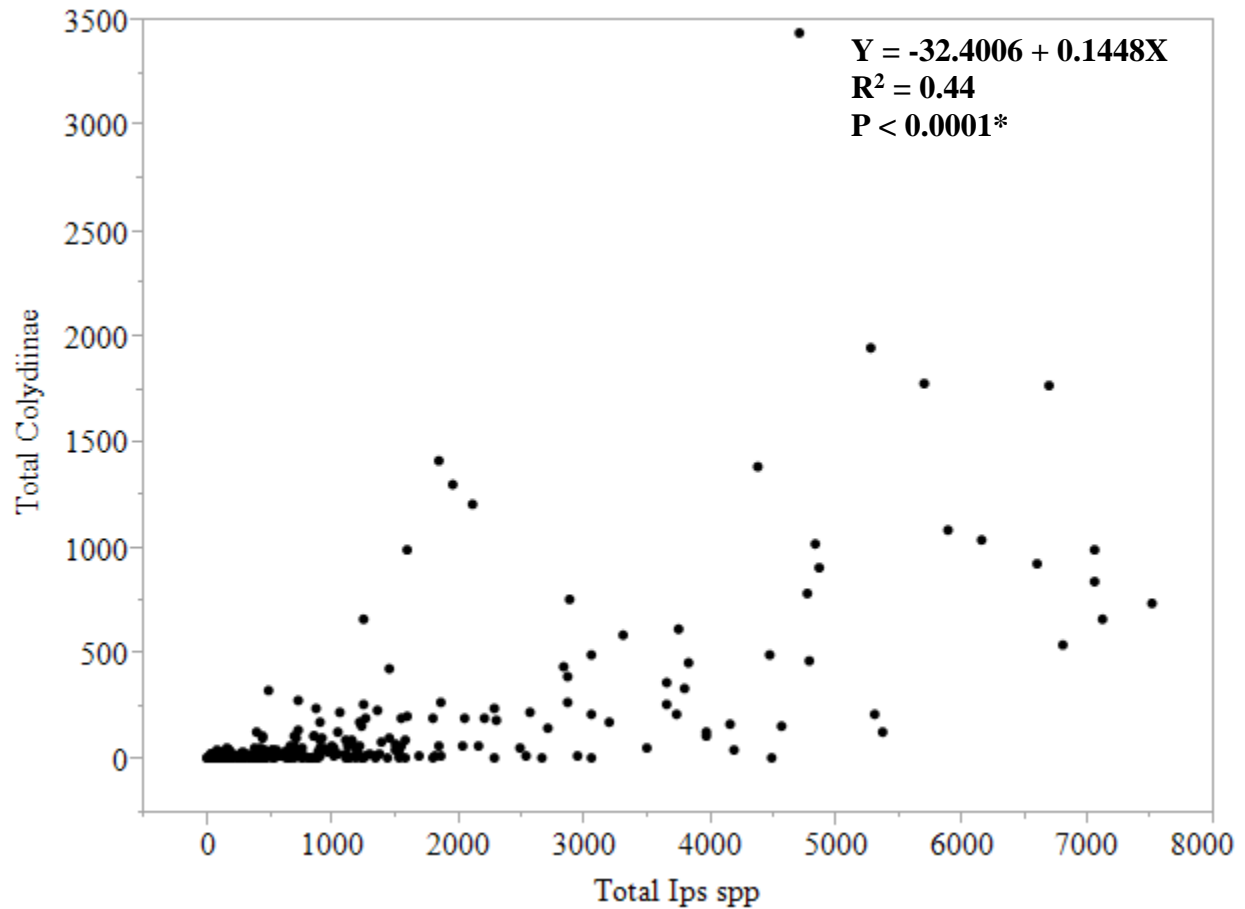
**Figure 10: Correlation between total *Ips* trap catches and Trogossitidae (n = 352). Each data point represents the sum of all *Ips* adults and all Trogossitidae adults collected in a single Lindgren funnel trap on a single collection date. Trapping spanned four collection periods: spring 2012, summer 2012, fall 2012, and spring 2013.**



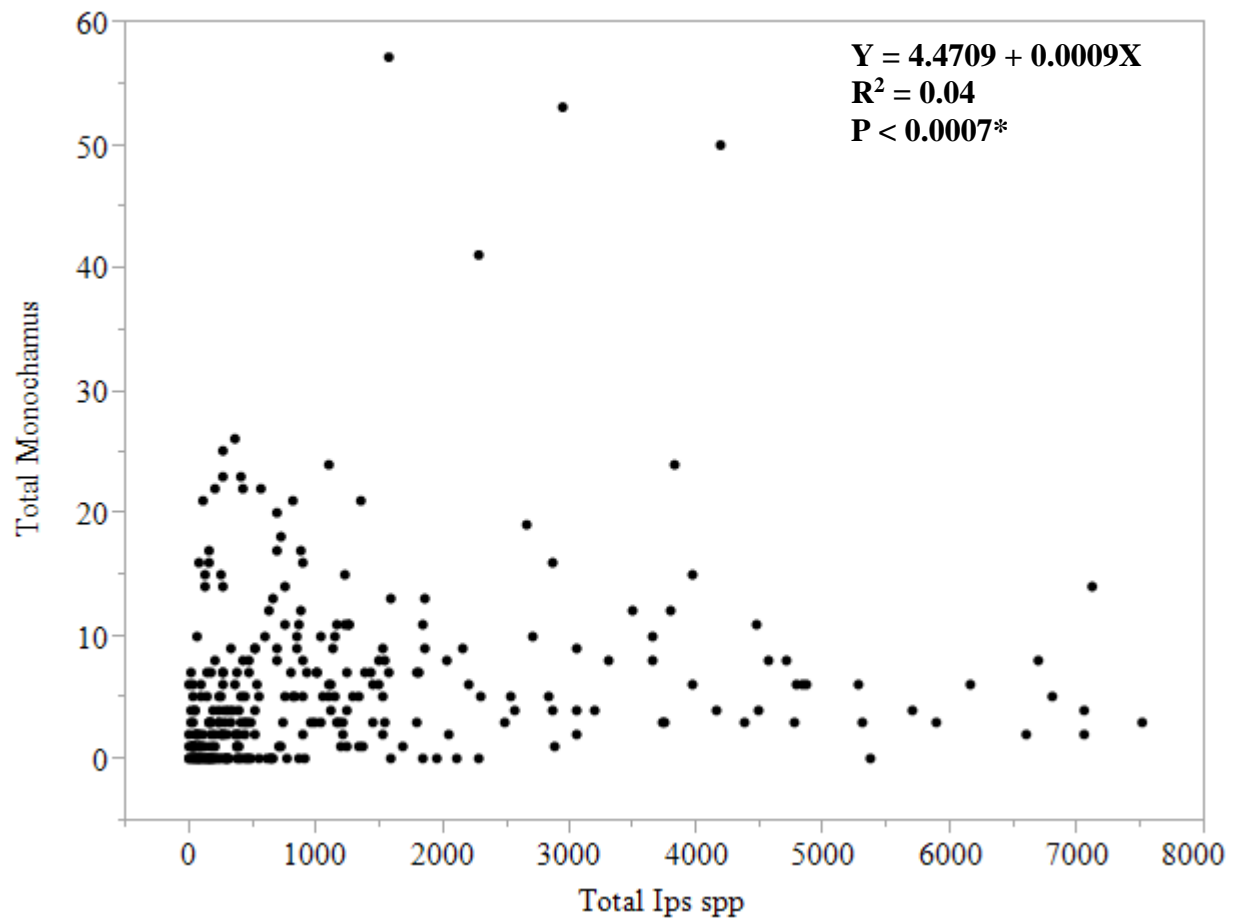
**Figure 11: Correlation between total *Ips* trap catch and total Histeridae trap catch (n = 352). Each data point represents the sum of all *Ips* adults and all Histeridae adults collected in a single Lindgren funnel trap on a single collection date. Trapping spanned four collection periods: spring 2012, summer 2012, fall 2012, and spring 2013.**



**Figure 12: Correlation between total *Ips* trap catch and total Colydiinae (Zopheridae) trap catch (n = 352). Most of the Colydiinae species were in the genus *Lasconotus*. Each data point represents the sum of all *Ips* adults and all Colydiinae adults collected in a single Lindgren funnel trap on a single collection date. Trapping spanned four collection periods: spring 2012, summer 2012, fall 2012, and spring 2013.**



**Figure 13: Correlation between total *Ips* trap catch and total *Monochamus* spp. trap catch (n = 262). The spring 2013 trapping season was excluded from this correlation due to the fact that *Monochamus* spp. did not emerge until after this trapping season. Each data point represents the sum of all *Ips* adults and all *Monochamus* adults collected in a single Lindgren funnel trap on a single collection date. Trapping spanned four collection periods: spring 2012, summer 2012, fall 2012, and spring 2013.**



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## CHAPTER 3 – COMPARING *IPS* TRAP CATCHES TO TREE MORTALITY SURVEYS

### INTRODUCTION

*Ips avulsus* (Eichhoff), *Ips grandicollis* (Eichhoff), and *Ips calligraphus* (Germar) contribute to widespread pine mortality in the southeastern United States. However, the scattered nature of their outbreaks makes control and salvage difficult, and records of their impacts are not maintained (Clarke et al. 2000). Several researchers have suggested that unreported *Ips* are responsible for more pine mortality than southern pine beetle (*Dendroctonus frontalis* Zimmerman), which is recognized as the most destructive pine insect of southern pines (Beal and Massey 1945, Thatcher 1960, Clarke et al. 2000). Despite their common influence in southern pine forests, there are no accepted methods for monitoring *Ips* populations and their impacts. In order to advance our understanding of the effects of *Ips* on pine stands, this study was designed to compare *Ips* pheromone trap catches to tree mortality within stands.

*Ips* pine engravers are present in all southern pine forests and their outbreaks may occur when area-wide conditions weaken trees. Trees generally are capable of defending against bark beetle attacks, and outbreaks are considered a symptom of poor stand health (Schowalter 2012). *Ips* pine engravers attack success is a direct result of reduced tree vigor. *Ips* typically reproduce in fallen or dying trees (Wilkinson and Foltz 1982, Bryant *et al.* 2006). However, they are capable of attacking living trees with the help of one or several exogenous disturbance agents (e.g. drought, fire, wind damage, severe storms, and physical damage by harvesting practices) (Wilkinson and Foltz 1982, Coulson *et al.* 1999, Clarke *et al.* 2000, Bryant *et al.* 2006, Schowalter 2012). In particular, droughts frequently precede *Ips* and other bark beetle outbreaks

(Mattson and Haack 1987, Christiansen and Bakke 1997, Pase III 2011, White 2014). Some biotic factors that increase susceptibility to bark beetle attack are vegetative competition, fungal pathogens, and enhanced phloem nutrition (Wilkinson and Foltz 1982, Eckhardt 2003, Fettig et al. 2007, White 2014).

As many disturbance agents are unseen, it is difficult to predict when *Ips* pine engraver outbreaks will occur. Landowners often see the aftermath of an outbreak, and have little or no time to react to the associated timber loss. Monitoring with semiochemical-baited traps may aid in alerting landowners of changes in bark beetle abundance. Similar trapping programs have successfully revealed regional population trends of southern pine beetle (SPB) (Billings 1988). A method for forecasting SPB infestations was developed that considers trap catches of *D. frontalis* and its predator, *Thanasimus dubius* (F.) (Billings 1988). In contrast, few studies have monitored the regional or local population dynamics of *Ips* pine engraver beetles of the southeastern United States.

The major goal of this study was to investigate the relationship of tree mortality to *Ips* abundance as determined by pheromone traps. In recent years, researchers have attempted to correlate bark beetle trapping numbers to abundance of beetle-killed trees. In Sweden, catches of *Ips acuminatus* (Gyllenhal) correlated strongly with numbers of infested trees (Faccoli et al. 2012). Also in Europe, the correlation of *Ips typographus* (L.) trap catches and tree mortality was investigated, and predictive models were developed to relate *I. typographus* trap catch to annual damage (Faccoli and Stergulc 2004). In the case of western pine beetle, *Dendroctonus brevicomis* LeConte, beetle-killed trees and daily trap catches of *D. brevicomis* were significantly correlated, but stand characteristics were a better indicator of tree mortality (Hayes et al. 2009).

Aerial surveying is frequently used for measuring the extent of bark beetle infestations. Many outbreaks of *Dendroctonus* spp. are discovered either through aerial detection flights or remote sensing. In the case of *D. frontalis*, digital aerial sketchmapping allows a surveyor in a plane to digitize outbreaks using onboard geographic information system (GIS) software (Steiner 2011). However, sketchmapping has not been implemented specifically for *Ips* in the southeastern U.S. The scattered distribution of those infestations makes estimation of most outbreaks unreliable by aerial surveys.

Foresters frequently estimate *Ips* damage by simply viewing trees from the adjacent road, but observed damage has not been related to the actual mortality in the stand. Furthermore, it is unknown if land managers can learn anything about bark beetle populations by observing damage at the margins of the stand. Sampling at stand edges, if proven useful, could offer a way to rapidly assess *Ips* pine engraver infestations. A primary concern with observing stand edges is that the survey may not translate to the interior portions of the stand. Microclimate and host susceptibility may be different in stand edges, and evidence suggests that bark beetle densities vary between the interior and edge portions of a stand (Peltonen *et al.* 1997). Roadside sampling techniques are useful for *Ips sexdentatus* infestations (Samalens *et al.* 2007). In the case of *I. sexdentatus*, the percentage of attacked trees on the edge does not differ from the percentage of the interior of the stand, and thus damage in stand edges can be used to estimate the mean percentage of attacked trees per stand. Also, counting trees within a 10-meter sighting distance from the road effectively estimates the percentage of attacked trees in the stand. Bias associated with maximum sighting distance, which varies depending on visibility and topography, is removed with a constant sighting distance.

The most thorough method for assessing tree mortality is a ground-based survey. However, physically investigating stand damage can be tedious and time-consuming. Many land managers simply do not have the time to enter a stand and record damaged trees. A ground-based survey may be used to verify the faster, more efficient survey methods like roadside or aerial surveying. In Europe, an “exhaustive ground sampling” technique determines the condition of all dead and infested spruce trees (Samalens *et al.* 2007). This ground-based survey allows a comparison of the estimation of damaged trees reported by the road surveys with actual totals of tree mortality.

In this chapter, we analyzed the results from vehicle-based road surveys and ground-surveys of tree mortality. Also, we compared *Ips* trap catches to estimates of tree mortality. The following questions were addressed. 1) Can a quick observation of the incidence of tree mortality provide any information about the abundance of *Ips*? 2) How useful are vehicle-based road surveys? 3) Does the number of dead or infested trees correlate with the abundance of *Ips*?

## **METHODS**

Trapping and tree mortality surveys were conducted in three regions, which include the Ozark National Forest in Arkansas, the Ouachita National Forest in Arkansas, and east Texas (Sam Houston and Davy Crocket National Forests) (see Chapter 2, Figure 1). Within each of these regions, two sites were chosen, a ‘high incidence’ stand and a ‘low incidence’ stand. These designations were made subjectively prior to implementing the study, and they suggest the level of dead and infested trees within the stand. In a high incidence site, tree mortality appeared more abundant than in the nearby low incidence stand. Due to regional differences, a high incidence

stand in one region may not necessarily relate to a high incidence stand in another, and likewise for low incidence stands.

Each study site was located within a relatively homogenous pine stand that was even-aged in character. All sites were intersected by a drivable USDA Forest Service road. The stand was at least one km in length with greater than 100 meters of pine forest extending away from both sides of the road. Sites were selected that had similar stand structures (i.e. pine dominance, sawtimber-sized diameter, and roughly comparable pine basal area per hectare) (see Chapter 2, Table 1).

Three methods were implemented to monitor the *Ips* infestations within stands. A pheromone trapping system collected flying insects and two mortality surveys were used to quantify the amount of dead and infested trees. The first was a vehicle-based road survey and the second was a ground-based 100% tally of tree mortality.

The pheromone trapping system, described in detail in Chapter 2, used *Ips*-specific pheromones to target all three *Ips* species. Five, 12-unit Lindgren® funnel traps were used within each stand. Trap contents were collected weekly during a three-week trapping period. This was repeated in four seasonally based trapping periods: spring 2012, summer 2012, fall 2012, and spring 2013.

The vehicle-based road survey was a rapid visual estimate of tree death and damage. This was accomplished by tallying trees while driving 10 to 15 kilometers/hour along a one-kilometer segment of the road that intersected the stand. The passenger observed trees on one side of the road and continued the tally on the other side when the vehicle made a return pass. Trees were tallied in two categories: dead trees with grey foliage or no needles and fading trees with symptoms of attack. Foliage discoloration, associated with partial or total crown dieback, was

regarded as a symptom of reduced tree vigor and subsequent *Ips* attack. This category included trees with flagging and top-killed crowns. All trees that could be seen from the road were recorded; therefore, the maximum sighting distance varied slightly between stands as the stand density was different at each location. To lessen surveyor bias, a second passenger also performed the survey and the average of the two surveyors was recorded. This survey was completed in spring 2012, fall 2012, and spring 2013.

The ground-based 100% tally of tree mortality was accomplished by an experienced forestry field crew. The survey took place throughout a forest extending 100 meters on both sides of the one kilometer road, resulting in a total survey area of 20 hectares. Every standing dead or fading tree was recorded in the study area. Fading trees were any trees with visible crown discoloration or dieback, symptoms of stress and bark beetle attack. When the color change in the crown only affected one or a few branches, this symptom was termed ‘flagging’. Flagging was a symptom that typically appeared in the initial stages of crown dieback. On some trees, the whole crown faded simultaneously. Once a fading tree was located, it was examined for signs of *Ips* attack (i.e. presence of pitch tubes or boring dust and galleries under the bark). The bark and phloem of each tree was also examined. The absence of living phloem indicated that the tree was dead. Each living tree with still fresh phloem was recorded by GPS for revisits in future surveys. A Garmin™ GPSmap 60CSx, rated as accurate to within 10 meters, was used to record coordinates for living trees. The following information was recorded for all trees: tree species, diameter at breast height (DBH), crown class, crown condition (i.e. color and appearance of foliage), and bole condition (Table 1). Except for DBH, all data recorded were categorical or nominal. The ground-based survey was repeated spring 2012, fall 2012, and spring 2013.

## Statistical analyses

All analyses were completed with JMP Pro 10 (SAS Institute Inc., Carey, NC). The significance level for all analyses was set at  $\alpha = 0.05$ .

The first analysis was designed to determine whether a subjective assessment of stand-level tree mortality was correlated to *Ips* abundance in the stand. Trap catches from the high and low incidence stands were compared. A two-tailed t-test was used to test the null hypothesis that mean trap catches between high and low incidence stands were not different. Only the spring 2012 and spring 2013 seasons were analyzed; mean trap catches in fall 2012 were too low in the Arkansas sites making them impractical for this analysis. There were a total of six t-tests to compare means.

The estimation accuracy of the road survey was compared to the actual number of dead and symptomatic trees recorded during the ground-based survey. This was accomplished with a simple linear regression, in which the predictive value of the road survey was assessed. For this regression, the combined number of dead and symptomatic trees was used for each survey type.

The relationship between the number of dead or fading trees and the number of *Ips* captured was investigated. More specifically, the correlation of tree mortality to the mean number of *Ips* of each species caught was examined. The *Ips* trap catch was also compared to the number of dead trees and the number of fading trees separately. All three *Ips* species were graphed simultaneously for each survey period, so that differences among species could be noted.

## RESULTS

The total numbers of dead and fading trees recorded in the vehicle-based survey found at each site during the spring of 2012 and 2013 are presented in Figure 1. The east Texas high incidence site had the greatest amount of tree mortality recorded by the vehicle-based survey in both spring of 2012 and 2013 with 111.5 and 96 trees respectively. The Ouachita Mountains low incidence site had the lowest amount in both years with 17 trees in 2012 and 14 trees in 2013. The number of fading trees was very low in spring 2013 for all locations.

The total numbers of dead and fading trees recorded in the ground-based survey are similar to the vehicle-based survey (Figure 2). The percentage of fading trees recorded in ground surveys was much lower in spring 2013 than in spring 2012 (Table 2). The percent of fading trees recorded in the tally in spring 2012 was an average of 25.1% for all sites, but in spring 2013 the percent of fading trees decreased to an average of 8.3%. In the 2013 survey, the percentage of trees that were in the suppressed crown class category increased in all sites except the east Texas high incidence stand.

Based on total trap catches, *Ips* abundance appeared to decline between spring 2012 and spring 2013 (Table 3). With the exception of east Texas, the trap catches in fall of 2012 were the lowest. *Ips grandicollis* was the most abundant species in this study and *Ips calligraphus* was the least abundant.

Trap catches of *Ips* species in the East Texas region were significantly greater in the high incidence site compared to the low incidence site in both the springs of 2012 and 2013 ( $P < 0.0001$ , Figure 3). This was also true for the two types of sites in the Ouachita Mountain regions in 2012 ( $P < 0.0002$ ). No significant differences were detected between high and low sites

in the Ouachita Mountains in 2013 ( $P < 0.1468$ ), nor were differences detected among the two site types in the Ozark Mountain in either year (Figure 3).

The total numbers of fading and dead trees estimated from the vehicle-based survey was significantly correlated with total trees determined by the ground-based survey ( $R^2 = 80.4$ ,  $P < 0.0001$ , Figure 4). The vehicle-based survey recorded approximately half of the total trees recorded in the ground-based survey, but the derived linear model indicates that total dead and fading trees could be rather accurately predicted from a road-based survey.

The total number of dead trees was correlated strongly with mean trap catches of each *Ips* species in the spring 2012 trapping season (Figure 5). The relationship in the fall 2012 trapping season was also significantly correlated. In the spring 2013 trapping season, when *Ips* abundance was lower than the previous year, the relationships between total number of dead trees and mean *Ips* trap catches were not significant.

The total number of fading (infested) trees was not correlated with most mean trap catches of each *Ips* species (Figure 6). Only the relationship between *Ips calligraphus* trap catches and the total number of fading trees in spring 2013 was significant ( $R^2 = 0.80$ ,  $P < 0.0157$ ).

## DISCUSSION

Information recorded in the ground-based survey in spring 2012 clearly showed that an *Ips* outbreak was taking place. However, *Ips* trap data suggest that abundance declined over the course of the study (Table 3), and in the spring 2013, there were very few faded and infested trees present (Table 2). The increase in the percentage of standing dead trees and the decline of the percent fading trees support the suggestion that the outbreak had subsided. Fading trees with crown conditions of “1” through “3” were more frequent in 2012 than in 2013. These crown

conditions could indicate that a stand is actively declining. Despite the lack of actively fading trees, some stands did still have trees with ‘flagging’ in spring of 2013. We have no evidence to suggest that the flagging symptom provides any indication of outbreak or high *Ips* abundance. Further studies could benefit from determining if ‘flagging’ is a symptom related to the early stages of bark beetle attack.

Another interesting observation from the results relates to the percentage of *Ips* infested trees classified as suppressed (Table 2). The total number of trees recorded by the ground-based survey did not appear to change much, but the percentage of suppressed trees increased in most stands over the course of the study. This may suggest that as hosts in the canopy become less available, *Ips* rely on weakened suppressed trees to sustain populations when an outbreak is not occurring.

For this study, two locations were selected in each region such that one stand had high tree mortality (high incidence) and the other featured less visible mortality (low incidence). High incidence stands characteristically had *Ips*-associated mortality seen from the road. The low incidence stand was thought to be healthier than the high incidence stand. Before the mortality surveys were completed, it was not known if this was an accurate description of the stands. Both the vehicle-based and ground-based surveys revealed that more total tree mortality had occurred in the high incidence stand of each region compared to that measured in the low incidence stand (Table 2, Figures 1 and 2). These results indicate that quick assessments from the road adjacent to the stand can provide some insight to the health of the stand.

Despite clear differences in tree mortality, the trap catches between high and low incidence sites were not always significantly different. During the spring 2012 trapping season, trap catches from the high incidence site were significantly greater in two of the three

comparisons (Figure 3). However, the trap catches between the two Ozark Mountain stands were not significantly different, although the ground-based survey revealed that the low incidence stand had slightly less mortality than the high incidence stand in that region. In the spring 2013 season, only the east Texas high incidence site was significantly greater than its counterpart ( $P = 0.0001^*$ ). In many cases the number of *Ips* captured varied greatly between collection dates of the same location. For example, in spring 2012 in the Ozark Mountain high incidence stand, several trap catches were well above the range of trap catches in the low site while some were much lower. Extreme weekly variation may be attributed to weather conditions that prevent bark beetle dispersal.

Vehicle-based road surveys do provide a good predictive model of actual tree mortality, provided that there is adequate visibility throughout the stand. All stands in this study featured low density pine of sawtimber sizes. The usefulness of road surveys may diminish in younger, denser stands. Another caveat is the fact that recording a fading tree does not necessarily mean that the tree is colonized by *Ips* bark beetles. When investigating each tree during the ground-based survey, it became apparent that some trees with the flagging symptoms recovered without any infestation. Alternatively, some red-crowned trees did not have any fresh phloem and no longer hosted bark beetle brood, so they were classified as dead.

The total number of fading and standing dead trees was positively correlated with the mean *Ips* catches in spring and fall of 2012 (Figure 5). However, in spring 2013, these correlations no longer existed as *Ips* trap catches were much lower than the previous year and the number of standing dead trees did not change much. Interestingly, the number of fading trees alone did not correlate with the mean *Ips* catches (Figure 6). A significant relationship only occurred in one relationship, the *Ips calligraphus* catches in spring of 2012, which, counter

intuitively, showed an inverse relationship between the mean *Ips* catch and the number of fading trees alone. The number of standing dead and fading trees may provide some insight to the abundance of *Ips*, but only when the outbreak is active (i.e. trees are actively fading throughout the stand).

## CONCLUSION

This study was able to track the decline of *Ips* abundance following a period of visible outbreak. As such, it was difficult to evaluate the predictive value of trap catches in relation to tree mortality surveys. Pheromone trapping may be able to alert landowners to rising *Ips* densities; however, this study provided little evidence to suggest this as *Ips* trap catches only correlated with dead and fading trees in one year of the study. Additional years of data collection would be necessary to evaluate if detection of rising *Ips* densities is possible with monitoring. From our results, it appears that the spring season is best for monitoring *Ips* pine engravers. We observed actively fading trees more frequently in the spring, and *Ips* dispersion appears greater in spring than in fall. The strongest relationships between *Ips* trap catches and tree mortality were found in spring of 2012 when the outbreak appeared to be still active. Future studies can benefit from short periods of trapping during the early portion of the spring season.

## TABLES

**Table 1: Description of information recorded during the ground-based survey.**

<b>Data Type</b>	<b>Categorical Data</b>	<b>Description</b>
Tree Species	Loblolly	Tree identified as <i>Pinus taeda</i>
	Shortleaf	Tree identified as <i>Pinus echinata</i>
DBH		Tree diameter (cm) measured at 1.37 meters (4.5 feet) above the ground
Crown Class	D	Tree dominant in canopy
	CD	Tree codominant in canopy
	I	Tree intermediate in canopy
	S	Tree suppressed within canopy
Crown Condition	1	Foliage with minor needle discoloration (i.e. yellow or red color caused by disruption of phloem), mostly green needles. Flagging symptoms. Tree may recover.
	2	Tree featured a high degree of needle discoloration, but some green needles remained. Tree will not recover.
	3	No green needles. Crown may have red or grey needles or a combination of both.
	4	No needles in crown. Typically, the tree is dead.
Bole Condition	1	Fresh phloem and no attacks in lower bole.
	2	Attacks visible in lower bole. Species of insects are recorded.
	3	No phloem present. Tree is dead.
Damaging Factors		Any conditions that may have contributed to the tree's injury or death are noted.
<i>Ips</i> species present		If the bole condition is a 2, then the <i>Ips</i> present in the phloem are recorded.
GPS coordinates		If the bole condition was a 1 or 2, then the GPS coordinates were recorded.

**Table 2: Summary results of ground-based 100% tally of tree mortality in spring 2012 and spring 2013. The column labeled “Live Trees per Ha.” provides an estimate derived from technical forest mensuration. Percentage data represents the percentage of the total trees recorded that meet the column’s description.**

<b>Year</b>	<b>Region</b>	<b>Subjective Tree Mortality</b>	<b>Total Trees Recorded</b>	<b>Dead or Fading per Ha.</b>	<b>Live Trees per Ha.</b>	<b>Dead Trees</b>	<b>Fading Trees*</b>
2012	East Texas	Low	121	6.05	83	71.1%	28.9%
		High	245	12.25	172	86.1%	13.9%
	Ouachita Mountains	Low	35	1.75	185	65.7%	34.3%
		High	126	6.30	152	85.7%	14.3%
	Ozark Mountains	Low	60	3.00	163	71.7%	28.3%
		High	80	4.00	203	70.0%	30.0%
2013	East Texas	Low	139	6.95	83	96.4%	3.6%
		High	196	9.80	172	98.5%	1.5%
	Ouachita Mountains	Low	30	1.50	185	86.7%	13.3%
		High	100	5.00	152	97.0%	3.0%
	Ozark Mountains	Low	49	2.45	163	87.8%	12.2%
		High	69	3.45	203	84.1%	15.9%

\* Fading trees are those with needle discoloration and fresh phloem.

**Table 2 (cont.): Summary results of ground-based 100% tally of tree mortality in spring 2012 and spring 2013. Percentage data represents the percentage of the total trees recorded that meet the column's description.**

Year	Region	Subjective Tree Mortality	Total Trees Recorded	Crown Condition				Canopy Trees**	Suppressed Trees
				1	2	3	4		
2012	East Texas	Low	121	3.3%	16.5%	24.0%	56.2%	81.8%	18.2%
		High	245	3.7%	7.3%	13.1%	75.9%	98.0%	2.0%
	Ouachita Mountains	Low	35	22.9%	2.9%	20.0%	54.3%	94.3%	5.7%
		High	126	0.8%	2.4%	11.9%	84.9%	82.5%	17.5%
	Ozark Mountains	Low	60	6.7%	6.7%	26.7%	60.0%	66.7%	33.3%
2013	East Texas	Low	139	1.4%	0.7%	0.0%	97.8%	78.4%	21.6%
		High	196	1.0%	0.5%	0.0%	98.5%	98.5%	1.5%
	Ouachita Mountains	Low	30	13.3%	0.0%	0.0%	86.7%	86.7%	13.3%
		High	100	2.0%	0.0%	3.0%	95.0%	74.0%	26.0%
	Ozark Mountains	Low	49	6.1%	4.1%	4.1%	85.7%	51.0%	49.0%
		High	69	13.0%	0.0%	1.4%	85.5%	73.9%	26.1%

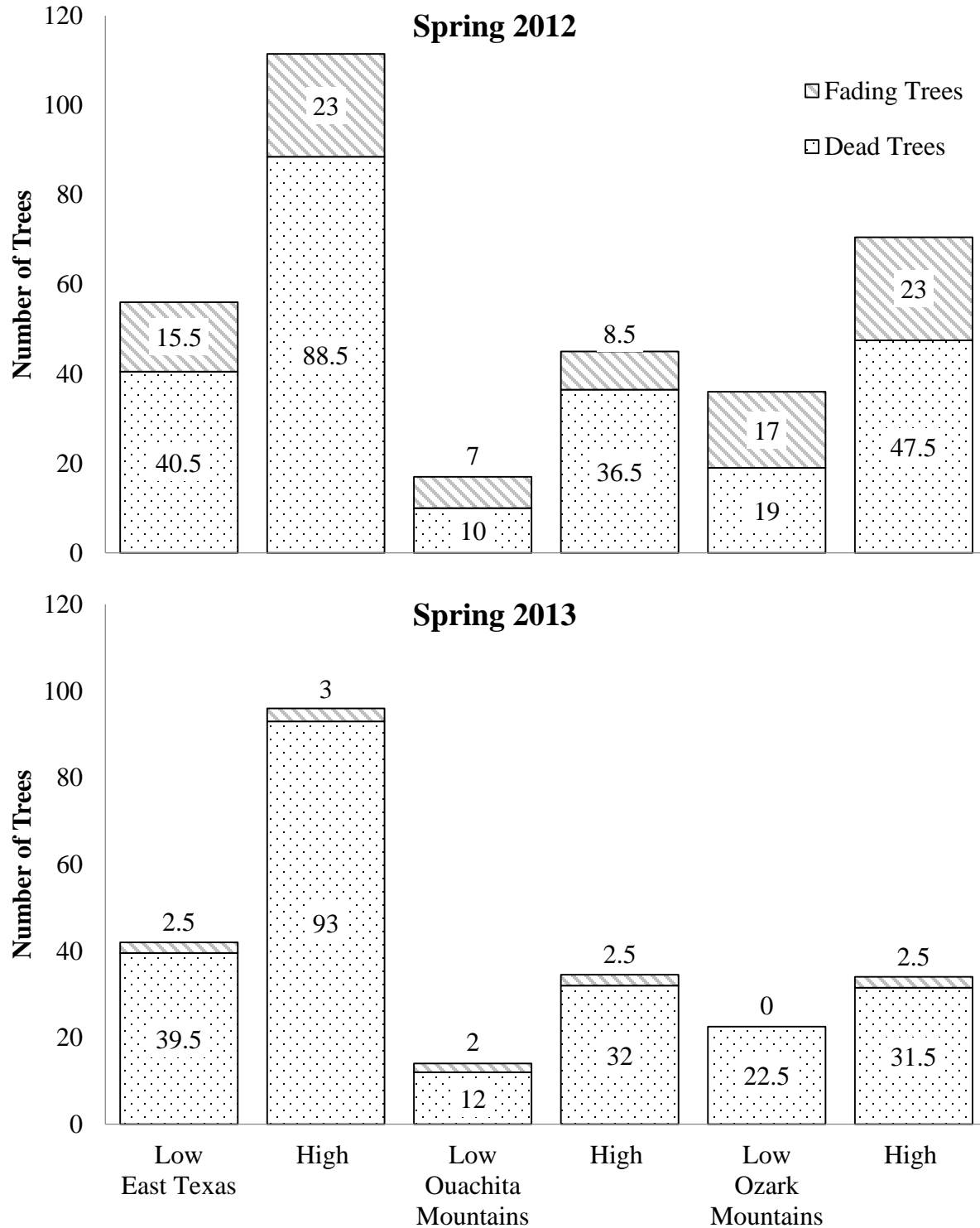
\*\* Represents a combination of trees with dominant, codominant, and intermediate crown classes.

Table 3: Total trap catches of *Ips* species (each value is a sum of 15 traps; five traps over three weekly collections), from four seasonal trapping periods and three regional locations. High (H) and low (L) describe the general level (incidence) of tree mortality that was estimated before trapping and surveys were initiated in each of the locations. Abbreviations for regions are as follows: OZ is Ozark National Forest, OU is Ouachita National Forest, and ET is eastern Texas (Sam Houston and Davy Crockett National Forests).

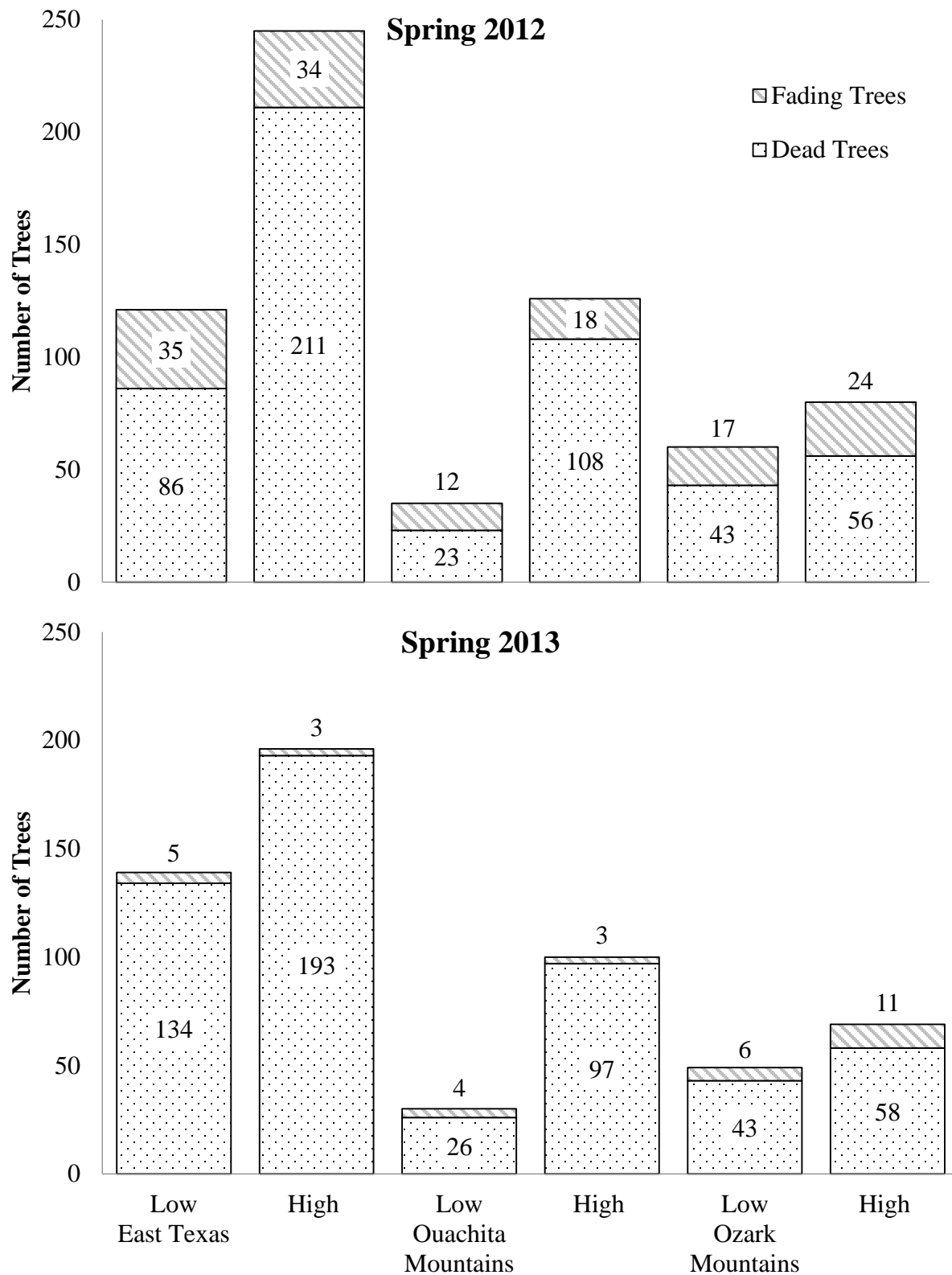
		Spring 2012			Summer 2012			Fall 2012			Spring 2013			Total
		OZ	OU	ET	OZ	OU	ET	OZ	OU	ET	OZ	OU	ET	
<i>Ips avulsus</i>	H	4383	13205	28795	3371	12948	4035	231	614	12665	711	2622	2842	86422
	L	2804	3760	7955	8574	6362	4153	243	110	3292	1193	1919	1036	41401
<i>Ips grandicollis</i>	H	6235	14396	30119	22054	47831	4728	170	484	658	1698	2033	891	131297
	L	5152	5035	7009	38348	24923	2467	200	152	143	1432	1234	244	86339
<i>Ips calligraphus</i>	H	29	160	5801	108	871	8803	10	178	1250	32	574	442	18258
	L	34	45	1021	78	319	4196	24	64	1020	97	431	703	8032

## FIGURES

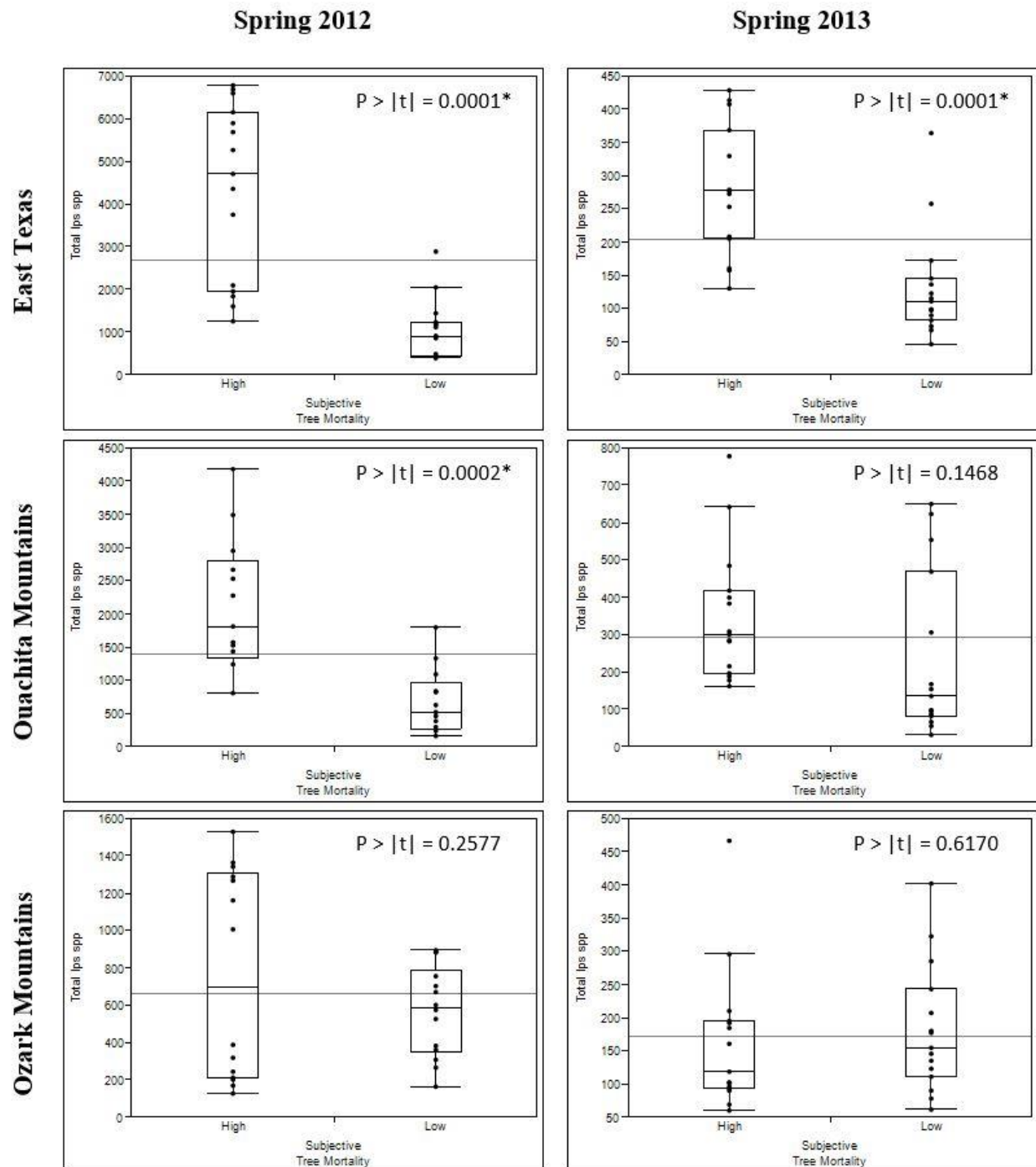
**Figure 1: Number of dead and fading trees at each location estimated by vehicle-based road survey in spring 2012 and spring 2013. Numbers are an average of two surveyors. The number is the total tree tally from both sides of the 1-kilometer segment of the road.**



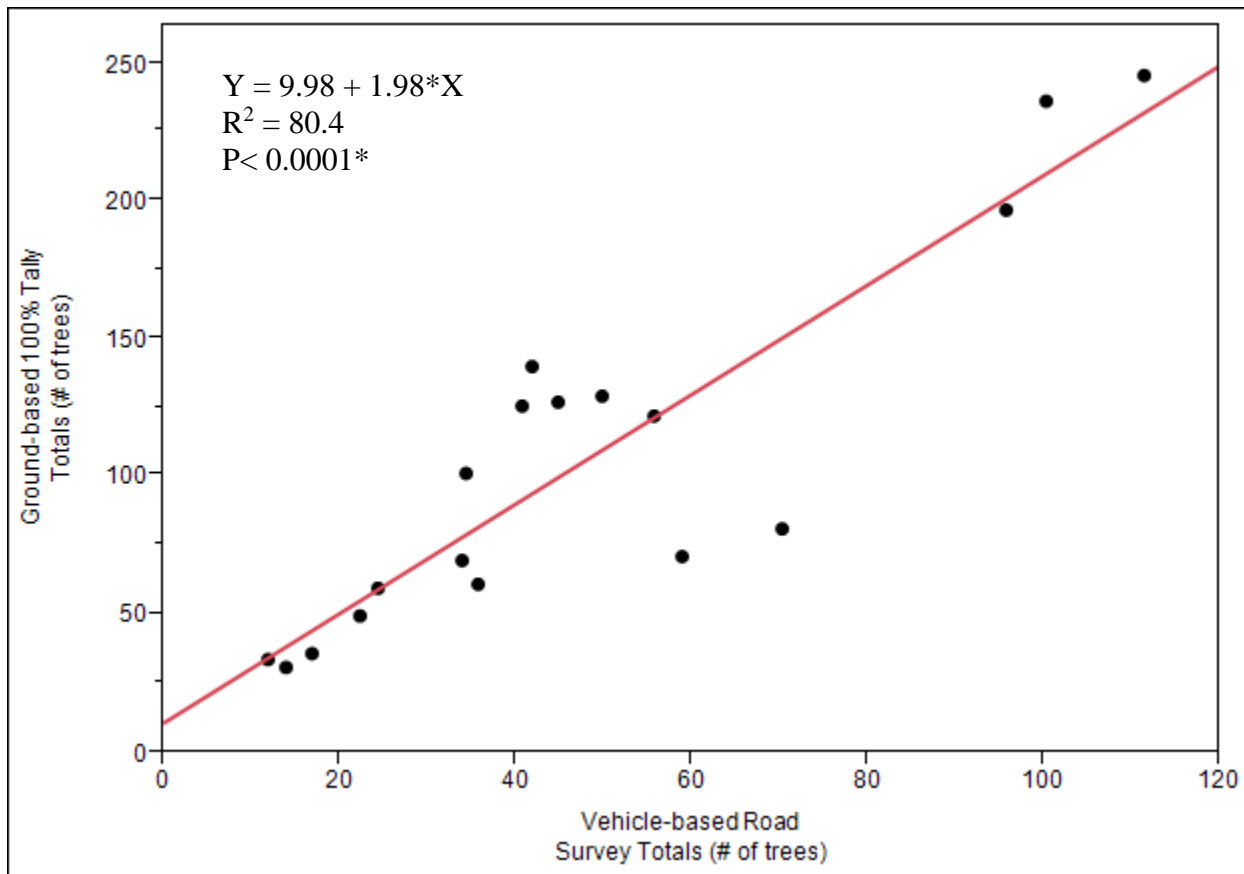
**Figure 2: Number of dead and fading trees at each location determined by ground-based 100% tally in spring 2012 and spring 2013. Each number is the total number of trees recorded within the 20 hectares of each site.**



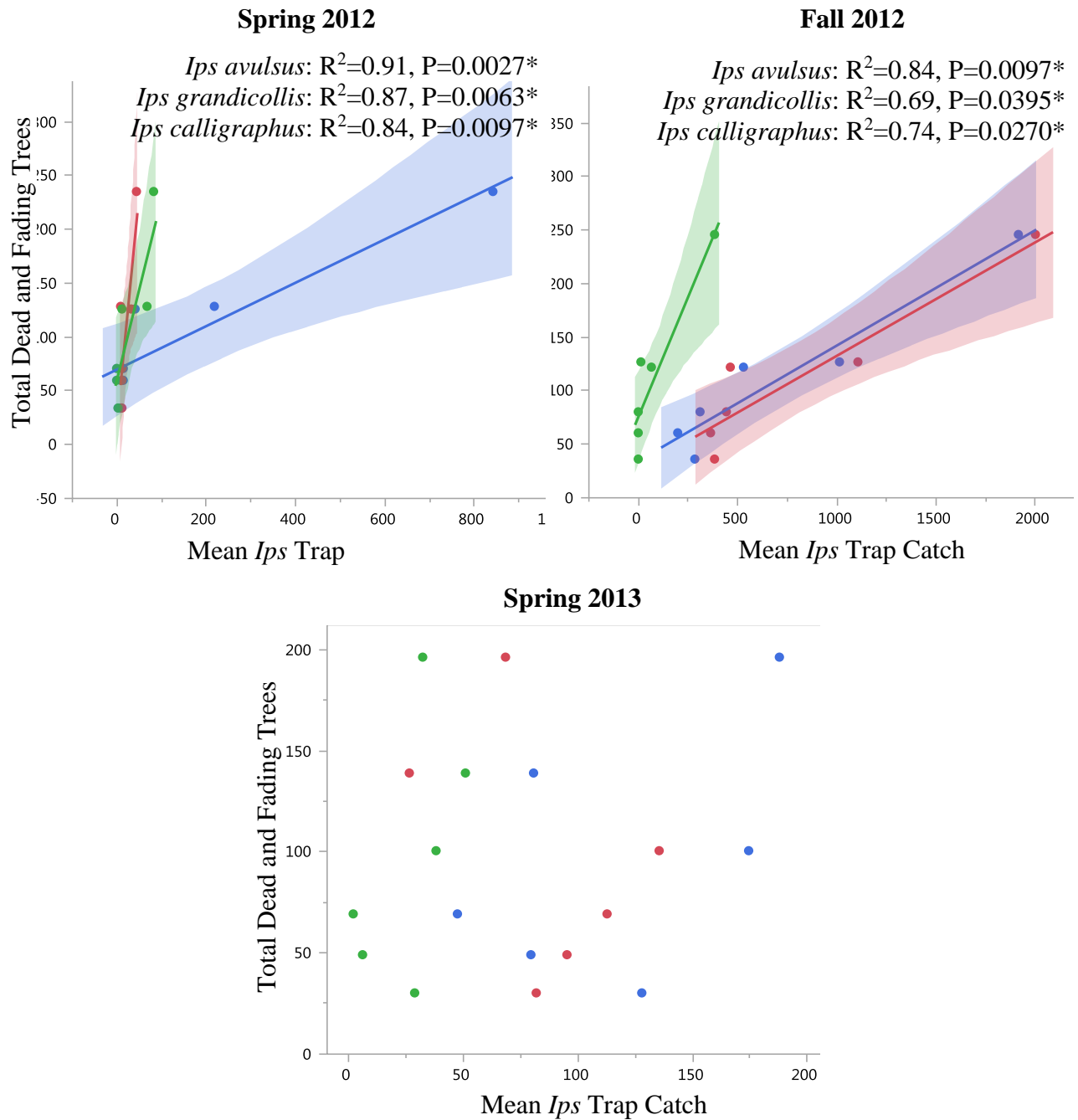
**Figure 3: Comparison of total *Ips* trap catches between stands with subjectively high or low estimates of tree mortality. Figures created using JMP Pro 10 (SAS Institute Inc., Cary, NC).**



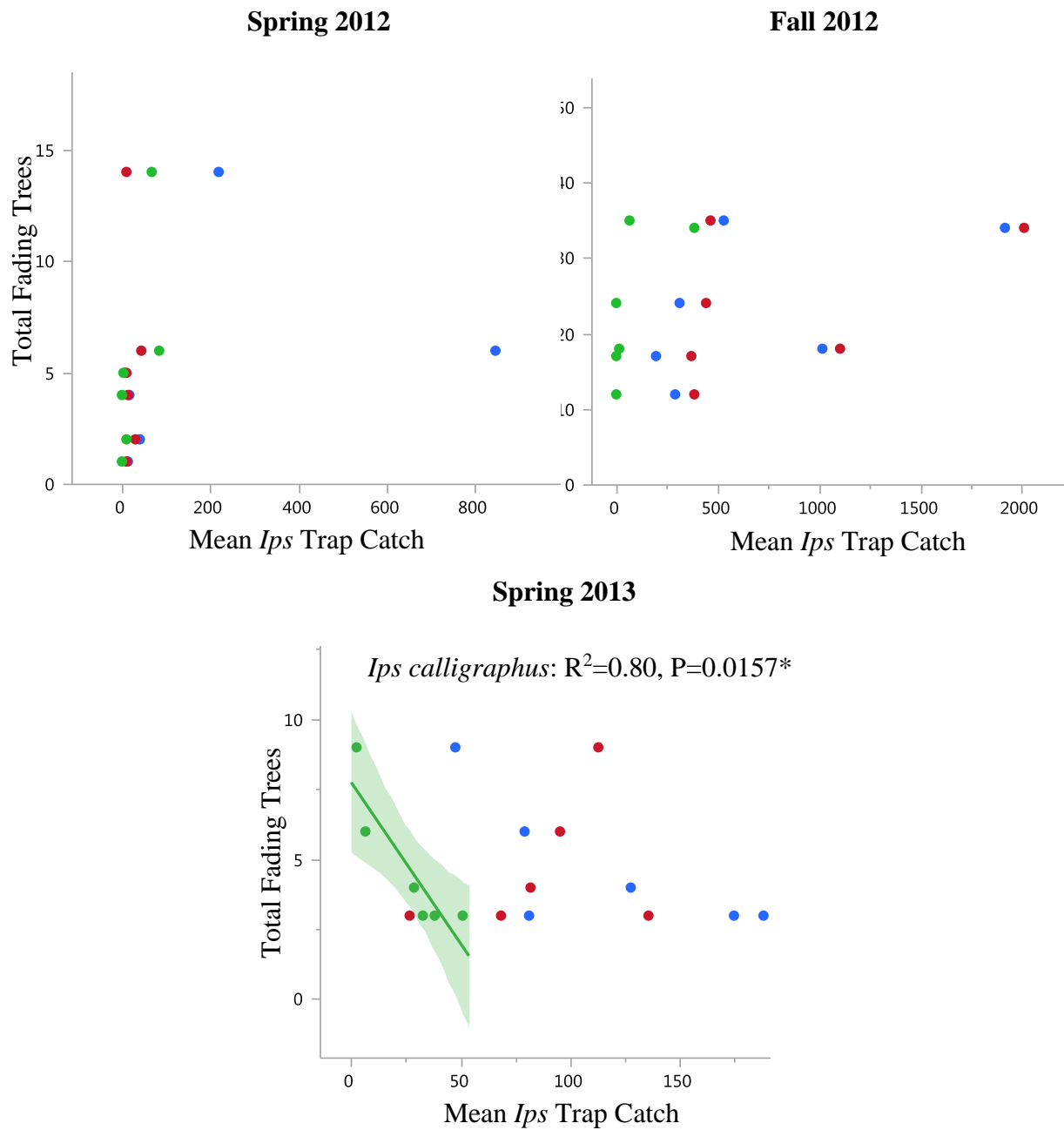
**Figure 4: Correlation of tree mortality estimated by vehicle-based road survey to actual tree mortality determined by ground-based survey (n = 18; six locations with three surveys each). Surveys completed in spring 2012, fall 2012, and spring 2013. Figure created using JMP Pro 10 (SAS Institute Inc., Cary, NC).**



**Figure 5: Relationship between the total of dead and fading trees recorded by ground-based survey and mean *Ips* trap catch (n = 6 for each *Ips* species). *Ips avulsus* = Blue, *Ips grandicollis* = Red, and *Ips calligraphus* = Green.**



**Figure 6: Relationship between number of fading trees determined by ground-based survey and mean *Ips* trap catch (n = 6 for each species). *Ips avulsus* = Blue, *Ips grandicollis* = Red, and *Ips calligraphus* = Green.**



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## **CHAPTER 4 – A CASE STUDY OF A NATIVE CURCULIONID OUTBREAK: *PISSODES NEMORENSIS* IN SOUTHERN PINE FORESTS**

### **INTRODUCTION**

*Pissodes nemorensis* (Germar) (Coleoptera: Curculionidae), the eastern pine weevil, is a common phloem-feeding beetle native to conifer forests of eastern North America (Jones 1965, Phillips *et al.* 1987). Though rarely considered a pest, unusually high densities are capable of causing tree mortality in southern pine plantations (Ollieu 1971, Overgaard & Nachod 1971). *Pissodes nemorensis* is an associate of other damaging pine-inhabiting insects, such as southern pine engraver beetles (*Ips* spp.) and black turpentine beetle (*Dendroctonus terebrans* (Olivier)). Our objective is to compile information of life history and impact of this often overlooked insect. Additionally, we report the methods and results of a trapping study that was implemented to assess current abundance of *P. nemorensis* on intensively managed pine plantations in southern Arkansas. We also hypothesize as to the role of *P. nemorensis* in southern pine forests and address gaps in our current knowledge.

Owing to a wide host range of conifer species, *P. nemorensis* can be found throughout much of eastern North America (Smith and Sugden 1969). In the northeastern United States and eastern Canada, *P. nemorensis* prefers, but is not limited to, spruce (*Picea* spp. Mill.), eastern white pine (*Pinus strobus* L.), and red pine (*P. resinosa* Aiton). In the southeastern U.S. states, it can colonize all southern pines, including loblolly (*P. taeda* L.), shortleaf (*P. echinata* Mill.), longleaf (*P. palustris* Mill.), and slash pine (*P. elliottii* Engelm.). *Pissodes nemorensis* also infests imported cedars, such as deodar cedar [*Cedrus deodara* (Roxb.) G. Don f.], atlas cedar [*C. atlantica* (Endl.) Manetti ex Carrière], and cedar of Lebanon (*C. libani* A. Rich).

Previously distinct species of *Pissodes* are now synonymous with *P. nemorensis*. A. D. Hopkins (1911) described *Pissodes deodarae*, a weevil breeding in exotic deodar cedar. He differentiated it from *P. nemorensis* by slight morphological characteristics. However, a comparative study between *P. nemorensis* and *P. deodarae*, suggested they were the same species (Dietrich 1931). This connection to imported cedar trees is where the common name, deodar weevil, is derived. Another important synonymy is in regard to the northern pine weevil, *Pissodes approximatus* Hopkins, and *P. nemorensis*. Diagnostic morphological characteristics do not exist to differentiate individuals of the two species although the populations were said to be distinguishable by a discriminate analysis (Godwin et al. 1982). When the morphological characters, seasonal flight activity, and reproductive development across the entire east coast were reevaluated, *P. approximatus* and *P. nemorensis* were synonymized as a single species with intraspecific variation across its range (Phillips *et al.* 1987). In light of this synonymy, the more recent common name, eastern pine weevil, acknowledges the expansive range of the weevil.

The lifecycle of *P. nemorensis* is well understood. Throughout its range it is univoltine. Adult weevils feed on host tree phloem by inserting their rostra through the bark. Based on anecdotal evidence, feeding occurs on small diameter shoots in the tree crown, in crevices of thicker bark, or where injury has occurred on the bole of the tree. The only evidence that feeding has occurred is inconspicuous 0.5 mm size holes. Females oviposit one or two, or occasionally more, eggs into a feeding site (Dietrich 1931, Fontaine and Foltz 1985, Atkinson *et al.* 1988a). Larvae construct galleries that are characterized by more vertical than horizontal orientation. The galleries are sometimes described as “H” shaped (Jones 1965). Jones discovered that, *in vitro*, the grub-like larvae grow to an average length of 6.9 mm, but may reach a maximum length of 12 mm. However, this average length may be greater in the field. After completing four instars,

5th stage larvae construct pupal chambers, aptly named a “chip cocoons” based on their construction from strips of sapwood (Ollieu 1971, Atkinson 1979). The pupae mature into adults in approximately two weeks (Jones 1965). Adult weevils emerge by chewing through the chip cocoon and the bark. Circular emergence holes may be observed with an average diameter of approximately 3 mm.

*Pissodes nemorensis* has dissimilar life histories in the northern and southern U.S. states (Figure 1). Northern *P. nemorensis* reproduce in late spring and larvae develop during the summer. Typically, adults overwinter in the duff layer of the ground. In contrast, the southern *P. nemorensis* life history appears to have “shifted” on account of the hot temperatures of summer (Phillips *et al.* 1987, Atkinson *et al.* 1988a). In the Gulf States, *P. nemorensis* aggregate and oviposit on host trees between September and March (Jones 1965, Fontaine and Foltz 1982). The peak of dispersal is typically observed in October and November (Jones 1965). In Florida, this peak occurs in late-November and early-December (Atkinson 1979, Fontaine and Foltz 1982). Oviposition slows during cold winter months, but can resume when temperatures are sufficiently warm in early spring. *Pissodes nemorensis* in the southern states is the only North American *Pissodes* species with a breeding season during fall and winter (Atkinson *et al.* 1988a).

Despite the clear distinction between the life histories of northern and southern states, their ranges are not allopatric and reproductive isolation does not occur. In central states such as Virginia, adult weevils may be caught in traps in both spring and fall months (Phillips *et al.* 1987). Consequently, the long oviposition periods of *P. nemorensis* allows introgression of mating between “fall-active” and “spring-active” adults, a fact which further supports the synonymy of *P. approximatus* and *P. nemorensis* (Phillips *et al.* 1987). Furthermore,

mitochondrial DNA of *P. nemorensis* collected from Florida and New York shows very low intraspecific sequence divergence across that range (Langor and Sperling 1995, 1997).

*Pissodes nemorensis* in the Gulf States are elusive during summer months. Between April and August, injured pines and pheromone-baited traps fail to attract adult weevils (Dietrich 1931, Jones 1965, Fontaine *et al.* 1983, Atkinson *et al.* 1988a, Atkinson *et al.* 1988b). Consequently, few researchers have observed the summer habits of *P. nemorensis*. It was assumed that *P. nemorensis* undergo a period of aestivation, remaining inactive until temperatures are less extreme (Dietrich 1931, Jones 1965, Ollieu 1971). When temperatures exceed 85°F, activity “slows to a standstill” and survival greatly declines (Jones 1965). The more recent suggestion is that weevil adults become quiescent in leaf litter during hot conditions and feed on small diameter pine shoots during cool periods of the day (Fontaine *et al.* 1983) (hatched pattern in Figure 1). Much of their behavior during summer months remains unknown, and it is uncertain if they take flight during this period.

In Gulf States, when *P. nemorensis* emerge, they are not reproductively mature and require an extended preoviposition period. A period of maturation feeding is necessary for females to produce eggs (Fontaine *et al.* 1983). Since *P. nemorensis* do not respond to host volatiles or pheromones during the summer, it is believed that their preoviposition period occurs from spring emergence until late summer. Under lab conditions and a temperature fluctuation of 20 – 27.2°C, the preoviposition period only lasts an average of 12.2 days (Jones 1965). A period of reproductive maturation is observed in which newly emerged adults are incapable of reproduction for several weeks (Fontaine and Foltz 1985). Females, at 25°C, develop oocytes and eggs for the first time after three and five weeks respectively, but males do not begin mating until after six weeks post-emergence. When weevils, which emerged in May, were placed inside

outdoor cages, they exhibit slower reproductive maturation than their lab-reared counterparts (Fontaine and Foltz 1985). Also, females in outdoor cages do not have sperm in their spermathecae until late August.

Successful aggregation to host trees is accomplished by a male-released pheromone. Traps that incorporate feeding adult male *P. approximatus* weevils attract additional males and females, suggesting the presence of an aggregation pheromone (Booth and Lanier 1974). The same aggregation effect is observed with *P. nemorensis* (Atkinson 1979). A monoterpene alcohol, grandisol, and its corresponding aldehyde, grandisal, are the aggregation pheromones that have been isolated *in vitro* (Booth *et al.* 1983). Both of these pheromones and freshly cut pine bolts synergistically attract a significantly higher numbers of weevils than any treatment that lacks one of those components (Phillips *et al.* 1984).

Due to their very effective aggregation pheromone, *P. nemorensis* achieves dense colonization of host trees. However, information about what causes the primary attraction, or kairomonal response, of *P. nemorensis* to host trees remains limited. *Pissodes nemorensis* has been reported as the sole inhabitant within attacked trees (Conner Fristoe, Plum Creek Inc., personal communication 2011, Ollieu 1971, Overgaard & Nachod 1971, Jones 1965). This was observed when stand-wide densities peak. Occasionally, they may also be found in close association with pine engraver beetles (*Ips* spp.) and *D. terebrans*; however, the observations of larval maturity during this study indicate that *P. nemorensis* could potentially be the primary colonizer in those instances.

Outbreaks of *P. nemorensis* in the Gulf States are rarely reported in scientific journals or statewide damage reports. However, land managers in East Texas observed significant losses in sapling-sized loblolly plantations during the late 1960s (Ollieu 1971). In 1970, forest health

specialists in Louisiana reported nearly 200 dead loblolly pines in 13- to 15-year-old plantations (Overgaard & Nachod 1971). There have been no recent reports in the scientific literature of *P. nemorensis* in the southeast. However, that does not necessarily mean that *P. nemorensis* has no effect in this region. Timber mortality was observed on Plum Creek timberlands of southeastern Arkansas between 2009 and 2011 (Conner Fristoe, Plum Creek Timber Company Inc., pers. comm, 2011). Scattered losses did not exceed 5% for most stands, but heavier losses were observed in stands with greater than 1730 stems/hectare. Also, foresters in Georgia reported loblolly and longleaf pine mortality in Calhoun and Mitchell County between 2011 and 2013 (Laurie Reid, South Carolina Forestry Commission, pers. comm. 2013, Chuck Norvell, Georgia Forestry Commission, pers. comm. 2013). The stands in Georgia also had scattered mortality not exceeding 5%. In both Arkansas and Georgia, the appearance of densely aggregated chip cocoons indicated that *P. nemorensis* contributed to the observed timber losses. The affected stands ranged from 9 to 21 years in age, were genetically improved loblolly or longleaf pine, had high stem-density (greater than 1000 stems/hectare), had complete canopy cover, and had not been precommercially or commercially thinned (Chuck Norvell, Georgia Forestry Commission, pers. comm., 2013).

This study investigated tree mortality in loblolly plantations belonging to Plum Creek Timber Company Inc. located in southeast Arkansas, within Ashley, Lincoln, and Drew counties. The objectives were to gain preliminary information about the relative abundance of *P. nemorensis* and other stem feeding beetles during the reported peak oviposition period and to assess the densities of *P. nemorensis*. Additionally, the trapping efficacies of two trap designs were compared.

## METHODS

Two traps were installed at five different sites in southeastern Arkansas during fall 2011 (Figure 2). Three sites were located in Ashley County, one site in Lincoln County, and one site in Drew County. All sites supported even-aged, loblolly pine plantations belonging to Plum Creek Timber Company, Inc. Stand ages ranged from 11 to 21 years old. The two traps at each site were a Lindgren® eight-unit funnel trap and an APTIV Intercept™ black panel trap. A distance of no less than 14 kilometers separated the five different trapping sites. At each site, traps were at least 25 meters apart and hung at a height such that the reservoir at the bottom was suspended approximately 0.5 m above the ground (Figure 3).

Both traps were baited with an inexpensive lure composed of a 1:1 mixture of ethanol and turpentine (Klean-Strip® Green™ Pure Turpentine from Tree Resin). A 250 ml Nalgene® bottle with a small hole drilled in the screw-on top was fitted with a braided cotton dental wick (15 cm long x 1 cm diam.) that extended 2.5 cm from the bottle. The wick facilitated volatilization of the lure. The combination of ethanol and turpentine is effective as a low-cost, broad-range attractant for bark infesting insects, including *P. nemorensis* (Phillips *et al.* 1987, Rieske and Raffa 1993).

The traps remained in the field for a six-week period between October 18 and November 29, 2011. Every two weeks, the contents of the traps were collected and the lures were replaced.

To supplement data from the fall 2011 trapping period, we examined the bycatch of traps from a previous study that used a similar trapping technique and lure system. This study took place during fall of 2009 and 2010 in two sites in Ashley County. The two Ashley Co. sites were those referenced above that were used in 2011. For this trapping study, three Lindgren® eight-

unit funnel traps were baited with 95% ethanol and Contech Inc. *Sirex* lure ( $\alpha$ - $\beta$  pinene) with the goal of attracting a native siricid woodwasp, *Sirex nigricornis* F. (Keeler 2012). Like the combination of ethanol and turpentine, ethanol and  $\alpha$ - $\beta$  pinene effectively catch a wide variety of phloem-feeding insects and their associates, including *P. nemorensis*. Insects caught in those traps were collected every two weeks. In 2009, the traps remained in the field between October 20th and December 1st, whereas in 2010, the traps were used between August 19th and December 2nd.

## RESULTS

Only 17 *P. nemorensis* were collected during the 2011 trapping period, but they were caught in four of the five sites. None were captured after November 15 in 2011. With the traps designed to capture *Sirex*, 316 *P. nemorensis* were trapped between October 20th and December 1st of 2009, but only 18 were trapped between September 23 and December 2 in 2010. The trap catch trends for two Ashley County sites were compared for all three years (Figure 4). In all years, it appears that traps failed to catch *P. nemorensis* near the end of November. Analysis of variance indicated that panel traps and Lindgren® traps did not differ in efficacy for catching *P. nemorensis* (Figure 5).

## DISCUSSION

The trapping study in 2011 had limited success in capturing *P. nemorensis*. The locations were chosen based on previous evidence of mortality associated with *P. nemorensis*. Outbreaks were reported in 2009 and 2010 by Plum Creek Timber Co. Following those apparent outbreaks, it appears that a population crash occurred before trapping was initiated in 2011. Owing to the

lack of weevils captured and absence of visibly infested trees, analyses related to relative abundance of weevils and stand characteristics could not be completed.

There is no evidence to suggest why the relatively high weevil densities observed in 2009 declined sharply the following year. Stand structure remained the same between 2009 and 2010. For most of the stands, thinning took place in summer of 2011, long after the 2010 traps yielded low counts. This suggests that thinning did not influence the weevil population crash. The population dynamics of *P. nemorensis* are poorly understood. In addition, there is limited knowledge about how natural enemies influence *P. nemorensis* densities in the southeastern United States. It may be hypothesized that *P. nemorensis* is vulnerable to a complex of predators, parasitoids, and competitors similar to those which affect bark beetles populations (i.e. *Ips* spp. and *Dendroctonus* spp.). For example, *Monochamus carolinensis* (Olivier) and *M. titillator* (F.) larvae may negatively affect *P. nemorensis* populations during fall months due to facultative predation (Dodds 2001).

The observed peaks in trapping data lead us to assume that the oviposition period was not missed during the dates of trapping. It appears that we were able to trap during the peak of oviposition in both 2009 and 2010. Our results confirm earlier findings that the highest numbers of *P. nemorensis* in Arkansas are captured between October and November (Jones 1965). The trapping in this study was stopped after November 29 due to a lack of weevils caught, which was likely the result of cold temperatures and extended periods of rain. Cold weather events beginning in mid-November in Arkansas may limit dispersal of *P. nemorensis*.

The population dynamics of *P. nemorensis* may also be affected by summer temperatures. Harsher summer temperatures may limit survival (Jones 1965). Also, we

hypothesize that increases in densities of *P. nemorensis* may correspond with a denser stand structure that provides a shaded understory.

When trap counts of Lindgren<sup>®</sup> and panel traps were compared, the counts of *P. nemorensis* did not vary significantly, but our trap counts were very low. The only species caught with a significant difference between trap types was *D. terebrans*. The panel traps caught more *D. terebrans*, and this agrees with a similarly finding by Miller and Crowe (2011). Other trap performance experiments were not conducted during the dispersal period of *P. nemorensis*.

Future research may benefit from other trap designs. A “lower-stem flight trap,” such as the one used in a study by Erbilgin and Raffa (2002), might be more effective for trapping adult *Pissodes* spp. The trap is made from a modified gallon-sized milk container. Three sides are removed from the container and one side remains as a striking surface. The container is inverted and fixed to the basal section of the tree. This trap is greatly more effective for catching *Pissodes* spp. than funnel traps or pitfall traps (Erbilgin and Raffa 2002).

In light of the low trap counts, it may be necessary to use the more specific aggregation pheromones in order to more effectively capture *P. nemorensis*. Though host volatiles are proven to attract *P. nemorensis* and its associates, *P. nemorensis* may preferentially fly toward aggregation pheromones. Host odors are important for close-range orientation of walking weevils, whereas aggregation pheromones may be more important for *P. nemorensis* in flight (Phillips *et al.* 1984). Another potential reason for the low trapping results of our 2011 study may relate to the volatilization rate of the lures. The concentration of volatiles may alter the ability of the lure to attract. *Pissodes strobi* appears to respond to a “concentration dependent threshold” with host volatiles (Phillips 1981). It is possible to hypothesize that the observed rate of volatilization may be too extreme for flying weevils to accept the lure as an acceptable host.

Unlike the lures used in 2011, the traps used in the 2009 and 2010 trapping studies utilized ethanol and  $\alpha$ - $\beta$  pinene packaged in perforated bags which controlled volatilization. Future research on primary attraction should compare the trap counts associated with various release rates of host volatiles.

Based on observations of densely aggregated colonization, *P. nemorensis* can sustain a population over a large area with only a few host trees. During endemic population levels, dense colonization patterns of *P. nemorensis* will recur in nearly all pine forests of Arkansas. Furthermore, their late-fall and winter oviposition period of is unusual for pine-infesting beetles of the south, and it may be advantageous for *P. nemorensis* in regards to limiting interspecific competition within the host trees.

The outbreak densities observed in 2009 may have been facilitated by stand conditions. It appears that higher *P. nemorensis* densities are found in stands with a greater availability of susceptible trees. The crown closure and dense spatial arrangement of trees may have provided a microclimate favorable for adult survival and increases in abundance. Tree susceptibility in our stands could have also been attributed to extreme changes in water availability, such as drought or, conversely, heavy rains. Additionally, the sites used in our study may have not historically hosted loblolly stands, and such human-mediated changes may inadvertently increase tree susceptibility to herbivore outbreaks (Clarke *et al.* 2000). Without pine management, these stands would likely feature a variety of woody plants and hardwood species that could tolerate changes in the water table.

Trapping studies which only use sex-pheromones that attract competitors, such as *Ips* spp., fail to capture *P. nemorensis*. *Pissodes nemorensis* are only captured when host volatiles or their own aggregation pheromones are used. This may be a mechanism for avoiding competition,

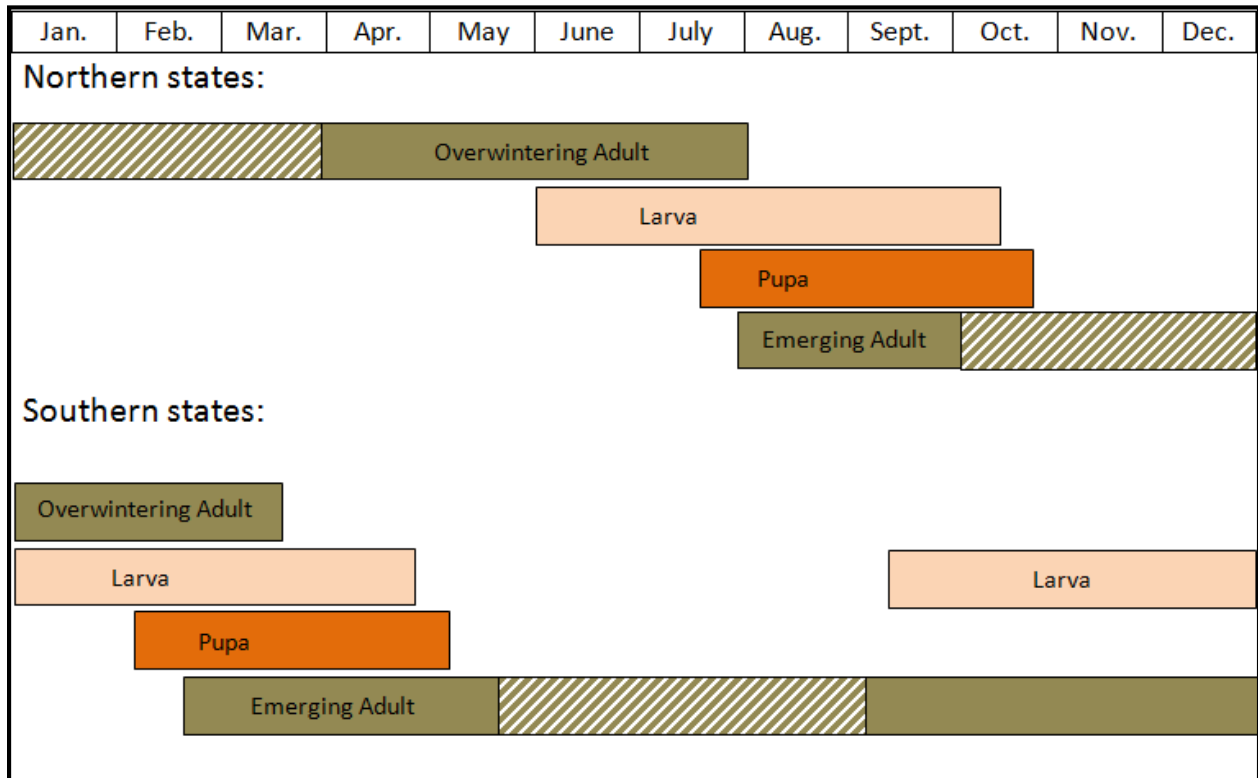
as it would be advantageous for *P. nemorensis* to be attracted to a stressed living tree that was not already colonized by its competitors. It is probable that hosts colonized by competitors inhibit *P. nemorensis* attraction, and therefore *P. nemorensis* preferentially oviposit on trees attacked by only *P. nemorensis*.

## CONCLUSION

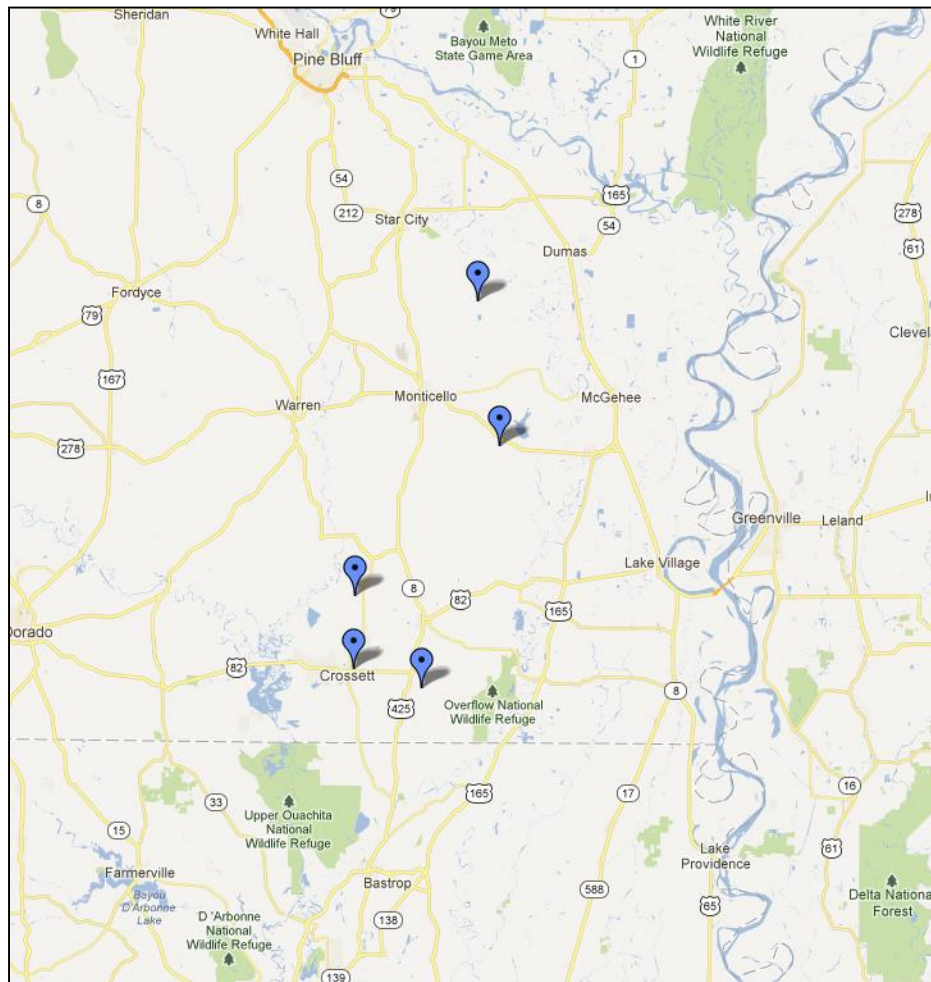
Relative abundance of *P. nemorensis* in southern Arkansas was high during 2009, but the populations crashed and trap counts were low in the following two years. Tree mortality appeared to occur in pine plantations with high stem density and complete canopy closure. There is not enough evidence to suggest that either Lindgren<sup>®</sup> or panel traps perform better for catching *P. nemorensis*; future trapping studies should test the effectiveness of the novel lower-stem flight trap.

## FIGURES

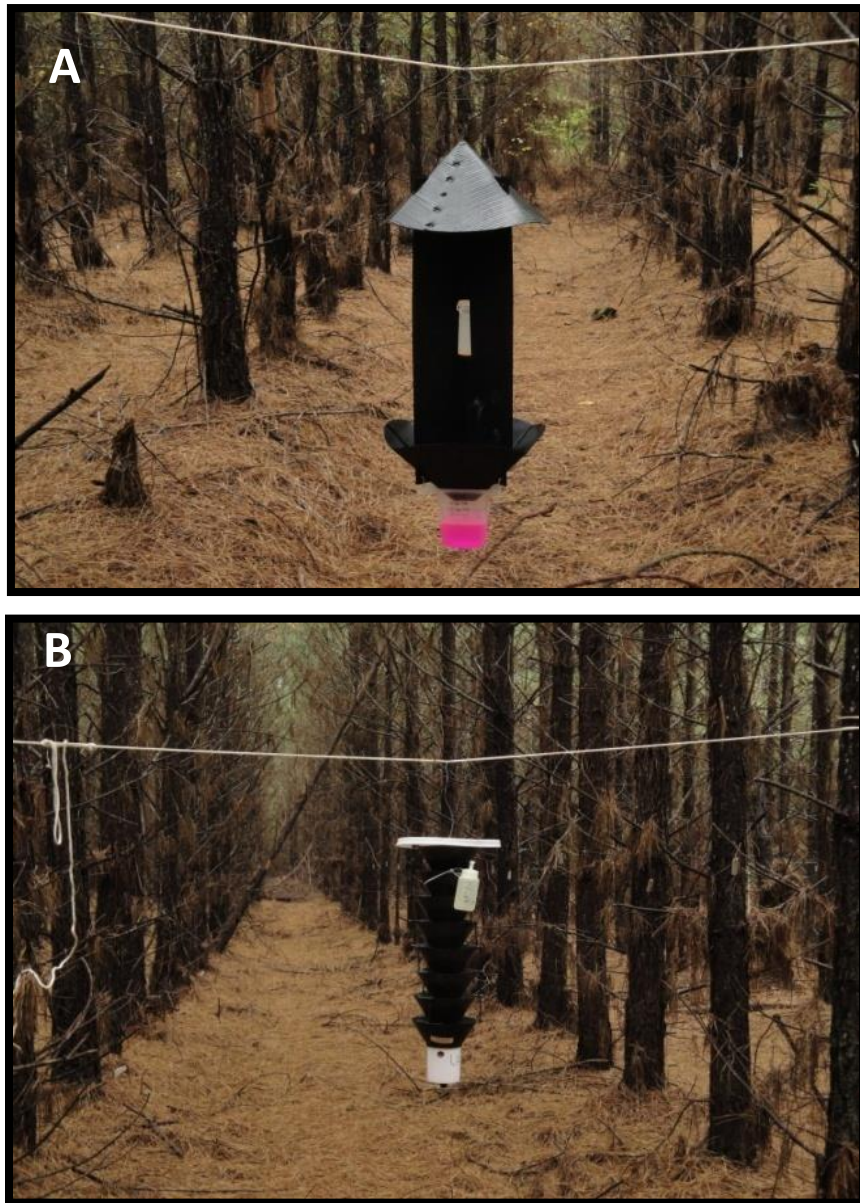
**Figure 1: Generalized life histories of *Pissodes nemorensis* found in northern and southern states; derived from Finnegan (1958), Atkinson (1979), Fontaine *et al.* (1983), Phillips *et al.* (1987), and Atkinson *et al.* (1988a). Hatched pattern indicates periods of limited adult activity.**



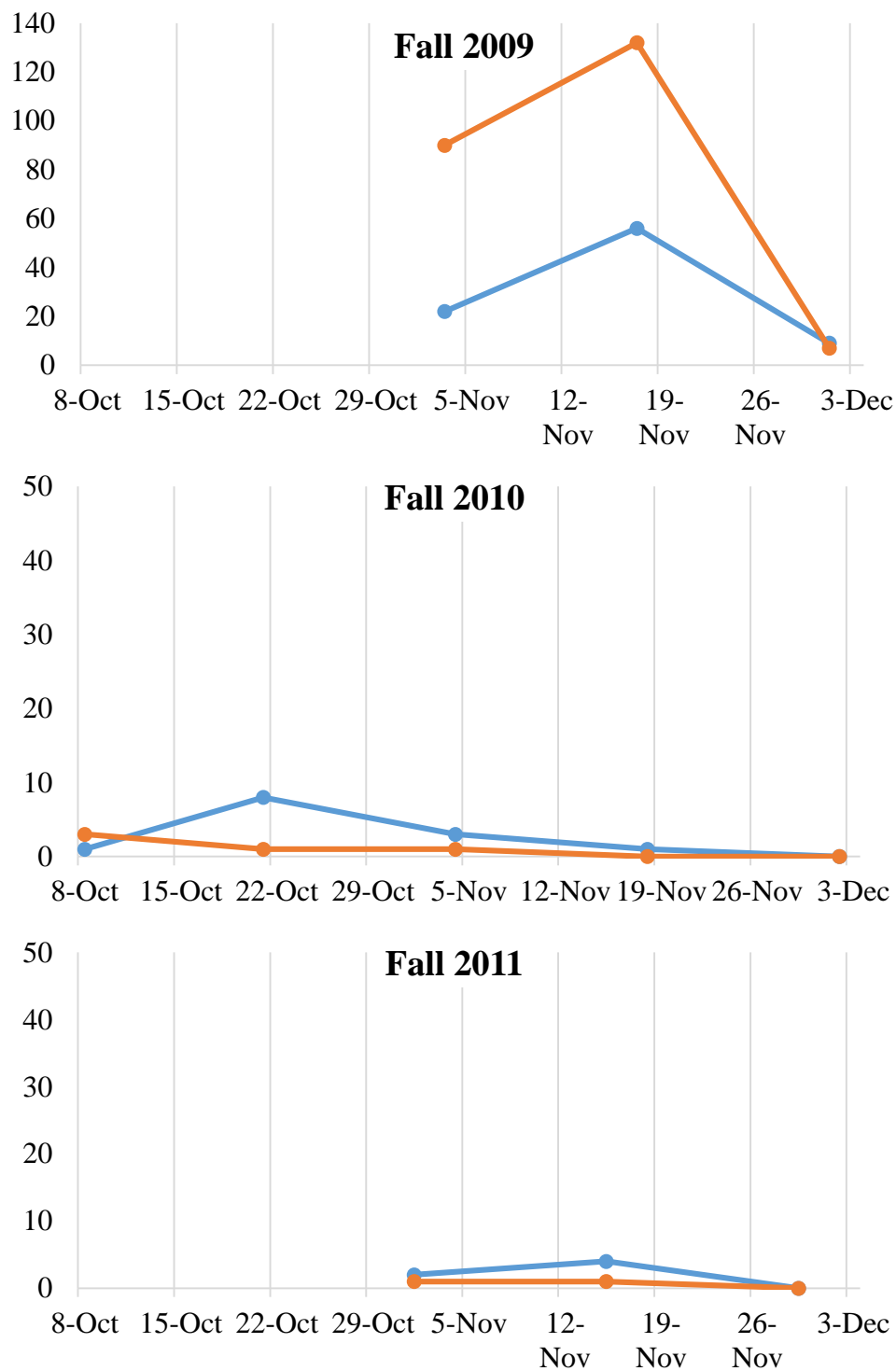
**Figure 2: Trap locations in southeastern Arkansas.**



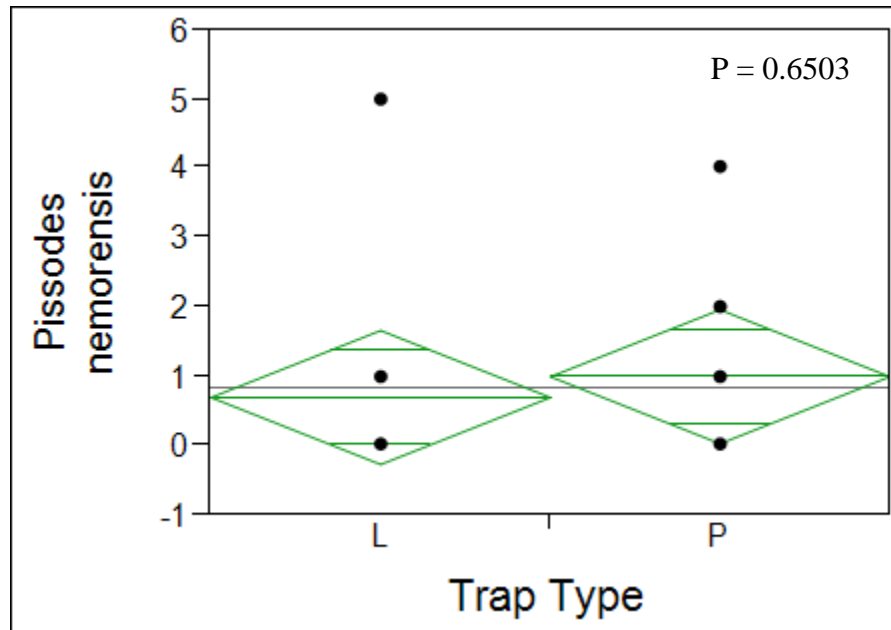
**Figure 3: Panel trap (A) and Lindren<sup>®</sup> eight-unit funnel trap (B). Photo credit: Chandler Barton.**



**Figure 4: Total number of *Pissodes nemorensis* caught in 2009, 2010, and 2011 at two sites located in Ashley County, Arkansas. For 2009 and 2010: each point is a sum of three traps at each location on that respective collection day. For 2011: each point is a sum of two traps.**



**Figure 5: Comparison of the distribution of *Pissodes nemorensis* trap counts for Lindgren funnel (L) and panel (P) trap. Figure created using JMP Pro 10 (SAS Institute Inc., Cary, NC).**



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## CONCLUSION

This research was designed to help develop methods for future *Ips* monitoring and tree mortality estimations. The pheromone lure system effectively captured all three species of southern pine engravers and their natural enemies. Between spring 2012 and 2013 (a total of 12 trapping weeks), 127,823 *Ips avulsus*, 217,636 *I. grandicollis*, and 26,290 *I. calligraphus* were captured. *Ips calligraphus* was the least numerous and this was especially true in the Arkansas sites.

*Ips* pine engravers were captured most abundantly during the spring and summer trapping seasons of 2012, with 135,938 and 194,169 respectively. In comparison, trapping during the fall yielded very few *Ips* (21,508). Also, during the spring of 2013, only 20,134 *Ips* spp. were captured, strongly suggesting that high population levels had subsided.

The ratio of males captured by the pheromone traps was sampled for each *Ips* species and season. The overall average male ratio was 0.55 for *I. avulsus*, 0.29 for *I. grandicollis*, and 0.31 for *I. calligraphus*. These ratios are similar to results found in other pheromone trapping studies. For *I. grandicollis* and *I. calligraphus*, the male ratios varied slightly among seasons, but the ratios for *I. avulsus* for all seasons were not statistically different.

Several coleopteran predators were captured in the pheromone traps. The most numerous were Cleridae (*Thanasimus dubious* F. and *Enoclerus* spp.), *Temnochila virescens* (F.), Histeridae (*Platysoma* spp. and *Plegaderus* spp.), and *Lasconotus* spp. Interestingly, *Lasconotus* spp., known as a predator/scavenger of *Ips* spp., was the most abundant over the course of the study with a total of 42,313. Of all the predators, the correlation between *Lasconotus* spp. trap catch and *Ips* spp. trap catch was the strongest ( $R^2 = 0.44$ ,  $p < 0.0001^*$ ). Cleridae trap catch did not correlate with *Ips* spp. trap catch ( $R^2 = 0.002$ ,  $p = 0.3581$ ). *Monochamus* spp., a competitor

for phloem and facultative predator of *Ips*, had a weak correlation with *Ips* spp. trap catch, though the relationship was statistically significant ( $R^2 = 0.04$ ,  $p = 0.0007^*$ ).

Prior to the study, the usefulness of vehicle-based tree mortality surveys was uncertain. The number of fading and standing dead trees recorded in the vehicle survey was compared to the ground-based survey method, and we found a strong correlation between the two survey methods ( $y = 9.98 + 1.98x$ ,  $R^2 = 80.4$ ,  $P < 0.0001^*$ ). However, there are some noticeable caveats to using vehicle surveys. First, the tree density and understory thickness alter the surveyor's ability to see into the stand from the road. Second, a vehicle-based method can only detect fading and standing dead trees, whereas the ground-based method allows for a closer evaluation of tree condition and cause of death.

The sites used in this study were selected based on subjective appearance of *Ips*-associated mortality. Within each of the three regions, a site with high incidence of mortality and a nearby site of low incidence was chosen, but it was unknown if the *Ips* abundance actually differed significantly. We found that despite the clear differences in tree mortality, the abundance of *Ips* did not always differ significantly. For spring 2012, *Ips* abundance was significantly different between high and low sites for two of the three regions. In spring 2013, only the sites in Texas were significantly different.

A key objective of this study was to find any relationships between *Ips* abundance and tree mortality. The strongest relationships between *Ips* spp. trap catches and the number of standing dead trees were found in spring of 2012 when the outbreak was considered still active. A statistically significant correlation existed in fall 2012 as well. Interestingly, a direct correlation could not be found between *Ips* abundance and the number of actively fading trees alone.

This study appeared to track the decline of *Ips* abundance following a period of visible outbreak. As such, it was difficult to evaluate the predictive value of *Ips* trap catch totals in relation to tree mortality surveys, or vice versa. Additional years of data collection would be necessary to evaluate if detection of rising *Ips* populations is possible with these monitoring techniques.

Future studies should focus efforts on trapping during the spring or summer seasons. One important difference between these seasons is the abundance of natural enemies. Spring traps caught fewer natural enemies than summer traps. If an assessment of natural enemy abundance is a goal of future studies, our data suggest that trapping should take place during the warmer, summer months. If a strict assessment of *Ips* abundance is an objective, then the spring months may be more suitable. Additionally, from our observations, the spring months were better suited for detecting fading trees from the road. Tree mortality surveys in the spring will more easily identify potential *Ips* outbreak sites.