

Trapping for *Sirex* Woodwasp in Brazilian Pine Plantations: Lure, Trap Type and Height of Deployment

Elder S. P. Batista¹ · Richard A. Redak² ·
Antonio Carlos Busoli¹ · Mariane B. Camargo³ ·
Jeremy Dean Allison⁴

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Abstract The *Sirex* woodwasp, *Sirex noctilio* (Hymenoptera: Siricidae) is considered a secondary pest of pine in its native range but has caused considerable economic losses in pine plantation forests in the southern hemisphere. In Brazil, trap trees are the primary tool used for early detection purposes but these are costly, labor-intensive to install and require stressing trees by herbicide application. Flight intercept traps baited with synthetic blends of host volatiles are an attractive alternative but have performed poorly in some settings. This study was carried out to look for alternatives to trap trees for use in Brazilian pine plantations for early detection of *S. noctilio*. Four field experiments were conducted in two consecutive flight seasons (2015–16 and 2016–17), in planted loblolly pine (*Pinus taeda*) stands, to compare captures among flight intercept traps baited with different lures, deployed at different heights and among different intercept trap designs. Two experiments compared different host volatile lures and a significant treatment effect was observed in one. No effect of trap design or height was observed.

Keywords European woodwasp · survey and detection · woodborer · invasive species

✉ Elder S. P. Batista
elderspb@gmail.com

¹ Departamento de Fitossanidade, Universidade Estadual Paulista, Unesp/FCAV, Via de Acesso Prof. Paulo Donato Castellane, s/n, Jaboticabal, SP 14884-900, Brazil

² Department of Entomology, University of California, Riverside, CA 92521, USA

³ Klabin Florestal, Av. Brasil, 26 – Harmonia, Telêmaco Borba, PR 84275-00, Brazil

⁴ Natural Resources Canada, 1219 Queen Street East, Sault Ste Marie, ON P6A 2E5, Canada

Introduction

Attractant-baited intercept traps are widely used in forest insect pest management surveys and detection programs [e.g., Early Detection and Rapid Response Program (Rabaglia et al. 2008)], to predict population trends of pest species [e.g., the southern pine beetle (Billings and Upton 2010)], to document the distribution of non-native and outbreak species, and to evaluate the impact of management efforts (Myers et al. 2000; Liebhold and Tobin 2008). Numerous trap design features can influence the abundance and diversity of forest insects captured. These include intercept trap type, type of collection cup or killing agent, trap surface treatment, modifications to the collection column and the visual cues presented (McIntosh et al. 2001; Allison et al. 2014; Dodds et al. 2015; Allison and Redak 2017). A large number of studies have quantified the impact of trap design and deployment methodology on the capture of forest Coleoptera and some patterns in trap design are beginning to emerge (Allison and Redak 2017). Regardless, existing strategies are almost always insect-specific and their breadth of utility unknown. Consequently, for numerous pest species, operational survey and detection tools do not exist and/or require further development.

The Sirex woodwasp, *Sirex noctilio* (Fabricius 1793) (Hymenoptera: Siricidae) is native to Eurasia and North Africa but has accidentally been introduced to several countries in the southern hemisphere and more recently into North America (both Canada and the USA, see Slippers et al. 2012). Although considered a secondary pest of pine species in its native range, the Sirex woodwasp has caused considerable economic losses in pine plantation forests in the southern hemisphere (Hurley et al. 2007). The negative impact of this invasive pest on plantation forests has been mitigated by the development of silvicultural treatments that reduce tree stress and by release of several biological control agents (most notably the nematode *Deladenus siricidicola*).

First detected in Brazil in 1988, as of 2012, *S. noctilio* had a distribution of ca. 450,000 ha in the Southern and Southeast regions of Brazil, and some estimates suggest that annual losses caused by this insect could reach 30 million US dollars (Iede et al. 2012). In response to the establishment of *S. noctilio* in Brazil, the National Fund for Woodwasp Control (FUNCEMA) was established with the primary objective of developing a management strategy for *S. noctilio* in Brazilian pine plantation forests. Brazilian management programs involve the silvicultural practice of thinning to promote tree vigor and resistance to attack, and biological control of the wasp. Surveillance tools are used to time thinning operations as thinning can result in increased susceptibility to attack if performed during the *S. noctilio* flight period (Iede et al. 2012). Similarly, the release of biological control agents relies on surveillance, because releases are most effective when they occur before wasp infestations reach high levels. Surveillance of *S. noctilio* in Brazil relies on the use of trap trees that have been stressed by herbicide application (Penteado et al. 2002). These trees are relatively attractive to colonizing female wasps. Although effective, the use of herbicide-stressed trap trees is labor intensive, expensive, and the use of herbicides in forest settings is not widely accepted by the general public.

Alternative surveillance techniques exist and have been used to monitor populations of *S. noctilio* in its invaded range. Ground and aerial surveys have been used extensively in Australia, South Africa and South America (Carnegie et al. 2005; Béeche et al.

2012; Hurley et al. 2012; Klasmer and Botto 2012). Additionally, remote sensing has been tested for use to detect differences in the spectral reflectance of stressed and healthy trees. Unfortunately, like aerial surveys, it is unable to discriminate between *Sirex* stressed/attacked trees and trees with altered spectral properties for reasons unrelated to *Sirex* infestations. Additionally, both remote sensing and aerial surveys have poor detection rates for low density *S. noctilio* populations, particularly those limited to understory trees (Haugen et al. 1990; Carnegie and Bashford 2012).

At low densities, *S. noctilio* almost exclusively infests stressed trees (Neumann and Minko 1981; Dodds et al. 2010; Haavik et al. 2016). This observation was the impetus for the development of trap trees for survey and detection of *S. noctilio* (Madden 1971; Madden and Irvine 1971; Neumann et al. 1982). It also suggests that stressed trees are preferentially attacked, and that attack is mediated by visual and/or olfactory cues associated with stressed pines. Several studies have attempted to identify volatiles that mediate attraction of *S. noctilio* females to stressed pines (Simpson 1976; Simpson and McQuilkin 1976). Field trials in Australia observed that pines treated with semiochemicals designed to represent a stressed pine had an incidence of attack four times greater than untreated pines (Bashford and Madden 2012). Although commercial semiochemical lures have been developed and are used operationally, they have not performed well in some cases. For example, in the southeastern US, native Siricidae (e.g., not *S. noctilio* which is not yet present there) are trapped in higher numbers in traps baited with fresh foliage and branches than commercially-available lures (Barnes et al. 2014). Although no direct comparisons between trap trees and traps baited with commercial *Sirex* lures were made, two North American studies have concluded that trap trees are superior to traps baited with commercial *Sirex* attractants (Zylstra et al. 2010; Barnes et al. 2014). Johnson et al. (2013) compared trap trees and semiochemical-baited traps for survey of *Sirex nigricornis*. Although statistically there were few differences among trap-tree and baited-trap treatments, trap captures were always higher in baited traps, and the authors concluded that semiochemical-baited traps were superior to trap trees. Hurley et al. (2015) report that unlike in North America (e.g., Dodds and de Groot 2012), commercial *Sirex* lures work well in South Africa and speculate that differences in population density among locations (high in South Africa; low in North America) may explain the variation in performance. It is also possible that the variation in performance is a result of qualitative differences between lures used in South Africa and North America.

Studies looking at the effect of trap type have also reported somewhat variable results with some reporting an effect of trap type (McIntosh et al. 2001; Bashford 2008; Allison and Redak 2017) on captures of woodwasps, while other studies have not detected difference in trap type (e.g., Haavik et al. 2014; Barnes et al. 2014; Hurley et al. 2015). Differences among studies in treatments and experimental protocols likely contribute to the varied patterns of treatment effects. This study was carried out in two consecutive years (2015–16) and designed to test the effectiveness of different lures, trap types, and heights of trap deployment on the capture of *S. noctilio* in Brazilian pine plantation forests.

Material and Methods

Multiple field trapping experiments were conducted to examine the effect of trap design, trap height and different sources of host volatiles on the number of individual

female *S. noctilio* captured in flight intercept traps. All field experiments were deployed in loblolly pine (*Pinus taeda*) stands with varying ages, sizes and levels of *S. noctilio* attack (see Table 1), estimated by sequential sampling methods according to Penteado et al. (2002). In all experiments traps were hung from a rope tied between two adjacent trees. Each trap was at least 1 m from the closest tree and the top of each trap was ca. 2 m above ground (except for the 8 m treatment traps in Experiment 4, which were 8 m above ground). All traps were at least 20 m from the closest trap and 15 m from the stand boundary. All traps were equipped with collection cups filled with 100 ml of 25% salt solution and a few droplets of scentless detergent (Ipê®). Collections were performed approximately every 2 weeks on 03-Nov, 16-Nov, 30-Nov, 07-Dec, 21-Dec 2015 and 04-Jan 2016 in 2015–16 (Experiments 1 & 3) and 15-Nov, 30-Nov, 08-Dec, 22-Dec 2016, 05-Jan, 18-Jan and 08-Feb 2017 in 2016–17 (Experiments 2, 3 & 4). At each collection date, the salt solution and host material lures were replaced. The turpentine and artificial lures were replaced once during Experiment 2, on 05-Jan 2017. Captured material was returned to the laboratory where it was processed, and all *S. noctilio* were identified, sexed and counted.

Effect of Natural Sources of Host Material

Experiment 1 was conducted, from 20-October, 2015 to 4-January, 2016 and tested whether there were any differences in *S. noctilio* captures among panel intercept traps baited with five different sources of host material lures: 1) *Pinus taeda*, 2) *P. elliotti*, 3) *P. eliliodurensis* [a hybrid of *P. elliottii* and *P. caribaea hondurensis* (hereafter Hybrid 1)], 4) *P. tecunumanii* X *P. caribaea hondurensis* hybrid (hereafter Hybrid 2), and 5) blank with no host material. Host material was incorporated into each lure through the use of mesh bags containing host foliage and small pieces of the main bole split into billets with the bark on of each species evaluated. The species/hybrids evaluated were used, because they are the main commercial pine planted in Brazil and are susceptible to *S. noctilio* attack (see below for a description of lure preparation). Panel traps constructed from 3 mm corrugated plastic by Cartonale Ind. e Benef. de Materiais Plásticos Ltda., Santana do Parnaíba, SP, Brazil (the same design and dimensions as the panel trap produced by ConTech Enterprises Inc., Victoria, BC, Canada) were assembled in the field and used for all treatments (see Fig. 1). At each of two sites, traps were deployed in a 5X5 grid consisting of five replicate blocks with one of each of the five bait treatments. Each trap was at least 20 m from its nearest neighbor.

Table 1 Loblolly pine (*Pinus taeda*) stand traits where trapping experiments were deployed

Experiment	Stand age (years)	Stand size (ha)	Sirex attack level (%)
1	14	13	3.5
2	11	56	9.0
3 (2015/16)	14	33	5.0
3 (2016/17)	11	56	9.0
4	11	56	9.0

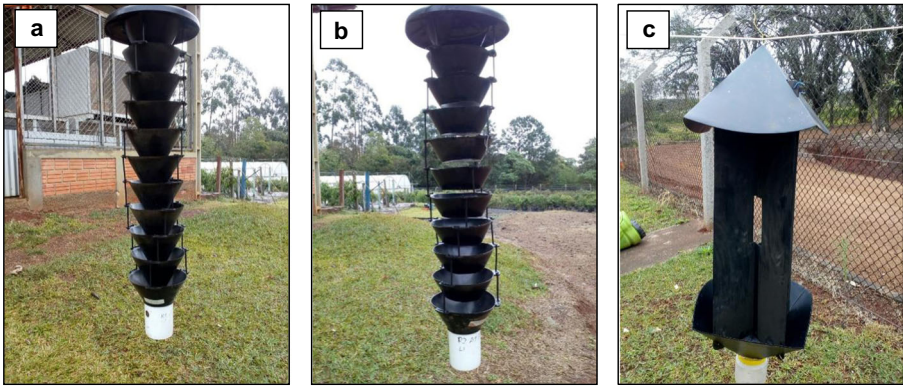


Fig. 1 Flight intercept traps types deployed. **a** Multiple funnel trap. **b** Modified multiple funnel trap. **c** Panel trap

Comparison of Natural and Semi-Defined Sources of Host Volatiles

Experiment 2 was conducted from 26-October, 2016 to 8-February, 2017 and, like Experiment 1, tested whether there were differences in *S. noctilio* captures in panel intercept traps baited with different sources of host material. Captures were compared in panel traps baited with: 1) *P. taeda*; 2) *P. elliottii*; 3) Hybrid 1; 4) turpentine; 5) commercial *S. noctilio* lure; and 6) blank traps with no lure. Host material lures (treatments 1, 2 and 3) were prepared as in Experiment 1. Slash pine (*P. elliottii*) turpentine was provided by Pinus Brasil Agro Florestal Ltda., Buri, SP, Brazil. Approximately 100 ml of turpentine was placed in 300 ml plastic bottles with 3 cm diameter top opening and no lid (Petty Embalagens Ltda., Jaboticabal, SP, Brazil). The bottles were hung outside the top funnel (i.e., under the overhang of the lid) to minimize rainwater falling into the open bottle. The release rate of turpentine from the bottles in the field is $\cong 0.5$ g/d (E. Batista, personal observation). Commercial *S. noctilio* lures were purchased from Alpha Scents Inc., West Linn, OR, United States. At each of two sites, traps were deployed in a 5X6 grid consisting of five replicate blocks with one of each of the six bait treatments. Again, each trap was at least 20 m from its nearest neighbor.

Effect of Trap Design

Experiment 3 was conducted, from 20-October, 2015 to 4-January, 2016 and then again from 26-October, 2016 to 8-February, 2017 and tested whether intercept trap design affected the number of female wasps captured in: 1) panel intercept traps; 2) commercial multiple funnel traps; and 3) modified multiple funnel traps (see Fig. 1 and Miller et al. 2013). Intercept panel traps were identical to those described above and the lure was made of *P. taeda* billets and foliage. Commercial multiple-funnel traps were purchased from ConTech Enterprises Inc. Modified multiple-funnel traps were multiple-funnel traps with the bottom 6 cm of each funnel removed in the laboratory [see Miller et al. (2013) for details] (Fig. 1). During both years at each of the two replicate sites, traps were deployed in a 5X3 grid consisting of five replicate blocks

with one of each of the three trap design treatments. Each trap was at least 20 m from its nearest neighbor.

Effect of Trap Height

Experiment 4 was conducted from 26-October, 2016 to 8-February, 2017, and tested the effect of height of trap placement on *S. noctilio* captures in intercept panel traps baited with *S. noctilio* lures (purchased from Alpha Scents Inc.). Panel traps were deployed in pairs, hung from the same tree with one trap at 2 m and the other at 8 m. At each of two replicate sites, trap pairs were deployed in a 5X2 grid consisting of five replicate blocks, each block with two traps (one at 2 m and one at 8 m, each trap was at least 20 m from its nearest neighbor).

Lure Preparation

To obtain the host material used to make the lures for Experiments 1, 2 and 3, live trees were felled (see Table 2 for stand characteristics). Portions of the main bole were cut into 30 cm sections, which were then split longitudinally into halves. Approximately 165 g of foliage and ten billets (small pieces of the main bole split with the bark remaining) were used to produce a single lure of 1.5 kg (mesh bag containing foliage and billets). Fresh trees were cut every 2 weeks to produce fresh lures, and ca. five lures could be produced from a single cut tree. Lures (0.5 cm × 0.5 cm mesh size; 1 m × 0.5 m bag size) were hung adjacent to the traps from the same rope on which the traps were suspended.

Statistical Analyses

In all experiments, captures were low resulting in many with less than five individuals per trap. Consequently, we were unable to analyze for the effect of stand or date regardless of experiment, and count data were pooled across date and stand for each treatment within each experiment. For each experiment, count data were analyzed using a standard G-test (Sokal and Rohlf 1981). For Experiments 1 and 2, no wasps were captured in the unbaited traps, and these treatments were not included in the analyses.

Table 2 Pine stands age and diameter at breast high (DBH) from where host material were collected to lure composition

Pine species / hybrid	Age (years)	Average DBH (cm) ^a
<i>Pinus taeda</i>	7	14
<i>Pinus elliottii</i>	7	14
Hybrid 1 (<i>P. elliottii</i> x <i>P. caribaea hondurensis</i>)	5	10
Hybrid 2 (<i>P. tecumumanii</i> x <i>P. caribaea hondurensis</i>)	5	10

^a only for felled trees

Results

In total 202 *S. noctilio* individuals, all females, were captured and included in statistical analyses in experiments 1–4 (51, 80, 36, and 62 individuals respectively). Only Experiment 1, which compared different sources (i.e., *Pinus* spp.) of host material as a lure, showed a significant treatment effect (Table 3). Traps baited with lures containing Hybrid 1 consistently captured the most individuals. This pattern was not repeated in Experiment 2 in which a commercial lure was used. Experiments 2–4 (comparing lure, trap design and trap height, respectively) did not observe any treatment effects.

Discussion

This study examined the effect of trap type, height and different lures on the capture of *S. noctilio* in Brazilian pine plantations. The number of wasps captured was low, likely due in part to the low population levels in the stands selected for field trapping experiments. All of the stands had previously received applications of the nematode biological control agent *Deladenus siricidicola*. Despite the low capture numbers, this study observed a significant effect of lure treatment in Experiment 1 and that only traps baited with host material or commercial lure captured wasps.

Immature life stages of endophytic insects like *S. noctilio* cannot migrate among potential host plants. Consequently, their reproductive success is a function of the suitability of the host plant as perceived by host-seeking females, and the realized suitability of the host plant for larval development (Jaenike 1978; Thomson 1988; Mayhew 1997). Several studies have demonstrated that volatiles emitted by and characteristic of stressed pines are attractive to female *S. noctilio* (Simpson and McQuilkin 1976; Simpson 1976; Böröczky et al. 2012; Hurley et al. 2015). Trap trees are effective for monitoring low-density populations of *S. noctilio* (Madden 1971; Penteado et al. 2002). It has been demonstrated that herbicide-stressed trees emit larger quantities of volatiles and are more attractive to female *S. noctilio* than untreated

Table 3 Female individual *Sirex noctilio* captures by experiment and treatment

Experiment 1		Experiment 2		Experiment 3		Experiment 4	
Lure	Captures	Lure	Captures	Design	Captures	Height	Captures
<i>P. taeda</i>	5	<i>P. taeda</i>	13	Panel	18	2 m	27
<i>P. elliotii</i>	11	<i>P. elliotii</i>	16	Multiple funnel	8	8 m	35
Hybrid 1	22	Hybrid 1	13	Modified multiple funnel	10		
Hybrid 2	13	Turpentine	11				
Blank	0	Commercial	27				
		Blank	0				
Total	51	Total	80	Total	36	Total	62
Expected	12.75	Expected	16	Expected	12	Expected	31
G-test	11.898	G-test	9.215	G-test	4.463	G-test	1.035
<i>p</i>	0.008	<i>p</i>	0.056	<i>p</i>	0.107	<i>p</i>	0.309

control trees (Böröczky et al. 2012). Field trapping studies have demonstrated that traps baited with synthetic host volatile blends capture more female *S. noctilio* than unbaited traps (Hurley et al. 2015). Barnes et al. (2014) observed that traps baited with fresh host material captured significantly more wasps than traps baited with commercial synthetic lures. Cumulatively, these results suggest that these types of volatiles are involved in the pre-alightment evaluation of potential oviposition sites by female *S. noctilio*. Post-alightment evaluation of host suitability by females involves olfaction as well as evaluation of the host during probing of the host with the ovipositor. Females are reported to reject host pines with high osmotic pressure in the phloem tissue and accept those with low osmotic pressure (Cutts and Dolezal 1969; Madden 1974; Madden and Cutts 1979). Although no pines appear to be resistant to attack from *S. noctilio* (Ryan and Hurley 2012), differences in *S. noctilio* rates of attack and emergence among pine host trees have been observed (Dodds et al. 2010; Eager et al. 2011; Ryan et al. 2012; Zylstra and Mastro 2012; Ayres et al. 2014; Foelker 2016; Foelker et al. 2016; Haavik et al. 2017). Further, tree resistance is an important determinant of early instar larval survival (Haavik et al. 2015). Patterns of oviposition and larval development reported for *S. noctilio* in *P. sylvestris*, *P. resinosa*, *P. banksiana* and *P. strobus* do not support the hypothesis that female perceived suitability of host pines is adaptive for larval development.

Intercept traps baited with semiochemicals are among the most effective tools available for survey and detection of forest insects (Dodds et al. 2000; Brockerhoff et al. 2006; Johnson et al. 2013; but see Mercader et al. 2013). Numerous trap design factors have been reported to affect intercept trap performance for woodboring insects (Allison et al. 2014; Allison and Redak 2017) and several studies have compared trap designs for woodwasps, including *S. noctilio*. While some studies report no effect of trap design on capture (Costello et al. 2008; Barnes et al. 2014; Haavik et al. 2014; Hurley et al. 2015) others did observe an effect (McIntosh et al. 2001; Bashford 2008). Allison and Redak (2017) conducted meta-analyses of the literature on trapping bark beetles, woodboring insects and their associates and reported that panel traps captured significantly more woodwasps than multiple-funnel traps. Sarvary et al. (2015) observed that female *S. noctilio* could be captured by both black intercept traps and a clear jar trap when both were baited with the putative male-produced pheromone (Cooperband et al. 2012; but see Hurley et al. 2015). When traps were not baited with the putative pheromone, only the black intercept traps captured females. Most studies that have reported no effect of trap type, including this one, have captured few insects. It is possible that patterns of trap design effects are not independent of the olfactory stimuli presented with traps.

Madden (1974) looked at the distribution of oviposition drills by *S. noctilio* on the bole of standing trees successfully and unsuccessfully attacked. On trees successfully attacked, oviposition tended to occur randomly on the tree bole with a tendency to become uniform. Conversely, on trees that resisted attack, oviposition tended to become aggregated suggesting variation in suitability/acceptability within individual trees among tissues. While oviposition occurs along the entire bole in young (i.e., small) trees, it occurs between 3 and 14 m above ground in older trees (Neumann et al. 1982). Observational studies of female *S. noctilio* oviposition report that oviposition is initiated near the base of the tree and that subsequently females work their way up the bole ovipositing every 7.5–50 cm (Rawlings and Wilson 1949; Morgan and Stewart

1966). Male *S. noctilio* are thought to form leks in the canopy of pine forests and that after emergence females are hypothesized to fly to the upper canopy where mating occurs (Morgan and Stewart 1966; Madden 1988). Cooperband et al. (2012) reported the identification of a male-produced pheromone that attracted both sexes in Y-tube olfactometer trials and speculated that it functioned to mediate lek formation among males and attraction of virgin females to leks. Consistent with this hypothesis, male wasps have been captured at higher heights than females, and when males were present in traps, females tended to be captured at higher heights (Martinez et al. 2014). Conversely, this study and Hurley et al. (2015) observed no effect of trap height on the capture of female *S. noctilio*. One potential explanation for the reported differences in trap height effects is that studies that report no effect of trap height baited traps with host volatiles while the study of Martinez et al. (2014) used unbaited sticky traps. Depending on the mating frequency of females it is possible that virgin and mated females orient along different flight paths with respect to the forest canopy. Virgin females may orient to the upper canopy in response to a light gradient and/or a male pheromone and mated females may orient to the lower bole in response to host volatiles.

Although trap trees can be effective for sampling *S. noctilio* they are expensive and labor intensive to use and can involve the use of herbicide. The development and use of a trap-lure system in place of trap trees has resulted in a 25–38% cost reduction in Tasmania, and semiochemical-baited intercept traps are used in other parts of Australia where *S. noctilio* is not yet widely present (Bashford and Madden 2012; Carnegie and Bashford 2012). Although trap captures were low the results of this study suggest that although host trees vary in their perceived suitability to host-seeking females (Exp. #1 but not Exp. #2), the synthetic lures available commercially in North America are at least as effective as host material in Brazil. Multiple-funnel and panel traps were equally effective sampling *S. noctilio* in Brazilian pine plantations. Given that panel traps are less expensive, survey and detection programs may opt for this trap design. Finally, this study observed no differences in the capture of *S. noctilio* females in traps placed at 2 and 8 m height. Future studies should explore the role of visual stimuli in the orientation behavior of male and female *S. noctilio* as well as the context-dependence of visual and olfactory stimuli.

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