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Survey of Subterranean Termite (Isoptera: Rhinotermitidae) Utilization of Temperate Forests

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Abstract

Both native and invasive subterranean termites (Isoptera: Rhinotermitidae), including the Formosan subterranean termite, are well known pests of urban areas, but little is known about their distribution or impact in forest ecosystems of the southeastern United States. Recently harvested timber stumps were mechanically inspected for the presence of subterranean termites in multiple locations across southern Mississippi and eastern Louisiana. A systematic line plot cruise with 100 x 200m spacing and 1/20th ha plots was implemented, and all stumps with a diameter greater than 7.6cm were inspected. In total, 7,413 stumps were inspected for the presence of subterranean termites, and 406 of those contained native subterranean termites (*Reticulitermes* spp.). Light traps were also placed at 8 sites to detect the presence of subterranean termite alates. While no invasive Formosan subterranean termites were found during mechanical inspection of tree stumps, alates were captured in light traps at three sites. The proportion of stumps infested with subterranean termites was negatively correlated with the number of stumps in each plot. Although 6.27% of pine stumps and 1.86% of hardwood stumps contained subterranean termites, no correlation was found between subterranean termite presence and type of stump (pine or hardwood) inspected. Subterranean termite presence in stumps ranged from 0.94% to 14.97% depending on site.

Introduction

Termites are ecologically and economically important insects, which can be found in nearly all forest ecosystems. Subterranean termites (Isoptera: Rhinotermitidae) play important roles in cellulose decomposition, nutrient cycling, and soil mineralization across a multitude of environments (Harris, 1966; Wood & Sands, 1978; Black & Okwakol, 1997). Although they are known to cause significant economic damage to wooden structures in urban areas throughout the U.S., native subterranean termites (*Reticulitermes* spp.) provide a valuable service to forest ecosystems as the predominant invertebrate decomposers of woody materials (La Fage & Nutting, 1978). The role of subterranean termites in forest ecosystem nutrient cycles is extremely important, and their presence can contribute up to 22% of total nitrogen input in tropical forest ecosystems (Yamada et al., 2006).

The non-native Formosan subterranean termite (*Coptotermes formosanus* Shiraki), is reported to infest 17 species of living trees (Chambers et al., 1988) and at least forty other species of plants (Lai et al., 1983). Formosan subterranean termites are known to cause significant economic damage to standing timber in many regions of the world (Harris, 1966). Greaves et al. (1967) reported that *Coptotermes* spp. were responsible for up to 92% of tree losses in virgin eucalyptus forests and approximately 64% of losses in younger forests, which were predisposed to fire stress. While studies have investigated subterranean termite ecology in tropical forest ecosystems, little is known about their ecology in temperate forests.

Loblolly pine (*Pinus taeda* L.) is a predominant species in the southern U.S., comprising 45% of commercial forest-land and contributing $30 billion annually to the economy (Schultz, 1999). Considering the large industry for loblolly pine in the southeastern U.S. and the lack of research on Formosan subterranean termites in forested areas, the impact of this invasive insect could be substantial. For instance, native subterranean termites, which are ubiquitously reported in the

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literature to rarely infest living trees, have been observed readily utilizing blue-stained portions of bark beetle-attacked trees before any foliage chlorosis could be detected (Little, 2013). While Formosan termites commonly infest living trees in urban settings, loblolly pines are more frequently infested than other tree species (Osbrink et al., 1999; Guillot et al., 2010). Additionally, Morales-Ramos & Rojas (2001) observed that Formosan subterranean termites consumed loblolly pine wood at a higher rate relative to other wood species in laboratory no-choice tests.

Biological differences between Formosan and native subterranean termites create the possibility that Formosan subterranean termites could impact forest ecosystems and individual trees differently than our native subterranean termites. For example, Formosan subterranean termite colonies are considerably larger and they are more aggressive feeders than native species (Su & Scheffrahn, 1988). Formosan subterranean termites also exhibited higher survival and wood consumption rates than native *Reticulitermes* spp. in laboratory assays (Smythe & Carther, 1970). They can also nest above-ground with no connection to the soil, which allows them to utilize resources previously inaccessible to native species. La Fage (1987) noted that in certain instances in New Orleans, LA, Formosan termites completely replaced native termites as the predominant termite species. More recently, Su (2003) confirmed that Formosan subterranean termites were outcompeting native *Reticulitermes* spp. in multiple urban locations.

Despite these concerns, little research has been done on the distribution or impact of Formosan subterranean termites in non-urban forested environments of the U.S. The first indication that Formosan subterranean termites were established in forested areas of the southeastern U.S. was when Sun et al., (2007) reported that Formosan subterranean termite alate catches were higher in forested settings than in urban areas. In addition, alate catches increased throughout the four year trapping study, which may have indicated that Formosan subterranean termite populations were well established and colony expansion was occurring in Mississippi (Sun et al., 2007).

Native and Formosan subterranean termite ecology in local forested settings of the southern U.S. has yet to be quantified. We hypothesized that elevated light trap captures of Formosan subterranean termite alates in rural forested areas by Sun et al. (2007) would be explained by infestations of living trees in surrounding forest stands. Therefore, the objectives of this study were to determine the extent of subterranean termite utilization of living trees within localized forested settings, quantify subterranean termite presence in forested areas, determine if the proportion of stumps infested with subterranean termites was correlated with stump density or size, and quantify the effect of tree type (hardwood or softwood) on subterranean termite utilization of woody resources.

Material and Methods

Stump Inspections

Eleven sites encompassing 476.8ha throughout southern Mississippi and Louisiana were inspected during 2011 and 2012 (Fig 1). There were nine sites surveyed in four Mississippi counties (Pearl River, Harrison, Jackson, and Lamar) and two additional sites were surveyed in St. Tammany Parish, Louisiana (Table 1). According to Sun et al. (2007), the four Mississippi counties inspected during this study were among the top five counties in the state for Formosan subterranean termite alate captures. Additionally, Formosan subterranean termites have been found in St. Tammany Parish, Louisiana since the late 1980s (La Fage, 1987; Messenger et al., 2002).

Clear cut pine plantations were inspected for subterranean termite presence in residual stumps within three weeks of harvest because they offered the best opportunity to discover the presence of subterranean termites in living trees without damaging standing timber. A variety of methods have been used over the years to detect subterranean termites in living trees (e.g. acoustic devices, dogs, CO_2 detectors, coring, etc.); however, most have limitations, and are often less effective than physical inspections with hatchets and shovels (Osbrink et al., 1999). Site selection criteria (recent timber harvest and proximity to established Formosan subterranean termite populations) led to sporadic site availability; therefore sites were inspected as they became available. To limit the possibility of post-harvest colonization of stumps, all inspections took place within three weeks of timber harvest; however, this only oc-

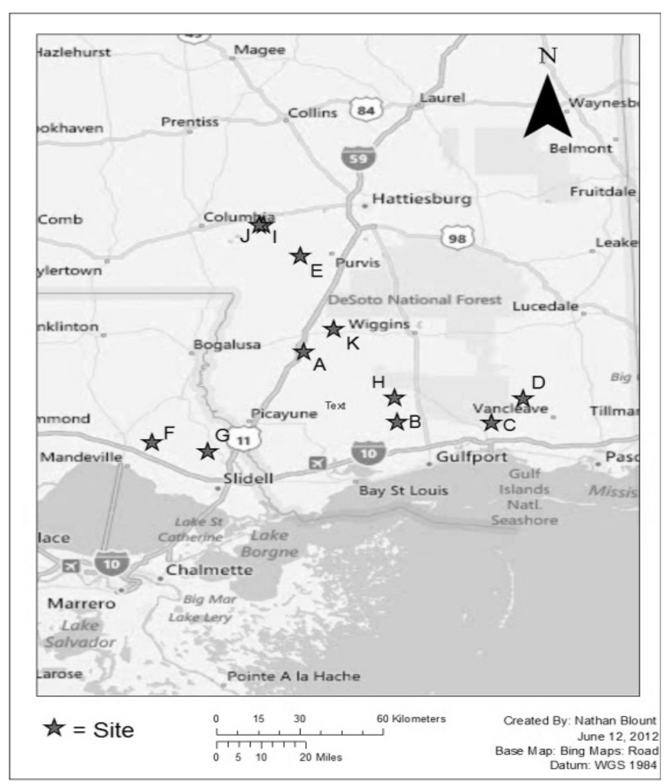

Fig 1. Map of inspecion sites.

curred for one site. The majority of sites were inspected while logging was still occurring, with the balance being inspected within a week of harvest. A 2.5% systematic line plot cruise (Avery & Burkhart, 2002) was implemented on post-harvest sites that were 20ha and larger to insure adequate sampling across each site and to limit sampling bias (with the exception of the first site inspected, A). Sites smaller than 20ha received a 5% cruise. Plots were 1/20th ha and circular, with 100m between plots and 200m between transects (Fig 2). A global positioning system (GPS) unit (GPSMAP 60CSx®, Garmin Ltd., Olathe, KS) was used to record plot centers.

All stumps greater than 7.6cm in diameter and located within plots were inspected with shovels and hatchets. Stumps less than 7.6cm in diameter were not sampled based on findings from prior studies, which indicated that native subterranean termites primarily occur in coarse woody debris with diameters above 7.6cm (Wang & Powell, 2001; Wang et al., 2003). One-quarter of each stump was inspected for subterranean termite presence from the cut surface of the stump down to 15cm below the soil line. This method has been previously used to successfully detect infestations of Formosan subterranean termites in living trees throughout urban areas, even when visual symptoms were not present (Osbrink et al., 1999). If termites were present in stumps, they were immediately identified to genera, *Reticulitermes* (native) or *Coptotermes* (Formosan), using the distinct morphological characteristic of head shape in the soldier caste (Gleason & Koehler, 1980). Stump diameter was recorded for each occurrence of termites in stumps, which was subsequently labeled as hardwood or pine (softwood). Each stump was then marked and labeled on a GPS unit.

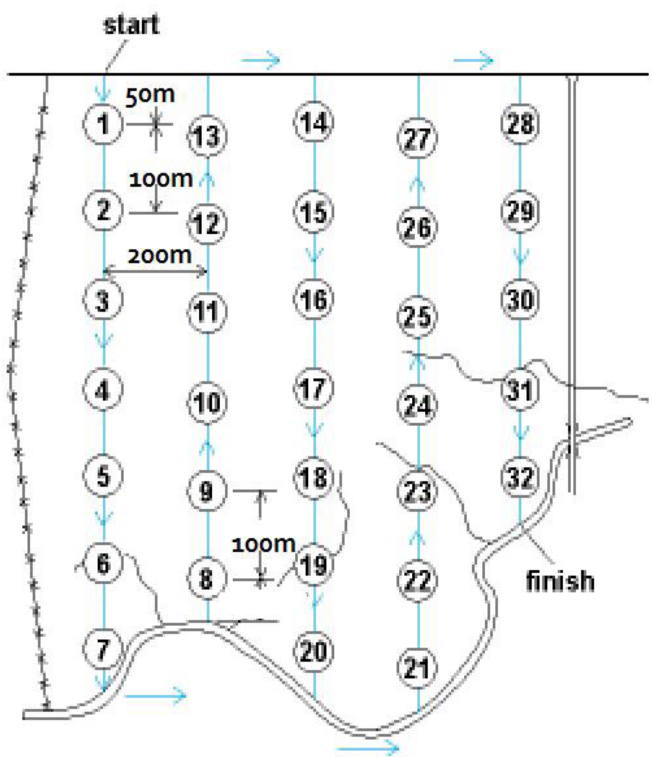

Fig 2. Example of systematic line plot cruise.

Alate Survey

Light traps were used during spring of 2012 to confirm the presence of Formosan subterranean termite alates near sites selected for physical inspections. Methods similar to Sun et al. (2007) were used to construct alate traps to ensure proper design. Traps were constructed using a 1.52m (5-foot) t-post, white polyvinyl chloride (PVC) pipe, 20ga 2.54cm (1-inch) poultry netting (Garden Plus), a solar powered light emitting diode (LED) light (model SPS2-P1-BK-T24, LG Sourcing Inc., N. Wilkesboro, NC), and a glue board (Trapper® LTD, Bell Laboratories, Inc., Madison, WI) (Fig 3).

To avoid non-target catches of vertebrates attempting to prey on captured insects, poultry netting was wrapped around the glue board and t-post to achieve cage dimensions of 16.5 (9.5-inches) x 35.6cm (14-inches). A wire top was fashioned to give easy access for glue board replacement. The t-post was driven into the ground to insure firm placement of the trap. The LED light came on approximately 20 min after sunset, and ran for six hours throughout the trapping season,

Table 1. Summary of Sites Inspected for This Study

Site	County/Parish	Hectares	% Cruise	GPS Coordinates	Soil Conditions	Site Designation
A	Pearl River	83.0	5	N30° 46’ 58” W89° 29’ 56”	Dry	Upland
B	Harrison	89.4	2.5	N30° 31’ 25” W89° 11’ 47”	Dry	Upland
C	Harrison	117.4	2.5	N30° 31’ 43” W88° 53’ 22”	Wet	Lowland
D	Jackson	25.9	2.5	N30° 36’ 43” W88° 47’ 17”	Dry	Upland
E	Lamar	13.4	5	N31° 8’ 23” W89° 30’ 52”	Moderate	Upland
F	St. Tammany	39.3	2.5	N30° 27’ 10” W89° 59’ 37”	Moderate	Lowland
G	St. Tammany	19.8	5	N30° 25’ 1” W89° 48’ 45”	Moderate	Lowland
H	Harrison	8.5	5	N30° 37’ 3” W89° 12’ 30”	Moderate	Lowland
I	Lamar	19.4	5	N31° 15’ 13” W89° 37’ 49”	Moderate	Upland
J	Lamar	39.3	2.5	N31° 15’ 11” W89° 38’ 20”	Moderate	Upland
K	Pearl River	21.4	5	N30° 52’ 5” W89° 24’ 16”	Moderate	Upland

Fig 3. Formosan subterranean termite alate light trap.

since peak Formosan subterranean termite alate flight activity occurs at dusk (Bess, 1970).

Formosan subterranean termite alates have been documented to swarm as early as mid-April in Mississippi, with peak activity often occurring near the latter part of May (Sun et al., 2007; Lax & Wiltz, 2010). Traps were placed on sites at the end of the first week in April, early enough to detect initial swarms. Glue boards were replaced once a week for a seven week period, with a final collection date of May 26. This sampling window allowed ample time to catch alates during their swarming periods determined by previous studies (Sun et al., 2007; Lax & Wiltz, 2010). All glue boards were dated and labeled by site when removed, wrapped in plastic wrap, and placed in a freezer for preservation. Alates were identified in the laboratory using morphological characteristics provided by Gleason & Koehler (1980).

A total of twelve alate light traps were placed on eight sites. The Lamar County sites (E, I, & J) were excluded from the alate trapping portion of this study due to travel constraints. Additionally, Lamar County had a low number of alate captures during a previous study (Sun et al., 2007) relative to all other counties sampled in Mississippi. The number of traps placed per site was determined by area, with five sites receiving one trap each (<40.5ha), two sites receiving two traps each (83 and 89.4ha respectively), and one site receiving three traps (117.4ha). Traps were placed at the approximated center of each site. On sites with multiple traps, the location was partitioned according to area, with traps located in the approximate center of each partition.

Statistical Analyses

The proportion of stumps infested with termites was compared to stump density and site using the BEINF family of the GAMLSS package (Rigby & Stasinopoulos, 2001; Stasinopoulos & Rigby, 2007; Stasinopoulos et al. 2009), in the R statistical package (R Core Team, 2012). While an arcsine transformation is commonly used for proportional data, this transformation has a number of limitations and has been superseded by a variety of alternative techniques (Warton & Hui, 2011). A zero-inflated distribution was required because the data demonstrated a bimodal distribution with an excess of zeros (Fig 4). The BEINF family of GAMLSS models the data using a beta distribution and allows for inflation of both zeros and ones. Inflation at both zero and one was chosen over zero-inflation alone because it was at least theoretically possible that all stumps in a plot could be infested with termites. This model assumes that there at least two processes that combined to produce the original data. The beta distribution models the range of proportion infestations between zero and one, while a binary process models an excess of plots with no infestations due to another process. The high number of plots with zero infestations and the potential for numerous infested trees within infested plots is potentially explained by the social/colonial nature of subterranean termites. Subterranean termite workers only feed within a finite distance of the colony. This biological process likely explains the zero-inflated distribution of our data.

Pearson's correlation coefficients (PROC CORR) were used to assess the relationship between infested pine and infested hardwood stumps in SAS 9.2 (SAS Institute, Cary, NC). Significance for all analyses was determined at α≤0.05.

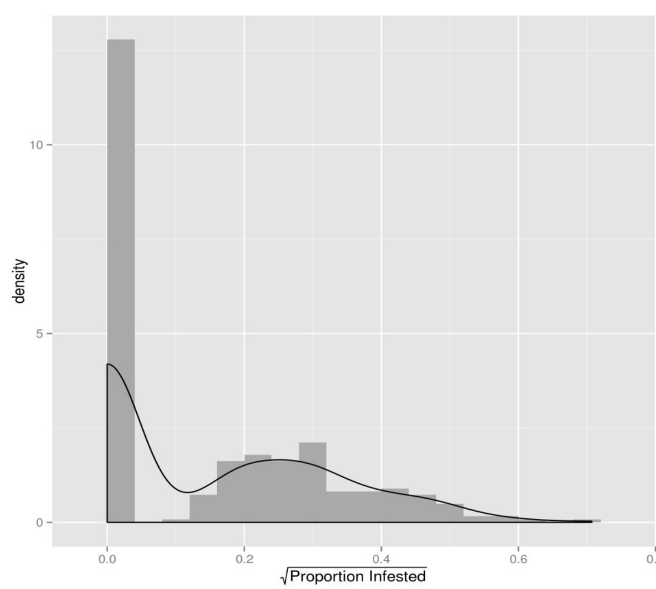

Figure 4. The distribution of the square root transformed proportion of stumps in a plot infested with termites compared to a kernel density distribution for the same variable. An excess of zeros is indicated by the large bar corresponding to zero infested stumps.

Results and Discussion

Stump Inspections

A total of 7,413 stumps were inspected, consisting of 6,072 softwoods (hereafter pines) and 1,341 hardwoods. No Formosan subterranean termites were found; however, 406 stumps containing *Reticulitermes* spp. were recorded, resulting in 5.48% of all stumps containing subterranean termites. These included occurrences in 381 pine (6.27% of total pines) and 25 hardwood stumps (1.86% of total hardwoods) (Table 2). Site F had the highest overall occurrences of *Reticulitermes* spp., with 14.97% of all stumps containing subterranean termites. Sites C and G followed, with 10.81% and 7.42% of stumps containing subterranean termites, respectively. Site I had the fewest subterranean termite occurrences among all sites with 0.94% of stumps containing termites, followed closely by sites D (1.33%) and K (1.58%).

Several sites had low occurrences of hardwood stumps. No hardwood stumps were encountered during inspections on site K, while site B had only 3 hardwood stumps in the plots. Sites D and F also had low numbers of hardwood stumps, 16 and 33 respectively. Five sites contained hardwood stumps that had no subterranean termites (B, D, H, I, and J).

For further analysis and uniformity, stumps that contained *Reticulitermes* species were converted to a per hectare basis using a conversion factor relative to the percent cruise conducted on each site. Overall, the mean number of subterranean termites per hectare across all stumps and sites was 27.41 (se=0.28, n=7,413). Mean number of pine stumps containing subterranean termites per hectare was 26.36 (se=0.30, n=6,072), with hardwood stumps at 1.05 (se=0.06, n=1,341). Subterranean termite occurrences per hectare for all stumps on a per site basis ranged from 5.06 (site A) to 79.39 (site F). Sites A, B, and I had fewer than 7.16 occurrences per hectare. Site C had the second highest occurrence of subterranean termites per hectare, 65.08, followed by site G, 38.72. Subterranean termite occurrence in hardwood stumps per hectare for site C was 7.16, which was the only site with more than 1.68 infested hardwood stumps per hectare.

Table 2. Summary of Stumps Containing Subterranean Termite Infestations by Site.

	Stumps Inspected			Stumps Infested			% Infestation		
Site	[1]P	[2]H	Total	[1]P	[2]H	Total	[1]P	[2]H	Total
A	530	360	890	20	1	21	3.77	0.28	2.36
B	411	3	414	16	0	16	3.89	0.00	3.86
C	1499	268	1767	170	21	191	11.34	7.84	10.81
D	585	16	601	8	0	8	1.37	0.00	1.33
E	181	166	347	15	1	16	8.29	0.60	4.61
F	488	33	521	77	1	78	15.78	3.03	14.97
G	258	52	310	22	1	23	8.53	1.92	7.42
H	223	204	427	11	0	11	4.93	0.00	2.58
I	304	121	425	4	0	4	1.32	0.00	0.94
J	387	118	505	19	0	19	4.91	0.00	3.76
K	1206	0	1206	19	0	19	1.58	0.00	1.58
∑	**6072**	**1341**	**7413**	**381**	**25**	**406**	**6.27**	**1.86**	**5.48**

[1]P = Pine; [2]H=Hardwood

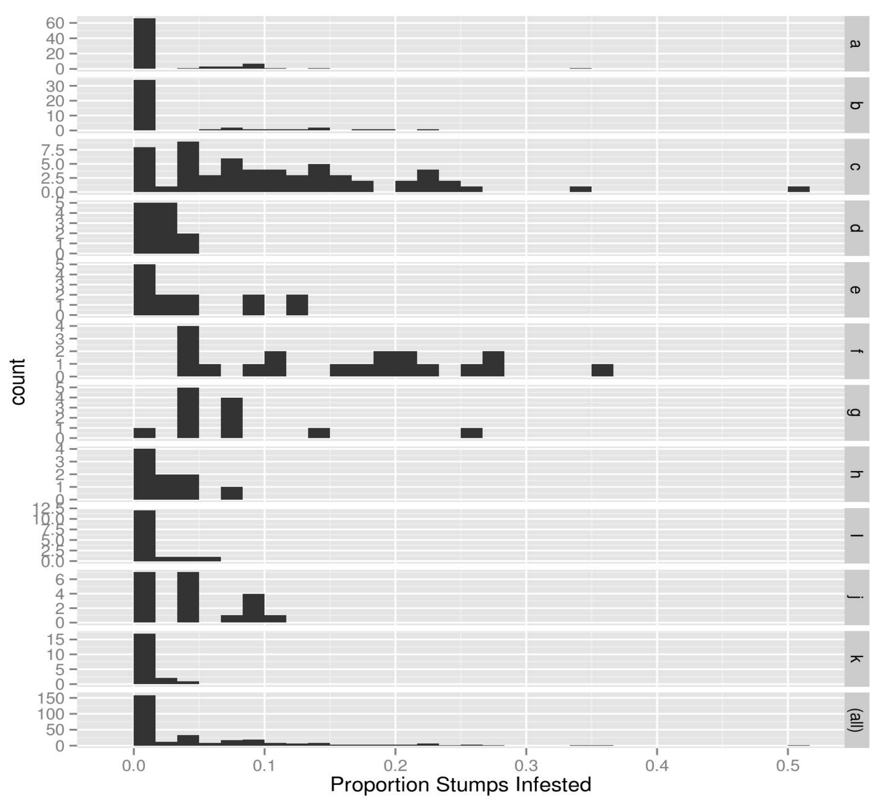

Fig 5. The distribution of the proportion of stumps in a plot infested with subterranean termites by site.

The distribution of the proportion of stumps infested varied with site (Fig. 4). In the inflated beta distribution analysis, both stump density (χ^2 = 8.49, df = 1, P = 0.0035) and sites (χ^2 = 38.42, df =10, P = 3.21e-5) were significant. These data suggest that across the range of overall densities experienced at various sites, native termite species infested a greater proportion of stumps in plots with lower stump densities and presumably larger stumps (Fig 6). In this agroforestry setting, lower stump densities indicate an older forest, with larger diameter trees. The BEINF family of GAMLSS fits four parameters. Two are the shape parameters for the beta distribution (μ = 0.1201, σ = 0.1942), while the other two are related to the probability of the additional processes leading to zero or one values (ν = 1.047, τ = 3.77e-09). These resulted in predicted probability densities of 0.511 at zero and 1.842e-9 at one.

The frequency of subterranean termite infestations in hardwood stumps was significantly correlated with the frequency of infestations in pine stumps across all sites (Pearson r = 0.68, P = 0.03). In other words, sites with higher termite abundance had more infestations of both pine and hardwood stumps. However, the frequency of infestation was always higher in pine stumps than in hardwood stumps. Our results agree with previous studies (Osbrink et al., 1999; Guillot et al., 2010) that reported subterranean termite prefer loblolly pine over hardwoods.

Although no Formosan subterranean termites were found in stumps of recently logged forested stands in Mississippi and Louisiana during this study, native *Reticulitermes* spp. were found across all sites inspected. Subterranean termite were more prevalent in pine stumps than hardwoods. Additionally, our results confirmed findings from previous studies, which documented that the number of subterranean termite occurrences decreased with increasing wood hardness, with pines generally having higher occurrences of termites than hardwoods (Lin, 1987; Peralta et al., 2004).

Alate Survey

During the seven week trapping period a total of 14 Formosan subterranean termite alates were caught among three sites (Fig 5). Two alates were detected on the May 11 trap check of site A in Pearl River County. On May 12, one alate was present on the site D trap in Jackson County. Alates were not present again until the May 25 check, when 10 alates were caught on site A and one alate was present on site G within St. Tammany Parish, LA. Both traps on site A caught alates during this study. Since *Reticulitermes* spp. primarily swarm during daylight hours, no native termite alates were captured during this study. Due to limited alate catches, no statistical analyses were performed.

Alates of Formosan subterranean termites were captured at three of the sites sampled. Site A had multiple alate captures among two traps, while only one alate was caught on

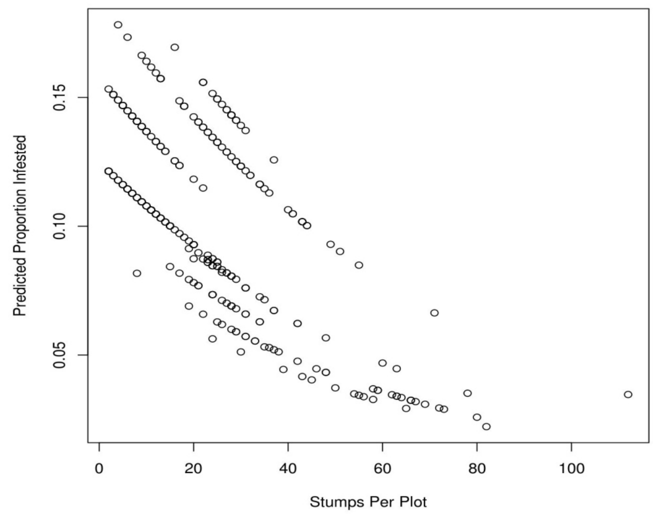

Fig 6. The predicted proportion of stumps infested in a plot based on site and stump density (the total number of stumps in the plot).

each of the other two sites. Formosan subterranean termite alates have the capability to disperse distances of nearly 900m in open areas (Messenger & Mullins, 2005); however, other studies have shown flight distances of 100m to be more common (Higa & Tamashiro, 1983). Although it is unknown if Formosan subterranean termite colonies were located on the inspection sites where alates were caught, it is likely that colonies were present within close proximity if common dispersal distances are taken into account. Additionally, since two sites only had an individual alate captured, it is possible that these alates dispersed a greater distance than the site with multiple alate captures. Multiple alates were captured on two different traps at site A, therefore it was likely that these traps were closer to parent colonies than the traps at other sites. It is also possible that Site A contained a larger colony or greater number of colonies than the other sites. Site D, which was located in a very rural location and completely surrounded by forest, only captured a single alate. The site was over 1.6km (1-mile) away from the nearest house or residential area. This increases the likelihood that the alate dispersed from an established colony in a forested area. Formosan subterranean termite populations may not currently be high enough in forested settings for easy detection, but our capture of them at secluded sites indicates that colonies may already be established in localized forested areas.

There are multiple plausible reasons as to why no Formosan subterranean termites could be located during physical stump inspections in our study. Formosan subterranean termites are known to commonly utilize living trees in urban areas (Osbrink et al., 1999). Urban environments, in general, contain far less downed woody debris than most forested areas. Less woody debris would concentrate subterranean termites into fewer resources, making them easier to detect. Soil moisture levels were also reduced during the hot summer months, which may have caused subterranean termites to travel deeper into the soil to prevent desiccation, thus avoiding detection.

Additionally, the trees on the sites inspected were mature prior to harvest, possibly limiting the import of any significant amount of infested woody debris for decades.

Given the propensity of Formosan termites for attacking living trees in urban trees in the U.S. and plantation forestry settings in other parts of the world, additional research is needed to ensure that Formosan subterranean termite populations do not pose a threat to native forests. More forested sites need to be inspected for the presence of Formosan subterranean termites, utilizing stricter site criteria such as distance to railroads and major roadways, documentation of previous alate trap catches, and proximity to urban areas. Imposing these additional requirements on inspection sites should increase the probability of locating Formosan subterranean termites in forested areas. However, due to the difficulty of locating potential sites, it may take several years to adequately complete the investigation. Continuation of alate monitoring within forested and rural areas is also very important as data collected can be useful in monitoring alate populations and range expansion. Further confirmation of Formosan subterranean termite establishment in localized forest settings would also raise the importance of determining feeding preferences and the likelihood of native subterranean termite displacement in forests, issues this study was not able to address.

We found no Formosan subterranean termites during physical stump inspections; however, only pine plantations were inspected during this study. Hardwood bottoms and older forests with less active management and higher proportions of downed woody material (DWM) may harbor Formosan subterranean termites. Furthermore, living trees might only be infested once populations in DWM are high enough and competition for woody resources is strong, a possibility that should be explored in future research. It is still debatable why more alates were caught in rural areas than in forested settings by Sun et al. (2007). Formosan subterranean termites may utilize DWM in rural forested areas. Another possibility is that light pollution may decrease alate trap efficacy in urban areas. As a consequence, traps placed in urban settings may underestimate alate numbers.

At the conclusion of this study, native *Reticulitermes* spp. were found in recently harvested timber stumps at the eleven sites inspected, but the introduced Formosan subterranean termite was not found. However, Formosan subterranean termites were located within the general area of 3 sites, as evidenced by the alate trap catches and proximity to homes. Due to lack of findings on inspected sites, feeding preference data for Formosan subterranean termites could not be collected, and native *Reticulitermes* spp. displacement is not a current problem within the study areas. Stump inspection data did reveal that *Reticulitermes* spp. occurred in a higher percentage of pine stumps than in hardwoods, but stump type did not significantly affect the number of occurrences per site. The proportion of stumps infested by native subterranean termites was negatively correlated with the number of stumps on a given plot. It is important to note that Formosan subterranean termites could be impacting forested stands in other locations, even though they were not located on the tracts inspected during this study. With rising populations, the invasive Formosan subterranean termite could still pose a major ecological threat to forested settings and native termite species, issues that need to be addressed with continued research.

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