

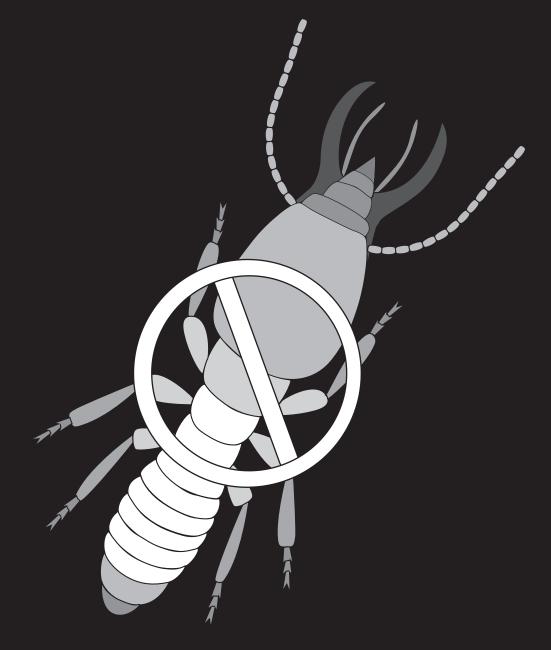
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# Areawide Management of Subterranean Termites in South Mississippi Using Baits



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Use of baits for areawide suppression of formosan subterranean termites (*Coptotermes formosanus* Shiraki) and native subterranean termites (*Reticulitermes flavipes* (Kollar) and *R. virginicus* (Banks)) was evaluated under field conditions at Keesler Air Base Force, Biloxi, MS. This test was designated to evaluate the feasibility of controlling subterranean termites by an areawide management strategy using a nutritionally based bait matrix to carry the active ingredient. The active ingredient, diflubenzuron, was delivered via the USDA-ARS bait matrix formulation. Quarterra interception and baiting systems were used for bait delivery. Test sites were selected based on 2,000 alate data. The test area consisted of 12 neighborhoods: 6 treated and 6 controls. One hundred and fifty stations per site were installed in a grid pattern around structures, trees, and shrubs. Every fifth station in each site was designated as an independent monitor. Pair comparison between treatment and control was done. Temperature, termite activity, and bait consumption were measured monthly from April 2001 to October 2003. Formosan subterranean termite activity stopped in April 2004.

**Keywords:** APM, bait, control, *Coptotermes formosanus*, formosan, pest, subterranean, termite.

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### Introduction

The formosan subterranean termite (*Coptotermes formosanus* Shiraki) is considered one of the most destructive urban pests in the United States, causing millions of dollar in losses (Su and Tamashiro 1987, Su and Scheffrahn 1990). Annual control cost estimates are around \$1,200 million (Su 1994, Su and Scheffrahn 1998) without taking into account expenses for repair of damaged structures.

The formosan termite is believed to have originated in continental China (Kistner 1985, Beal 1987). Kistner based his analysis on the presence of Sinophilus xiai (Coleoptera: Staphylinidae), which is associated exclusively with C. formosanus and has otherwise been found only in the People's Republic of China. Formosan subterranean termites were introduced into the United States at the end of World War II (Beal 1987) via infested wood in cargo ships and were officially recorded for the first time in 1965 in Houston, TX (Beal 1967). The same year, formosan subterranean termites were detected in Louisiana (New Orleans and Lake Charles) (Spink 1967), indicating the existence of two independent points of entrance (La Fage 1987). (In 1969, it was discovered that a museum specimen collected in Charleston, SC, in 1957 had been misidentified as C. formosanus (Chambers et al. 1988).)

All of the points of entrance of the formosan subterranean termite have been associated with naval bases; therefore, it is hypothesized that this termite was introduced via naval cargo ships returning from the Far East at the end of WWII (La Fage 1987, Su and Tamashiro 1987, Beal 1987).

By the mid 1980s, *C. formosanus* was reported in Texas, Louisiana, South Carolina, North Carolina, Florida, Mississippi, Alabama, Georgia, and Tennessee (Beal 1987, Su and Tamashiro 1987). In 1992, *C. formosanus* was found in San Diego, CA, but this is believed to be an independent introduction from Hawaii (Atkinson et al. 1993). One of the main points of entrance has been New Orleans, where this pest has caused extensive damage to wooden structures and live trees and millions of dollars in losses (La Fage 1987, Rojas et al. 2001). It is being dispersed to other States in infested wood and railroad ties.

#### **Dispersal of Subterranean Termites**

Woodson et al. (2001) reported that by the end of the 20th century the formosan subterranean termite had spread to 95 localities in 11 States. This rapid spread in the continental United States has been attributed to transport of infested materials made of cellulose and to other human activities (Su and Tamashiro 1987, Woodson et al. 2001).

Natural dispersion of the formosan subterranean termite is slow. Its foraging range has been estimated at about 100 m (King and Spink 1969, Tamashiro et al. 1980); therefore, foraging dispersion is inconsequential. Dispersion via alate reproductives is more efficient, but it has been calculated that the flight range of reproductives does not exceed 450 m under normal swarming conditions (Su and Tamashiro 1987). A more recent study provides an estimate of 892 m by alate release and recapture across the Mississippi River in New Orleans, LA (Messenger and Mullins 2005). Nevertheless, alates may be accidentally transported long distances in trucks and trailers. Dispersion of formosan subterranean termites in the continental United States has been mainly anthropogenic, and it has been confirmed that infested railroad ties are a source of dispersion, as they are reused to build fences and delimit flower beds (Henderson 2000).

## Biology of Formosan Subterranean Termites

*Coptotermes formosanus* colonies start with a pair of alate reproductives, a single pair of male and female. Nuptial flight swarms normally take place from the end of April until the end of June (King 1971, Higa 1981, Higa and Tamashiro 1983). Henderson and Delaplane (1994) reported that between 1989 and 1992, the *C. formosanus* swarm season started during the first week in May in New Orleans. Formosan subterranean termites only swarm in evenings with high relative humidity, high probability of rain, and minimal winds, starting around 8:30 p.m. and ending around 9:30 p.m. (Henderson and Delaplane 1994).

Once alates have swarmed, they fall to the ground, lose their wings (and are now called "dealates"). They then mate, becoming "reproductives." Pairs of dealates are considered primary reproductives and are known as "king" and "queen." Dealates build a nuptial chamber in a suitable substrate and mate during the first 72 hours

2

after nesting. Egg laying begins 4 to 9 days after mating (King and Spink 1974). During the first year, queens have two egg-laying periods separated by a resting period in which there is no egg production (King and Spink 1974). The number of eggs laid during the first period has been estimated to average 20.2 (King 1971) and 28.2 (Higa 1981). Nevertheless, these estimates did not account for overlap of different developmental stages and loss of eggs to cannibalism (Morales-Ramos et al 2003). Morales-Ramos et al. (2003) used a graphic integration method to estimate total eggs laid during the first period. According to his method the mean number of eggs produced during this period was 36.7,  $\sigma = 15.5$ .

At the end of their first year, *C. formosanus* colonies did not exceed 100 individuals (Bess 1970, King 1971, King and Spink 1974, Higa 1981). At the end of the second year, colonies contained between 200 and 300 individuals (Bess 1970, King 1971, King and Spink 1974). Only after reaching 3 years did the number of individuals in the colonies increased into the thousands (Bess 1970, Higa 1981). Bess (1970) reported the presence of 10,000 eggs associated with a 4-year-old queen with an enlarged abdomen (physogastric). It has been calculated that a 4-year-old queen can lay up to 250 eggs a day. Bess estimated that the incubation period of the eggs is 40 days.

Colonies of *C. formosanus* contain millions of individuals. A capture-mark-release-capture method has yielded estimates that field colonies may contain between 1 million and 4 million foraging workers (Tamashiro et al. 1980, Su et al. 1984). Su and Tamashiro (1987) compiled laboratory data from several researchers and estimated that a colony may reach a population of 2.5 million at between 5.1 and 7.8 years.

Su and La Fage (1984) estimated that the daily consumption rate of this termite species was between 46.0 and 55.8 mg of wood per gram of termite at temperatures ranging between 22 and 24 °C and that the average individual weight of termite workers was 2.8 mg. Based on this data, a million termites could consume between 134.0 and 160.7 g of wood daily at 22 to 24 °C. Morales-Ramos (2004, unpublished data) calculated that the daily consumption rate for groups of 250 termite workers was between 16 and 20 mg at a constant temperature of  $27\pm1$  °C. Based on these estimates, a million *C. formosanus* workers are capable of consuming between 64 and 80 g of wood per day. A field colony probably does not reach this consumption rate because of natural temperature fluctuations. Nevertheless, these estimates help to give us an idea of the destructive potential of this pest.

#### **Control Methods**

Control methods have been divided into defensive methods and suppressive methods.

Defensive methods protect structures from foraging termites. Defense includes use of chemical and physical barriers and wood treatments. Chemical barriers include repellent liquid termiticides applied in trenches around structures and injected under slabs. Chemical barriers do not control subterranean termite population; they only temporarily protect structures from foraging termites (Su and Scheffrahn 1998). Similarly, physical barriers only protect structures from foraging termites. The most common physical barriers are basaltic barriers made of uniform-size gravel, steel mesh, and chemically treated plastic sheets. These barriers have been demonstrated to be effective, but they require special installation before structure construction and they increase construction cost (Su and Scheffrahn 1998).

Suppressive methods reduce the population of subterranean termites. These methods include use of nonrepellent termiticides and slow-acting baits. Liquid insecticides are applied in the same manner as repellent termiticides; foams and gels are injected directly into the structure suffering an active termite infestation. These chemicals act slowly, which allows the termites that come in contact with the insecticide to distribute it to other members of the colony during normal grooming behavior.

Esenther and Beal (1974, 1978) introduced the concept of using insecticidal baits to eliminate termite infestations in residential buildings and structures. The first effective baits against subterranean termites consisted of decayed wood blocks impregnated with mirex (Esenther and Beal 1974, 1978). Newer bait systems incorporate slow-acting toxicants or growth regulators such as chitin synthesis inhibitors and metabolic inhibitors. Chitin synthesis inhibitors such as diflubenzuron, hexaflumuron, lufenuron, noviflumuron, and chlorfluazuron belong to the benzoylphenyl urea family (Beeman 1982). Metabolic inhibitors include hydromethylnon, sulfuramid, and borates (Su and Scheffrahn 1998).

Baits are placed in ground stations around structures (Su and Scheffrahn 1991, Su 1994) or in above-ground stations inside structures (Su and Scheffrahn 1997) infested by subterranean termites. The principle is that foraging termites feed on the bait and distribute the toxin or active ingredient to the rest of the colony viatrophallaxis (Su and Scheffrahn 1988, Lewis 1997). In a laboratory study, Suárez and Thorne (2000) determined the rate of distribution of alimentary fluid by trophallaxis in *Reticulitermes flavipes, R. virginicus,* and *Zootermopsis nevadensis* and concluded that slow-acting, short-life toxicants have potential for wide dissemination through colonies of these species.

Dry-bait matrices were initially used in commercial baiting systems. These systems consisted of cardboard or purified cellulose paper impregnated with the active ingredient (Su 1991, Su and Scheffrahn 1993, Shaheen 1997). Other commercial baiting systems used  $\alpha$ -cellulose powder mixed with the active ingredient.

A new generation of baits includes high-moisturecontent matrices formulated to be nutritionally attractive to termites (Morales-Ramos et al. 2003). One of these bait matrices, developed by USDA-ARS researchers, was based on the feeding preferences and nutritional requirements of the formosan subterranean termite (Morales-Ramos and Rojas 2003, Rojas and Morales-Ramos 2001, Rojas et al. 2003). The purpose of this formulation was to increase bait consumption by subterranean termites in order to enhance assimilation of active ingredients within colonies, consequently reducing the time required by treated termites to attain lethal doses (Morales-Ramos and Rojas 2003a, Rojas 2002a, b, Rojas and Morales-Ramos 2003). These bait matrix formulations in combination with feeding stimulants and masking agents (Rojas et al. 2004b) allow incorporation of less palatable but more widely available active ingredients, such as diflubenzuron, without compromising bait consumption by the termites (Morales-Ramos and Rojas 2003a).

Field observations have shown that treatment of individual infested structures is not an optimal approach to suppress colonies of subterranean termites, and many studies have shown inconsistent results using this approach (Lewis 1997, Su and Scheffrahn 1998). This is mostly because formosan subterranean termite colonies can occupy extensive terrain within their foraging territory, including many privately owned properties (Su and Tamashiro 1987). Control of formosan subterranean termites may be more effective if approached on an areawide basis instead of structure by structure.

In the early 1980s, Knipling and Rohwer published a new concept for control of insect pests—areawide management. This concept is based on four fundamental concepts: "Areawide management of pests a) should be executed in extensive geographic areas; b) should be coordinated by organizations instead of individuals; c) should be focused on the reduction and management of key pest densities at acceptable low levels; and d) may include a regulatory component to ensure full participation in the program" (Kogan 1998). Based on this concept, an areawide management program designed to control subterranean termites was implemented.

The primary objective was to assess the feasibility of areawide control by reducing or eliminating colony populations of subterranean termites using baiting systems with slow-acting toxicants and new natural products in the recently developed USDA/ARS bait matrix. Secondary objectives were—

- determining the potential of targeting tree species for placement of bait stations to maximize the probability of detecting termites within a neighborhood, and
- investigating potential ecological relationships between tree species and termite activity.

## Materials and Methods

## Monitoring Alate Swarms of Formosan Subterranean Termites

Alates were captured with glue boards (CatchMaster) mounted on  $30 \times 6$  cm wooden boards hung on nails beneath private utility lights and public street lights. Protective wire barriers were placed over the glue boards to prevent unintentional capture of wildlife (figure 1a). In forested areas, the glue boards were mounted on  $30 \times 6$  cm boards hung underneath solar-powered LED white lights (Brinkmann Solar).

The solar traps were mounted on trees or utility poles without lights and were placed within 275-800 m of the nearest manmade structure. Solar traps were placed more than 275 m from rural buildings or more than 100 m from an artificial light source in a rural location. The ideal location for a solar trap allowed sufficient exposure to sunlight to ensure that the batteries would be recharged. Therefore, the east side of trees and utility poles located under a thin or absent canopy was preferred (figure 1b).

In 2000, over 1,100 sticky traps were placed throughout the southern 13 counties of Mississippi from the middle of April to the middle of June. Traps were collected once a week for 10 weeks (Henderson and Delaplane 1994) and placed inside  $30 \times 30$  cm minigrip polybags (ZipLock). To inhibit fungal growth and decay, the traps were then refrigerated or frozen until they could be analyzed.

A protocol was established to count and identify the alates captured in each trap (figure 1c). All alates were observed in situ using a stereoscopic microscope (Nikon model C-PS, 10X/22). All captured alates were counted, and the number for each trap recorded. For traps with 5 or fewer individuals, all of the alates were identified as either *C. formosanus* or unknown; for traps with 6 to 50 individuals, 5 randomly selected individuals were identified; for traps with greater than

50 individuals, 10 randomly selected individuals were identified. Alates were identified as *C. formosanus* by the distinctive hairs found on the surface and margins of the unpigmented wings and the two distinct and sclerotized veins in the costal field that are visible along the entire length of the wing (Scheffrahn and Su 1994).

The random selection process involved use of clear acetate sheets with random sampling points punched out. There was a selection of random hole-sheets. with either 5 or 10 points, for the selected densities; the sheets were alternated with each trap. The sheets were placed over the plastic-wrapped glue trap and a fine-point marker was used to mark the sampling point on the trap. The alate closest to the point was then identified. For example, during the week of June 9, 2003, trap CC06 captured 27 alates. Of those, 5 were randomly selected for identification, of which 2 were C. formosanus. The 3 alates that did not key out to C. formosanus or could not be identified due to damage were listed as "unknown." A ratio of 2:5, or 40 percent, was then multiplied by 27 to establish an estimated count of 10.8 C. formosanus alates for trap CC06 on that date.



Figure 1. (A) Sticky board.



(B) Sticky board on light pole.



(C) Formosan termites on sticky board.

The total number of *C. formosanus* alates captured in that area was determined by adding the accumulated estimated totals for the 10-week sampling period. The average capture per trap was calculated using the estimated count and the number of traps per area that captured *C. formosanus* alates.

This survey confirmed the presence of formosan subterranean termites and their expansion into a relatively large geographical area in the State of Mississippi.

#### Phase 1 (March 2001 to October 2002)

**Site selection.** Data obtained from alate dispersion studies in spring 2000 were used to determine the location of test sites with best probability of having formosan subterranean termite colonies present. This determination was based on the assumption that formosan subterranean termite colonies existed within approximately 100 m of sticky traps that caught relatively high numbers of alates.

A total of 12 sites were selected inside Keesler Air Force Base (Biloxi, MS) and adjacent neighborhoods, and a total of 1,800 underground stations (figure 2a) were installed. A pattern of underground monitoring stations was established around those light traps that had shown a relatively high number of catches (figure 1c). Underground double-purpose monitoring-baiting stations (Quarterra, developed by Ensystex) were used for the study (figure 2b). These cylindrical stations have six wooden aspen interceptors (used for monitoring) placed inside and tightly secured around the inner wall of the station, allowing the termites easy access to the empty cavity in the middle; the bait is placed in the middle cavity. The design of these stations allowed easy monitoring and bait placement with minimal disturbance to the termites (figure 2b).

The stations were placed in a grid pattern within each neighborhood. This pattern followed the geometry of the neighborhood; as a consequence, the patterns of the stations differed among the test sites. To increase the probability that termites would visit the stations, positioning of houses was also taken in account in placing stations. From the total number of stations assigned to each site (150), 30 were strategically placed around trees and shrubs infested by termites and apparently healthy trees susceptible to their attack (McMichael 1998, Henderson 2000, Morales-Ramos and Rojas unpublished) (figure 3). The stations were installed in August 2000, and monthly monitoring started in September. Once the system was established and termite activity increased, baiting started in March 2001.

**Bait formulation.** The bait matrix was formulated according to Rojas and Morales-Ramos (2001), having a final composition of 69.7 percent water, 30 percent  $\alpha$ -cellulose powder, and 0.3 percent a nutritional supplement (Rojas et al. 2003). The nutritional supplement was dissolved in drinking water. A slow-action chemical chitin synthesis inhibitor belonging to the family of benzoylphenyl ureas, diflubenzuron (Ensystex), was dissolved in a minimum amount of acetone and mixed with water containing the nutritional supplement and 200 ppm of urea as a feeding stimulant (Rojas et al. 2004b). This liquid was mixed with  $\alpha$ -cellulose powder. The final mixture contained 250 ppm of diflubenzuron.

One hundred and fifty grams of this bait was enclosed in bags (provided by Ensystex). The bags were made of polyester net, lined with waxed construction paper to protect, manipulate, transport, and store the bait. Bags with bait were placed only into the preinstalled stations that contained subterranean termites, as recommended by the manufacturer (figure 2b). Ants, which kill termites, were controlled by use of a granular, nutritionally based bait matrix (Rojas and Morales-Ramos 2003); this bait also contained diflubenzuron as the active ingredient to avoid contaminating the test with other toxicants.

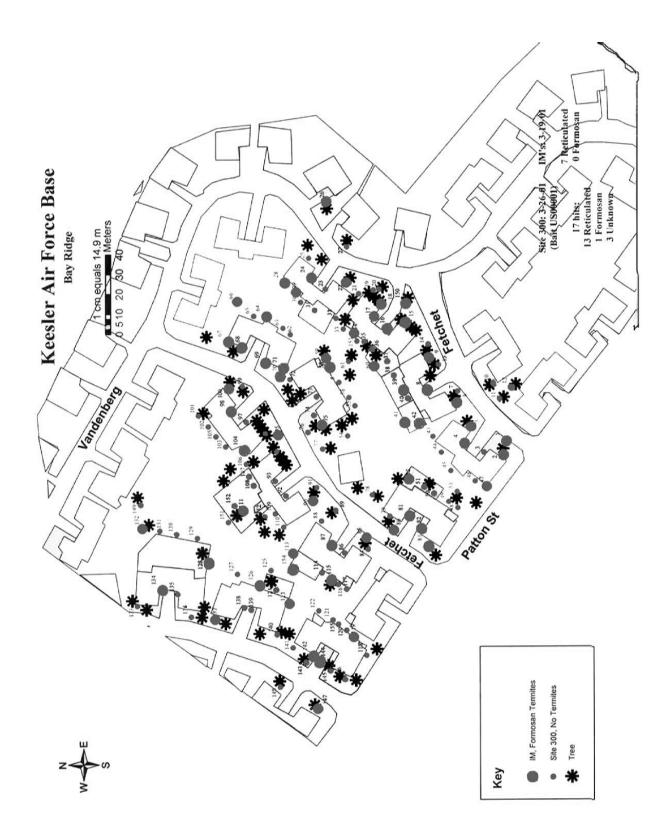
Monitoring. The test was evaluated by use of control areas and independent monitors. Six neighborhoods were designated as treatment areas and six as control areas. Because of the variability of the test sites, treatment and control areas were paired based on similar physical characteristics and outward appearances. In each pair, one site was treated and the other was left to regular pest maintenance by KABF personnel. These latter sites were designated "treated controls." Every fifth station was designated as an independent monitor (figure 3). This allowed 30 stations per site to serve as an added control in the treated sites. These independent monitoring stations were treated similarly to the rest of the underground stations but did not receive any bait throughout the duration of the study's phase 1. When termites were present in the independent monitors, additional wooden interceptors were added to maintain termite activity in the station by stimulating foraging.



Figure 2. (A) Underground station.



Figure 2. (B) Bait placement and monitoring.



Monitoring was done by two different crews. One crew was responsible for monitoring the control neighborhoods, independent monitors, and unvisited normal stations in treated neighborhoods. The second crew was responsible for monitoring termite activity in normal stations in treated neighborhoods that had been visited by termites and for placing bait and monitoring its consumption.

**Termite identification.** Samples of termite workers and soldiers present in the stations were collected in 16 ml glass scintillation vials (Wheaton) containing 70 percent ethanol. Vials were taken to the laboratory at the Coastal Research and Extension Center, Biloxi, MS, where specimens were identified using taxonomic keys (Weesner 1965, Krishna 1966, Scheffrahn and Su 1994, and Messenger 2001) aided by a stereoscopic microscope (Nikon model C-PS, 10X/22).

Data collection and analysis. As soon as a station became active with termites, it was immediately barcoded and designated as a visited station. Data were collected using either MS Excel paper data sheets or electronically using Symbol palm barcode scanners (SPT 1700) and Exterris software Ver. 1 provided by Ensystex. Later the software was exchanged for an updated version developed by Rojas et al. (2003, unpublished data). Data collected included station number, barcode number, site number, and collection date; tree number and species; presence or absence of termites and species; presence or absence of ants and species; bait added, consumed, or removed; wood interceptors eaten, rotted, and added; and temperature inside the station. Temperature was measured inside the stations using an infrared laser thermometer (Raytek, Raynger ST20). The data were downloaded to a computer as a text file and converted to spreadsheet format using Excel (Office 97, Microsoft). Stations visited by termites retained their status as visited for the duration of the study. Stations with termites present were designated as active or inactive and this status was listed for every monitoring date according to field observations.

Data were analyzed in two different ways. The first method consisted of comparing the number of active stations observed between treated and control neighborhoods. Mean numbers of active stations were compared using analysis of variance (ANOVA) and Student's *t*-test. The second method consisted of calculating the proportion of active stations based on the cumulative number of visited stations. This was calculated as  $active_{DS}/visited_{S}$ , where active was the number of stations with termite present at date 'D' in site 'S,' and visited is the cumulative number of stations visited by termites in site 'S.' Proportions of active stations were compared between treated and control neighborhoods by the Z-test for categorical data analysis (Ott 1984).

#### Phase 2 (November 2002 to January 2004)

Experimental area. Four of the original neighborhoods were retained in 2003 to continue with phase 2: Bay Ridge Fetchet and Orville Wright as treated and East Falcon A and B as controls. Starting in phase 2, all stations that showed termite activity were treated, including previously designated monitoring stations. This was done to increase the treated area in the treated neighborhoods and to treat all colonies that may have escaped treatment during phase 1. Treatment was evaluated by comparison with the control (untreated) neighborhoods. The selection of neighborhoods for phase 2 was based on presence of formosan subterranean termites in the underground stations and number of alate catches in sticky traps during the 2001 swarm season. At the end of phase 1, formosan subterranean termites were present in only one of the treatment neighborhoods, Bay Ridge Fetchet, but an abundance of formosan subterranean termite alates were persistent in all of the neighborhoods. Phase 2 started on December 2, 2002, and ended on January 30, 2004.

**Bait formulation.** The bait matrix was the same as in phase 1, but a new active ingredient, *N*-hydroxynaphthalimide (NHA) (Rojas et al. 2004a), was added to increase the effectiveness of the bait. The new bait formulation included 250 ppm diflubenzuron and 500 ppm NHA in the basic USDA-ARS bait matrix. Bait was delivered to the treatment neighborhoods as described in phase 1.

#### Phase 3 (February 2004 to August 2004)

This phase of the study tested the possibility of eliminating formosan subterranean termite activity from the only neighborhood in which this species was present—Bay Ridge Fetchet. All other neighborhoods were eliminated from the study. To achieve total reduction of formosan subterranean termite activity, a more potent chitin synthesis inhibitor, chlorfluazuron, was used. The bait was formulated as in the previous phases using 250 ppm of chlorfluazuron instead of the other active ingredients. The treatment period started on February 27, 2004, and ended on August 26, 2004.

Because the only objective of this phase was to eliminate formosan subterranean termites, there was no attempt to do any statistical comparison with untreated controls.

#### Phase 4 (January 2004 to May 2005)

Formosan subterranean termite alate captures during the swarm season of 2003 suggested their presence in the 12 selected neighborhoods. However, C. formosanus remained undetected in underground stations of 11 of the 12 neighborhoods during phase 1. Because underground bait stations were installed in a grid pattern around houses, our next hypothesis was that termite colonies were primarily infesting trees and that these colonies were too small to reach the underground stations located near the houses. Trees inside the study area were systematically inspected for termite presence to test the above hypothesis. The inspections consisted of searching for mud tunnels on the outer bark and base of the trees, shrubs, and stumps and searching into tree folds, cavities, and imperfections by prodding with a metal weed picker to find soft areas that might indicate the presence of carton material and termites. The species of trees and termites were recorded along with location (figure 4).

Once the presence of subterranean termites was confirmed, eight underground Quarterra stations were installed around each infested tree in a double ring pattern that placed the first set of four stations as close to the tree as possible oriented according to the cardinal points (north, east, south, and west) with station "A" always being placed on the north side of the tree. For the second set, four stations were placed approximately 5 feet farther from the tree than the first set at the half-cardinal points (northeast, southeast, southwest, and northwest.) The stations were installed during November 2003. Monitoring, following all previous protocols, began in January 2004. After enough time to allow establishment of the termites in the stations. treatment started at the end of March. At that time, 50 percent of the trees in the study had stations visited by formosan subterranean termites. The bait matrix contained chlorfluazuron at 250 ppm, formulated as before. Baiting, monitoring, and data collection were also performed as before.

Infested trees that had been hollowed by termite foraging, indicated by the presence of carton material, were inspected with a borescope at the beginning of phase 4 and 6 months later. Three holes were drilled in each tree using a 1/2-inch sterile drilling bit in an attempt to locate voids. If a void was located, then observations were done using a precision flexible borescope (Hawkeye model HFB2, gradient lens). The presence or absence of live termites inside the tree voids was recorded.

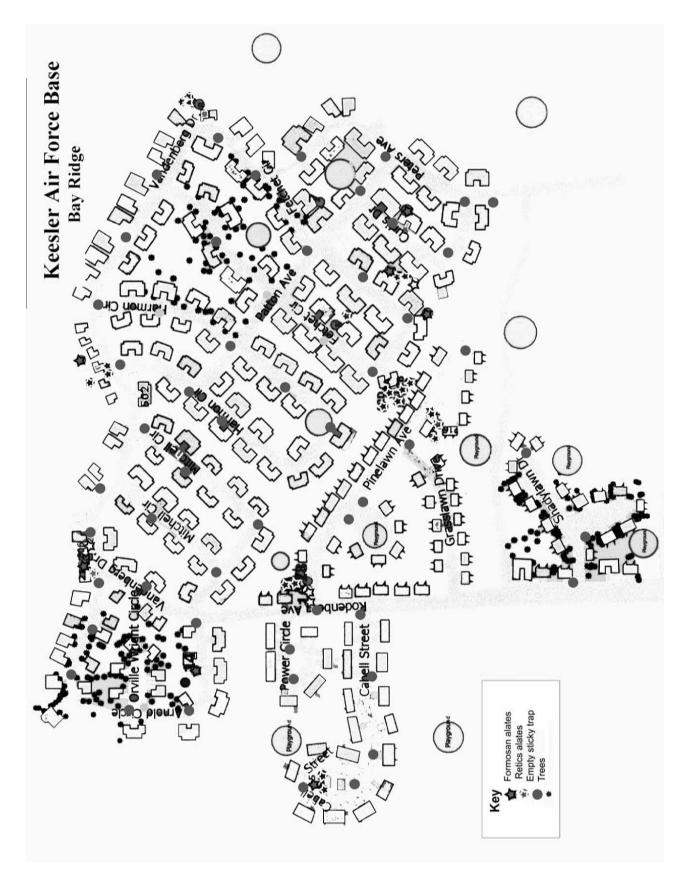
#### Laboratory Comparison of Active Ingredients

The relative toxicities of diflubenzuron and chlorfluazuron were compared under laboratory conditions in order to explain differences observed in suppression levels between phase 4 and phases 1 and 2.

Groups of 1,000 termite workers and 200 soldiers were placed inside transparent polystyrene boxes  $17 \times 12 \times 6$  cm in dimension. The boxes were filled with a substrate of 400 ml of a 1:1 mixture of topsoil and sand, 200 ml of distilled water, and 1 g of the water-absorbing polymer polyacrilamide. Each box was vertically connected to a polystyrene dish located on the cover of the box through a 0.5 cm diameter hole and linked to the termite substrate by 50 ml centrifuge tubes cut at the bottom and glued to the inner side of the box cover. Two pieces of cardboard were placed inside the centrifuge tubes to encourage the termites to go inside and up to the connected dish located on the outer side of the box cover. The top dish was used as a foraging area for the termites in order to avoid direct contact of the termite group to the active ingredients.

Termites were fed with blocks  $(3 \times 2 \times 0.4 \text{ cm})$  of red oak wood (*Quercus rubra* L.) placed inside the foraging dish. Red oak was chosen as a food source because oak trees were the most common trees in the treated neighborhoods. The wood blocks were dried in a vacuum oven and weighted before being provided to the termite groups. New wood blocks were added to the foraging dish as termites consumed the wood.

Termites were collected from six different colonies in New Orleans City Park. The method of collection consisted of burying buckets next to infested trees. Wet corrugated cardboard rolls placed inside each bucket lured the termites. Rolls were collected daily and brought to the laboratory where the termites were



separated from the cardboard by gentle aspiration. Termites were counted while being aspirated and placed directly inside the test boxes.

Baits were prepared as usual using 250 ppm of the active ingredient. Small centrifuge tubes (2.0 ml) were filled with 1.6 g of bait. Three different doses of each of the active ingredients were tested: 1.6, 3.2, and 4.8 g of bait per 1,200 termites. These doses corresponded to approximately 0.4, 0.8, and 1.2  $\mu$ g of active ingredient per termite worker. Treatments consisted of exposure to 1, 2, or 3 of tubes with bait from each of the two active ingredients, making a total of 6 treatments, in addition to a control group not exposed to any chemical. Each treatment and control was replicated 10 times for a total of 70 test boxes.

The test boxes were maintained at  $27\pm1^{\circ}$ C,  $95\pm5$  percent relative humidity, and total darkness for 16 weeks. The test boxes were monitored daily for food availability and bait consumption and weekly for termite group mortality. Only the death of whole termite groups was taken into consideration in the analysis. At the end of the study, remaining wood blocks and bait were dried and weighted to determine the full wood and bait consumption of each group.

#### Results

Phase 1 (March 2001 to October 2002)

**Termite population dynamics.** Three different species of subterranean termites were present in our study area. The most common were native eastern (*R. flavipes*) and southern (*R. virgicus*) subterranean termites. Introduced formosan subterranean termites (*C. formosanus*) were detected in only one of the neighborhoods despite heavy captures of alates in all locations during swarming season. Subterranean termites were more active during late summer: Activity generally peaked during August and September (figure 5). Increase in termite activity was associated with increase in station temperature, but after a delay of a few days.

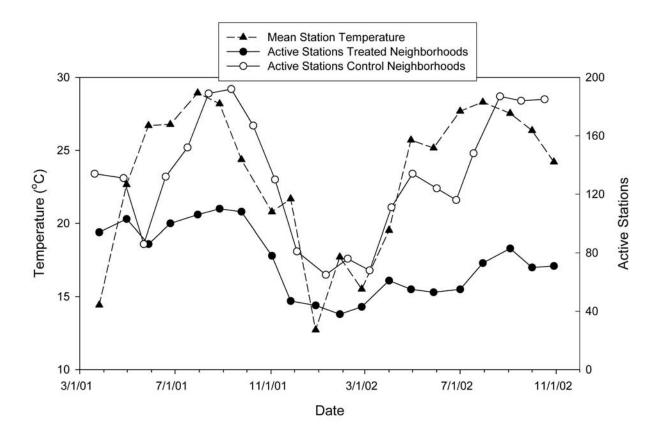


Figure 5. Mean station temperature and termite activity in underground stations in 12 Biloxi, MS, neighborhoods March 2001 to November 2002.

In many instances, it was also noted that more than one termite species was present in the same underground station simultaneously. In other instances the presence of one termite species was replaced by another termite species depending on season. Native termites were more tolerant of hot-cold temperature extremes than the formosan subterranean termite. This pattern of termite activity was also observed in living trees such as oaks and pines.

Ants frequently nested in the stations, mostly native tropical fire ants (*Solenopsis germinata* F.), red imported fire ants (*S. invicta* Buren), and odorous house ants (*Tapinoma sessile* Say). Ants were occasionally found sharing the space of the underground stations, base of trees, or tree cavities with subterranean termites. Under normal conditions no apparent competition for space was observed because ants occupied the empty cavity of the station, but termites still could access the interceptors or bait by building soil barriers and mud tunnels to protect themselves from predation. On trees, termites freely moved up and down via their characteristic mud tunnels, which allowed them to remain undetected by ants and other predators.

**Treatment effects.** During phase 1, termites visited a total of 870 stations, of which 409 were in treated and 461 in control neighborhoods (see appendix). Bay Ridge Fechet was the most infested site of the treated neighborhoods and East Falcon A the most infested of the control neighborhoods (table 1).

Termite activity in normal stations and independent monitors of control neighborhoods followed a normal cyclic pattern, with activity highest during late summer and minimal during mid winter (figure 6a). In the treated neighborhoods termite activity showed a similar cyclic pattern in the normal stations, but termite activity declined during the second year of the study (figure 6b). This phenomenon was observed in the independent monitors of treated neighborhoods to a lesser degree.

Neighborhood	Total stations	Monitoring stations	Stations visited by termites	Percentage visited	Stations visited by by FST*
Freatment					
Bay Ridge Fetchet	161	31	76	47.2	19
Bay Ridge Orville Wright	158	31	56	35.4	0
West Falcon A	154	34	61	39.6	0
West Falcon B	153	30	40	26.1	0
Shadylawn	161	32	46	28.5	0
Sun Coast Villa A	161	32	48	29.8	0
Control					
East Falcon A	150	30	89	59.3	0
East Falcon B	154	31	87	56.4	0
Pinehaven A	157	31	61	38.8	0
Pinehaven B	151	30	24	15.8	0
Oak Park	154	31	76	49.3	0
Sun Coast Villa B	153	29	58	37.9	0
All Neighborhoods	1,867	372	724	38.7	19

#### Table 1. General underground station information of the 12 neighborhoods for phase 1

\* Formosan subterranean termite (Coptotermes formosanus).

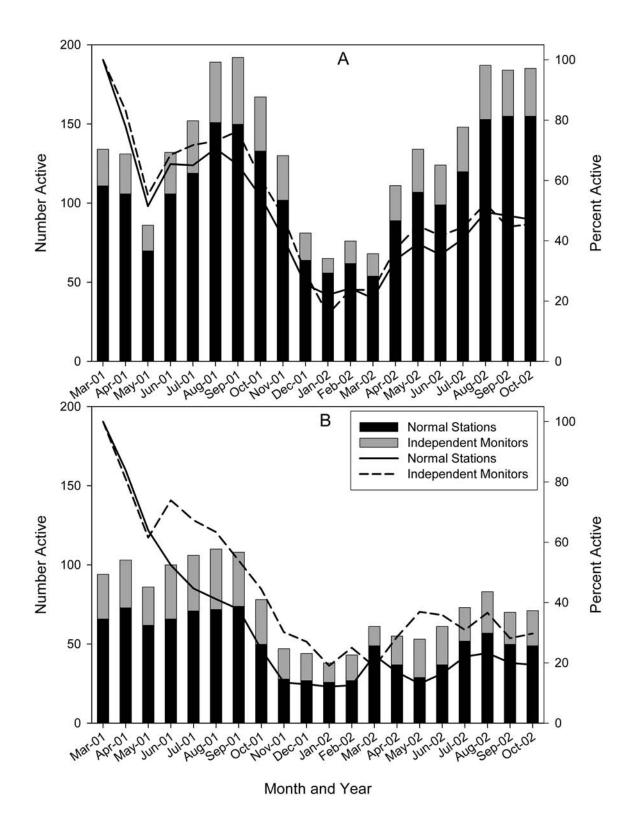


Figure 6. Termite activity in (A) untreated and (B) treated neighborhoods during phase 1. Bars represent number of active stations; lines represent the percentage of active stations calculated from the total number of stations visited by termites.

Data analyses indicated that independent monitors were nine times more likely than normal stations to have termite activity at the end of the test. Analysis of termite activity at the end of the study between treatment and control in terms of number of active stations (figure 7b) and percentage of active stations (figure 7c) indicated that termite activity was significantly higher in control stations than in treatment stations, since the calculated *t* and *Z* values were above the critical values of significance of the *t* and *Z* tests from tables, represented by the dotted lines in figure 7a and b. Following the same rationale, figure 8 compares termite activity between control and treatment in the monitoring stations. The data show that, during most of the course of the study, differences in termite activity between control and treatment monitoring stations were not significant. Nevertheless, at the end of the study, termite activity was significantly higher in control stations than in treatment monitoring stations (figure 8b and c).

At the end of the study, a total of 22 monitoring stations remained active—31 percent of the total number of active stations (71). Because only 22.6 percent of the stations visited by termites were monitors, termite activity is considered to be comparatively more persistent in monitoring stations than in normal stations. We attributed this difference in activity to termite colonies that escaped treatment because of the lack of termite presence in adjacent normal (treated) stations. In many cases the termites found in monitoring stations were of different species than termites found in adjacent normal (treated) stations.

Termites consumed a total of 95.5 kg of bait in all treated neighborhoods during phase 1. Figure 9 shows the distribution of bait consumption over time in the six treated neighborhoods. The neighborhood with the greatest bait consumption was Bay Ridge Fetchet, where termites consumed a total of 20.7 kg of bait (table 2).

**Tree associations.** A total of 26 genera of trees and shrubs had associated monitoring stations. Of these, 6 genera represented 82 percent of the trees and shrubs. These 6 plant genera and the percentage of stations associated with each are shown on table 3. The remaining 18 percent of the stations included 20 genera, each represented by fewer than 10 individual plants (Hollomon et al. 2001).

#### Phase 2 (November 2002 to January 2004)

Though still present at the end of phase 2, termite activity was significantly higher in untreated than in treated neighborhoods (figure 10a). Use of baits with diflubenzuron achieved a steady decline of termite activity in 2 years and 10 months of phases 1 and 2 in both Bay Ridge neighborhoods. Termite activity declined consistently every growing season during this period (figure 10a). A total of 43, 37, and 27 stations were active during August of 2001, 2002, and 2003, respectively, in both Bay Ridge neighborhoods. However, this activity represented 43.4, 29.8, and 19.7 percent, respectively, of the total visited stations.

Untreated neighborhoods (East Falcon A and B) showed little change in termite activity during the same three growing seasons. A total of 86, 89, and 79 stations were active during August of 2001, 2002, and 2003. These active stations represented 69.9, 52.6, and 43.8 percent of the total stations visited by termites. These differences represent 50.0, 58.4, and 65.8 percent reduction in termite activity in the treated neighborhoods compared to the untreated neighborhoods.

Statistical analysis of termite activity and percentage of active visited stations show significantly higher activity in untreated neighborhoods from August 2002 through the end of phase 2 in December 2003 (Figure 10b and c). Phase 2 started in November 2002; therefore, these two treated neighborhoods started showing significant reduction in termite activity during phase 1. Termite activity data from phase 2 showed a moderate increase in the level of reduction of termite activity in treated neighborhoods compared to the control neighborhoods (figure 10). At this point of the study, termite suppression levels seemed to have reached a plateau at which further gains in level of suppression appeared to be growing smaller every year.

Termites consumed a total of 52.5 kg of bait during phase 2 (table 4). Figure 11 shows bait consumption over time in the two treated neighborhoods. Mean bait consumption per active station during phase 2 showed a slight increase over phase 1 (tables 2 and 4).

#### Phase 3 (February 2004 to August 2004)

Six stations in the Bay Ridge Fechet neighborhood had remained consistently active with formosan subterranean termites through phases 1 and 2, a

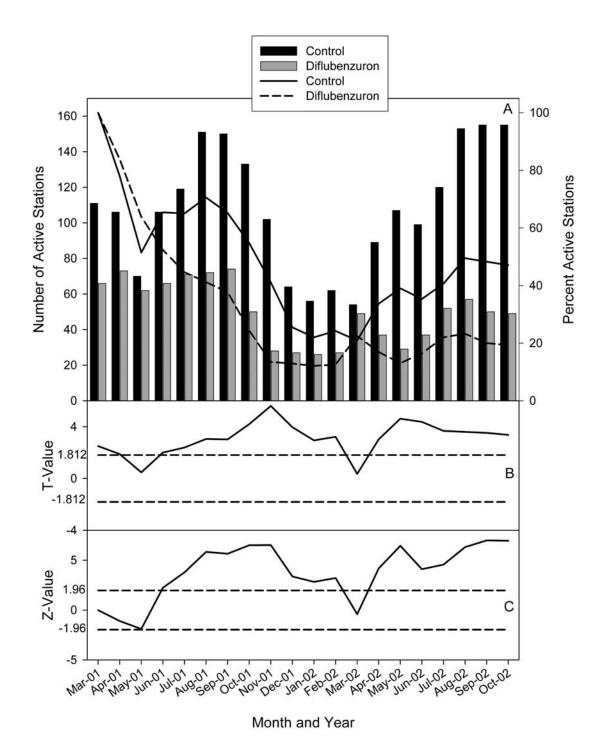
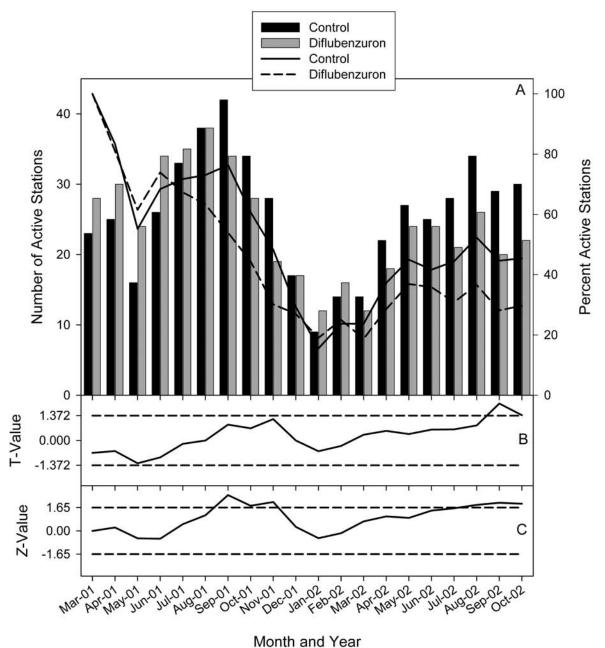


Figure 7. Comparison of normal station activity between six treated and six untreated neighborhoods during phase 1. (A) Bars represent number of active stations, and lines represent percentage of active stations calculated from the total number of stations visited by termites. (B) Student's *t*-test comparing numbers of active stations between treated and untreated neighborhoods; solid line represents calculated *t*-values and dashed lines represent the critical values of *t*. (C) *Z*-test comparing the percentages of active stations between treated and untreated neighborhoods; solid line represent the critical values of *Z*. In B and C, values of *t* or *Z* between the dashed lines indicate no significant differences between treated and untreated neighborhoods; values above the dashed lines indicate significantly greater activity in untreated neighborhoods.



Month and Tear

Figure 8. Comparison of independent monitor station activity between six treated and six untreated neighborhoods during phase 1. (A) Bars represent number of active stations, and lines represent percentage of active stations calculated from the total number of stations visited by termites. (B) Student's *t*-test comparing numbers of active stations between treated and untreated neighborhoods; solid line represent the calculated *t*-values and dashed lines represent the critical values of *t*. (C) *Z*-test comparing the percentages of active stations between treated and untreated neighborhoods; solid line represent critical values of *Z*. In B and C, values of *t* or *Z* between the dashed lines indicate no significant differences between treated and untreated neighborhoods; values above the dashed lines indicate significantly greater activity in untreated neighborhoods.

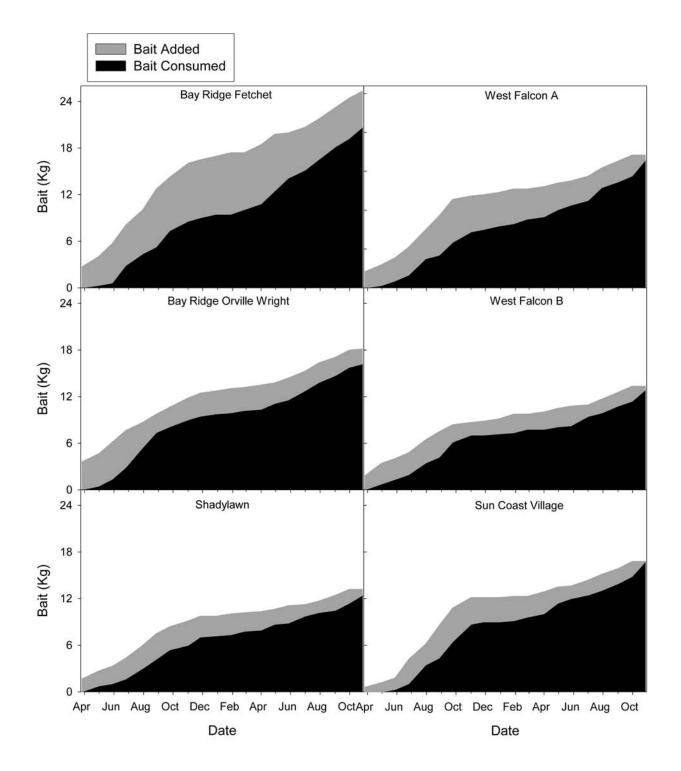


Figure 9. Total bait added to underground stations (grey) and consumed by termites (black) in six treated Biloxi, MS, neighborhoods March 2001 to November 2002.

Neighborhood	Normal stations visited by termites	Monitoring stations visited by termites	Total bait consumed	Bait consumed per visited normal station
			kg	g
Treatment				
Bay Ridge Fetchet	63	13	20.7	$328.6 \pm 26.0$
Bay Ridge Orville Wrigh	t 42	14	16.2	$385.7 \pm 39.0$
West Falcon A	48	13	16.5	$343.8 \pm 32.8$
West Falcon B	30	10	12.9	$430.0 \pm 48.6$
Shadylawn	35	11	12.4	$355.7 \pm 50.4$
Sun Coast Villa A	35	13	16.8	$480.0 \pm 55.3$
Control				
East Falcon A	71	18	0	0
East Falcon B	73	14	0	0
Pinehaven A	53	8	0	0
Pinehaven B	21	3	0	0
Oak Park	64	12	0	0
Sun Coast Villa B	47	11	0	0

## Table 2. Termite activity and bait consumption in 12 neighborhoods at the end of phase 1

### Table 3. Major types of trees and shrubs having monitoring stations during phase 1

	Genus	Percentage
Oak	Quercus	54
Pecan	Carya	7
River birch	Betula	6
Sweetgum	Liquidambar	6
Tallow	Sapium	5
Pine	Pinus	4
Total		82*

\* 3% various other genera.

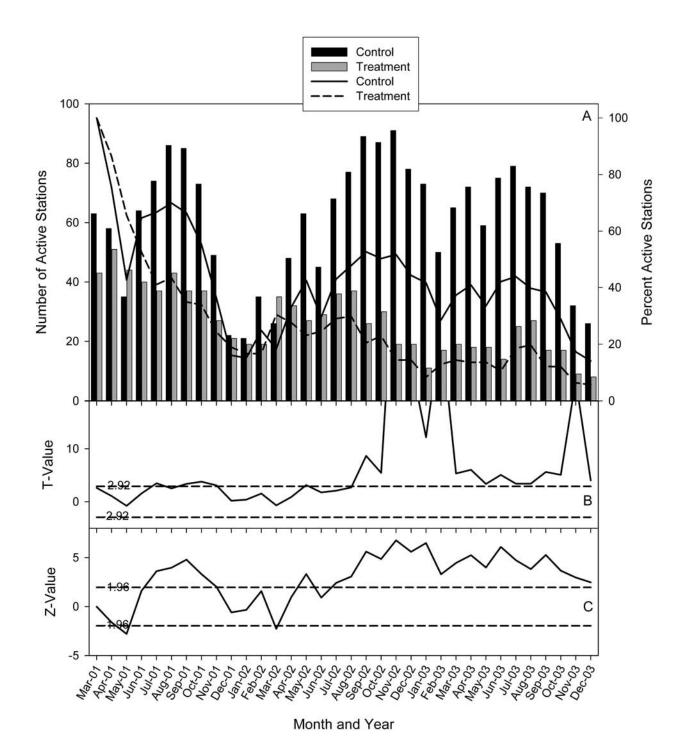


Figure 10. Comparison of station activity between two treated and two untreated neighborhoods during phases 1 and 2. (A) Bars represent number of active stations, and lines represent percentage of active stations calculated from the total number of stations visited by termites. (B) Student's *t*-test comparing numbers of active stations between treated and untreated neighborhoods; solid line represent the calculated *t*-values and dashed lines represent the critical values of *t*. (C) *Z*-test comparing the percentages of active stations between treated and untreated neighborhoods; solid line represent the calculated *Z* values and dashed lines represent the calculated neighborhoods; solid line represent the calculated reated and untreated neighborhoods; solid lines represent the calculated *Z* values and dashed lines represent critical values of *Z*. In B and C, values of *t* or *Z* between the dashed lines indicate no significant differences between treated and untreated neighborhoods; values above the dashed lines indicate significantly greater activity in untreated neighborhoods.

Neighborhood	Total number of stations visited by termites	Stations visited anew during phase 2	Total bait consumed	Bait consumed per visited station
Treatment			kg	g
Bay Ridge Fetchet	85	8	30.4	$371.3 \pm 27.3$
Bay Ridge Orville Wright		4	22.0	$413.9 \pm 41.0$
Control				
East Falcon A	91	3	0	0
East Falcon B	94	8	0	0

## Table 4. Termite activity and bait consumption in 4 neighborhoods at the end of phase 2

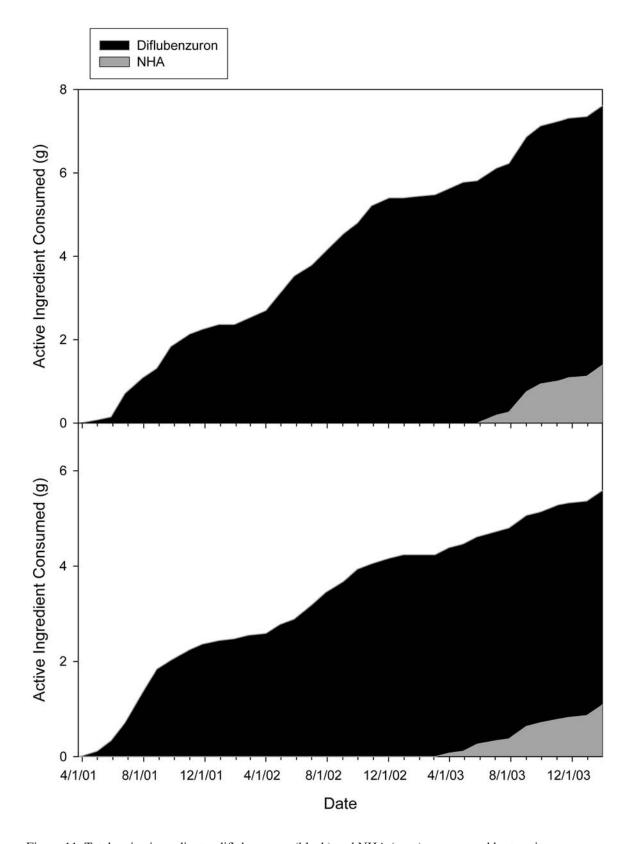


Figure 11. Total active ingredient—diflubenzuron (black) and NHA (gray)—consumed by termites in underground stations in six treated Biloxi, MS, neighborhoods March 2001 to November 2002.

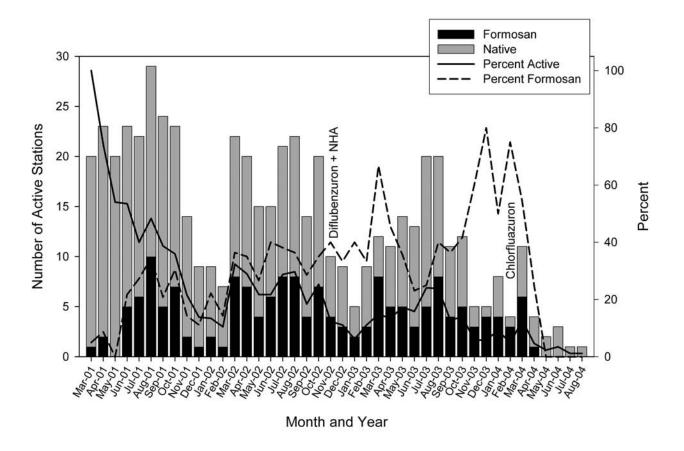


Figure 12. Formosan (black) and native (grey) subterranean termite activity observed in Bay Ridge Fetchet neighborhood during phases 1, 2, and 3, March 2001 to August 2004. Solid line represents percentage of active stations with either species of termites calculated from the total number of stations visited by termites; dashed line represents percentage of formosan subterranean termite activity from total number of active stations.

period of 2 years and 10 months, and consumed several kilograms of diflubenzuron bait. All formosan subterranean termite activity in these stations ceased within 3 months of introducing the new chlorfluazuron bait (figure 12), and formosan subterranean termites remained undetectable to the end of phase 3.

The chlorfluazuron bait was less successful in eliminating activity of native subterranean termites from all active stations. Some stations had sporadic activity of native subterranean termites to the end of phase 3 (figure 12). Active stations showed the presence of different species of native termites during phase 3, evidence of the presence of numerous but relatively small colonies. Nevertheless, only one station remained active with subterranean termites (*R. flavipes*) by the end of phase 3 (figure 12).

A total of 5.5 kg of chlorfluazuron bait was consumed during phase 3 in a total of 14 stations, 6 of which had formosan subterranean termites that remained active to the end of phase 2 and beginning of phase 3.

Phase 4 (January 2004 to May 2005)

Formosan subterranean termites were found in a total of 28 trees located mostly in Bay Ridge. Most of the infested trees were live oak (*Quercus virginiana* Mill.), but they included a total of 11 different species and one unidentified stump (table 5). The majority of trees infested with *C. formosanus* were outside the experimental areas selected for phase 1. The distribution pattern of infested trees was inconsistent with the pattern of alate captures around Keesler Air Force Base (figure 4).

Termite activity was detected in stations installed around 22 (78.5 percent) of the 28 trees selected for phase 4. Formosan subterranean termites were detected in underground stations installed around 14 (63.6 percent) of the 22 trees that showed termite activity in the stations (table 5). This is equivalent to only a 50.0 percent success attracting formosan subterranean termites to at least one station installed near an infested tree, considering that *C. formosanus* were seen in all 28 trees selected for phase 4.

Approximately 11 kg of bait was consumed from a total of 75 bait stations visited by termites. Termites consumed a mean of  $147\pm14.4$  g of bait in each station and a mean of  $551.2\pm75.8$  g of bait in all of the stations installed in a tree. The mean bait consumed per

station was considerably lower in phase 4 than in phases 1 through 3, probably because phase 4 stations were placed closer to the trees.

Formosan subterranean termite activity disappeared from all stations installed in the 14 infested trees before the end of summer (figure 13). Boroscope inspection of trees revealed no termite activity inside the trees in September 2004. New formosan subterranean termite activity was detected in two stations from different trees in November 2004, and baiting resumed in these two stations. Termite activity disappeared in one of the two stations in February 2005, but the other station, located in tree "B," remained active to the end of phase 4 in April 2005. Native subterranean termite activity declined dramatically by July 2004, but some stations remained sporadically active to the end of phase 4 (figure 13). Only 3 of the 22 trees showed any termite activity by the end of phase 4. This represents a total of 86.0 percent reduction of subterranean termite activity and 92.8 percent reduction of formosan subterranean termite activity in treated trees within 14 months.

## Laboratory Comparison of Active Ingredients

At the end of 4 weeks, survival in all treatment groups was similar to that in the control group. At the end of week 6, diflubenzuron-treated groups continued showing similar survival levels as the control groups in all dose treatments. Chlorfluazuron groups showed 60.0, 30.0, and 50.0 percent survival for one, two, and three doses respectively and an overall survival of 46.6 percent at the end of this period. The control group showed 70.0 percent survival at the end of the 6th week (table 6).

At the end of 8 weeks the survival of the diflubenzuron groups continued to be similar to that of the control groups. The chlorfluazuron groups, on the other hand, showed higher mortality than the control groups at the end of 8 weeks (table 6). The overall survival for diflubenzuron groups at the end of 8 weeks was 63.3 percent and for chlorfluazuron groups, 16.6 percent; survival of the control group was 60.0 percent (table 6).

At the end of 10 weeks, diflubenzuron groups showed only a slight reduction of survival compared to the control groups. At the end of week 10 there was only one surviving termite group that was given chlorfluazuron, and it received only one dose. No surviving termite groups remained after 10

	Tree Trunk		Visited	Termite species observed*		
Tree species	ID	diameter	stations	Cf	Rf	Rv
		ст				
Quercus virginiana	А	68.33	1	+	-	-
Quercus virginiana	В	92.96	4	+	-	-
Quercus virginiana	С	89.41	2	+	-	-
Quercus virginiana	D	107.95	3	-	+	-
Acer sp.	Е	28.45	4	-	+	-
Carya illinoensis	F	38.10	2	-	+	+
Broussonetia papyrifera	G	33.78	0	-	-	-
Magnolia grandiflora	Н	52.32	0	-	-	-
Liquidambar styrasiflua	Ι	48.26	0	-	-	-
Quercus virginiana	J	98.04	3	+	+	-
Quercus virginiana	Κ	74.17	0	-	-	-
Quercus virginiana	L	101.35	4	+	+	-
Jnknown (stump)	Μ		0	-	-	-
Quercus virginiana	Ν	104.39	1	-	+	+
Quercus nigra	0	49.78	4	+	+	+
Quercus virginiana	Р	53.34	2	+	+	+
Fagus grandifolia	Q	48.77	5	+	+	-
Fagus grandifolia	R	33.53	4	+	+	-
Quercus nigra	S	50.80	3	+	+	-
Quercus nigra	Т	41.40	8	+	+	-
Pinus sp.	U	44.70	0	-	-	_
Carya cordiformis	V	40.13	4	-	+	-
Quercus virginiana	W	54.36	1	-	+	-
Quercus virginiana	Х	44.45	1	-	+	_
Quercus virginiana	Y	46.48	4	-	+	_
Quercus virginiana	Ζ	98.04	2	+	-	_
Quercus virginiana	AA	74.17	6	+	-	_
Quercus alba	BB	85.85	7	+	+	_

## Table 5. Termite activity in stations installed near trees during phase 4

Cf = Coptotermes formosanus, Rf = Reticulitermes flavipes, Rv = R. virginicus

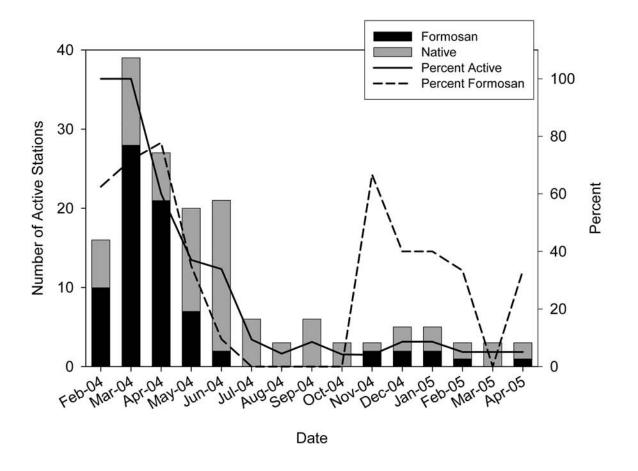


Figure 13. Formosan (black) and native (grey) subterranean termite activity observed in underground stations installed near trees during phase 4, February 2004 to April 2005. Solid line represents percentage of active stations with either species of termites calculated from the total number of stations visited by termites; dashed line represents percentage of formosan subterranean termite activity from total number of active stations.

	Su	rvival at	the end o	f	Dry weight consumption <sup><math>\dagger</math></sup>		
Treatment*	6 wk	8 wk	10 wk	16 wk	Wood	Bait	
		(	%		mg	g	
Control	70	60	60	60	3,975 ± 1,139a		
Diflubenzuron:							
1 dose	60	60	60	40	$2,535 \pm 654ab$	$360 \pm 47d$	
2 doses	90	70	50	40	$2,406 \pm 686$ abc	$660 \pm 95$ bc	
3 doses	90	60	40	0	$961 \pm 419$ bcd	$612 \pm 77$ bcd	
Chlorfluazuron	:						
1 dose	60	20	10	0	$856 \pm 235$ cd	$420 \pm 41$ cd	
2 doses	30	0	0	0	$371 \pm 108d$	$720 \pm 103$ ab	
3 doses	50	30	0	0	$273 \pm 130d$	972 ± 177a	

Table 6. Survival, wood consumption, and bait consumption of groups of 1,000 termite workers and 200 soldiers exposed to either diflubenzuron or chlorfluazuron at three dose levels

\* One dose was 1.6 g of bait containing 250 ppm active ingredient.

<sup>†</sup>Mean ± standard error of the mean. Means with the same letter are not significantly different at  $\alpha = 0.05$  after Student's *t* test.

weeks treatment with two or three doses of chlorfluazuron. The overall survival of the diflubenzuron, chlorfluazuron, and control groups was 50.0, 3.00, and 60.0 percent, respectively, at the end of 10 weeks. At the end of week 11 no termite group survived in any of the chlorfluazuron treatments.

At the end of the experiment (week 16), diflubenzuron treatment with three doses showed no survival of termite groups. However, diflubenzuron treatments with one and two doses still showed 40.0 percent survival. The control group showed 60.0 percent survival (table 6).

Wood consumption was different in the diflubenzuron groups and the chlorfluazuron groups. Diflubenzuron groups with one and two doses consumed more than twice the wood (by dry weight) than chlorfluazuron groups (table 6). The control group consumed significantly more wood during the same period than any of the treatment groups except diflubenzuron with one and two doses (table 6). Bait consumption (dry-weight basis) was higher in chlorfluazuron groups than in diflubenzuron groups (table 6). Chlorfluazuron groups with two and three doses consumed significantly more bait than wood (|t| = 2.35 and 3.13, respectively; df = 9) by the end of the 16-week experimental period. In contrast, diflubenzuron groups with one and two doses consumed significantly more wood than bait (|t| = 3.32 and 2.52, respectively; df = 9).

Total food consumption (wood + bait) in the chlorfluazuron groups was significantly lower than wood consumption in the control groups (F = 3.4; df 6, 63; P = 0.0057). This may be the result of significant mortality of termite workers in the chlorfluazuron groups. Only the three-dose diflubenzuron groups consumed significantly less food than the control groups. Diflubenzuron treatments with one and two doses had total consumption similar to that of the control groups. This may indicate

considerably lower mortality of workers in these two diflubenzuron treatments.

## Discussion

Results from this study show that bait systems can be used effectively to reduce subterranean termite populations areawide. Termite activity was significantly lower in treated neighborhoods than in untreated neighborhoods at the end of phase 1. However, complete areawide elimination of subterranean termites could not be achieved by this method. Results of phase 2 showed that the rate of termite suppression decreases every year, suggesting a plateau effect in suppression levels as time progresses. The change of the active ingredient from diflubenzuron to chlorfluazuron in phase 3 sharply increased suppression levels. This indicates that more powerful active ingredients can improve the effectiveness of areawide control of subterranean termites by baits.

Use of baiting systems as tools for areawide management of formosan subterranean termites is promising. Results of phase 3 suggested that formosan subterranean termites may be more vulnerable to baits than native subterranean termites. Their higher foraging activity and larger colonies produce larger foraging territories, increasing the probability of finding a given bait station. The higher consumption rates of formosan subterranean termites than their native counterparts expose their colonies to larger doses of the active ingredients in shorter periods. Rapid consumption of chlorfluazuron bait produced the collapse of large colonies of formosan subterranean termites in three months during phase 4.

This long-term study has shown the strengths and weaknesses of baiting systems as tools for areawide management of subterranean termites. Baiting systems allow detection of termite colonies before they produce structural damage and provide an effective and environmentally friendly means of controlling subterranean termites. However, baiting systems are labor intensive because of the need to install and continuously monitor the underground baiting stations. Current baiting systems do not attract termite workers to the stations; instead they rely on termites finding the stations through their constant foraging activity. As a result, only a small percentage of the underground stations are ever visited. Effectiveness of baiting systems depends in great measure on the strategic positioning of bait stations and their distribution around the infested area. This study showed that attention must be given to the presence of large trees when positioning bait stations. Our results showed that formosan subterranean termite infestations were strongly correlated with the presence of large trees (especially hardwood species) and that most infestations appear to have started in such trees. During phase 4, detection of formosan subterranean termites improved dramatically by directing search efforts to the inspection of large trees.

Laboratory studies have shown that formosan subterranean termites tend to prefer hardwood species as food over southern yellow pine species (Morales-Ramos and Rojas 2001). Also, laboratory studies on incipient colonies showed that formosan subterranean termites colonies are more likely to survive if they feed on oak, maple, sweet gum, sycamore, willow, etc. than if they feed on yellow pine during the first 3 years after the colony's foundation (Morales-Ramos and Rojas 2003b). Assuming that formosan subterranean termites are able to locate their preferred food sources, colonization is most likely to begin in large hardwood trees. Therefore, it is reasonable to assume that formosan subterranean termites probably infest large hardwood trees first and attack housing structures only after their colonies become large enough to expand to new locations.

This study shows that the pattern of distribution of formosan subterranean termites on the Keesler Air Force Base was strongly correlated to the presence of large trees and not to the presence of houses. Stations installed in a regular pattern around houses failed to detect formosan subterranean termites in all but one of the 12 neighborhoods in phase 1.

Effectiveness of baiting systems is also related to the choice of active ingredient. Our laboratory study showed that chlorfluazuron was considerably more effective than diflubenzuron in reducing termite survival in a shorter period. Phase 4 showed a dramatic collapse of formosan subterranean termite activity within only 3 months of introducing the chlorfluazuron bait. If chlorfluazuron had been used during phase 1, the levels of subterranean termite suppression could have been much greater.

Though both chemicals belong to the same family of compounds and have the same mode of action,

chlorfluazuron seems to act considerably quicker than diflubenzuron. Laboratory studies confirmed this by showing significantly higher mortality of formosan subterranean termite groups subjected to chlorfluazuron than of those subjected to diflubenzuron. Data also show that mortality occurred earlier in groups treated with chlorfluazuron than in those treated with diflubenzuron.

The slower action of diflubenzuron may allow termite colonies to recover during periods of low foraging activity. Termite foraging activity and bait consumption decline during winter, but formosan subterranean termite colonies living in trees have access to a large food supply during this time.

Another mechanism of recovery may be gradual depletion of those termite workers affected by the treatment. This diminishes foraging activity and eliminates bait consumption in the colony before the reproductive pair is killed. This would result in a relatively rapid recovery because younger termite workers may still be able to forage inside their toxin-free tree.

A third factor in effectiveness of baiting systems is the relative palatability of the bait matrix. Simple matrices such as cardboard and paper may be effective at eliciting termite foraging where the alternative food choices are limited, such as in housing structures. To entice termite colonies living in trees, though, the bait matrix has to be much more sophisticated. The bait matrix used in this study was developed after research into formosan subterranean termite feeding preferences and chemical analyses of the most preferred wood species (Morales-Ramos and Rojas 2003a).

Successful delivery of a toxic active ingredient via bait in an areawide management system will depend greatly on formulating a matrix that can compete for foraging termites with a variety of highly preferred tree species present in the urban environment. This also applies to the wood species used for monitoring termite activity in the stations. Morales-Ramos and Rojas (2001) determined that yellow pine wood is not preferred by the formosan subterranean termite. Monitoring stations using yellow pine installed near infested trees may not stimulate termite foraging and so would remain inactive, allowing termites to escape treatment. Changing yellow pine to aspen wood increased the number of stations visited by formosan subterranean termites by more than threefold in a field study near oak trees in New Orleans (J. A. Morales-Ramos, 2007, unpublished).

Choice of active ingredient may also affect palatability of the bait. Our laboratory experiment showed that termite groups consumed more red oak wood than diflubenzuron bait, but similar groups consumed significantly more chlorfluazuron bait than red oak. The same study showed that termite groups consumed significantly more chlorfluazuron bait than diflubenzuron bait. This seems to indicate that diflubenzuron, while not repellent, reduces palatability of the bait. Chlorfluazuron, on the other hand, does not seem to affect palatability.

This study also provided the opportunity to compare two methods for evaluating termite control treatments: use of uniformly distributed independent monitor stations versus use of untreated areas as controls. Independent monitors have been used successfully to evaluate bait treatments in structures. Such studies were preceded by mark-release-recapture methods to determine colony identity. Areawide studies cannot depend on determination of colony identity by the mark-release-recapture method because of the size of the area treated and the potentially high number of termite colonies present. In an areawide management study, treatment comparisons measure the effect on populations of colonies rather than on a few colonies.

At the end of phase 1, termite activity was significantly higher in untreated neighborhoods. However, independent monitors showed a considerable discrepancy in termite activity than normal stations in the treated neighborhoods (figure 6). This discrepancy was not observed in untreated neighborhoods. As a consequence, results of statistical comparisons of termite activity between treated and untreated neighborhoods differed between normal stations and monitoring stations (figures 7 and 8). There are two possible explanations for this discrepancy:

- Activity in monitoring stations represents foraging activity by different colonies than those present in normal stations and therefore those colonies remained untreated through the study; or
- Normal stations show reduced activity because termites avoid them after treatment.

Observations of termite activity in individual treated stations through phases 1 and 2 showed that a large number of stations remained active for more than a year during treatment. Furthermore, other stations in which termite activity ceased after treatment showed renewed activity a few months later. This evidence contradicts the notion of termite avoidance of treated stations. Because treatment in most of the neighborhoods was directed against native subterranean termites, the first explanation is more likely because termites of the genus Reticulitermes produce smaller colonies than *C. formosanus*. In many cases, normal stations adjacent to active monitoring stations were never visited by termites. In other cases, monitoring stations were active with a different species of Reticulitermes than those present in adjacent normal stations.

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# Distribution of Stations and Termite Activity in Neighborhoods

#### page

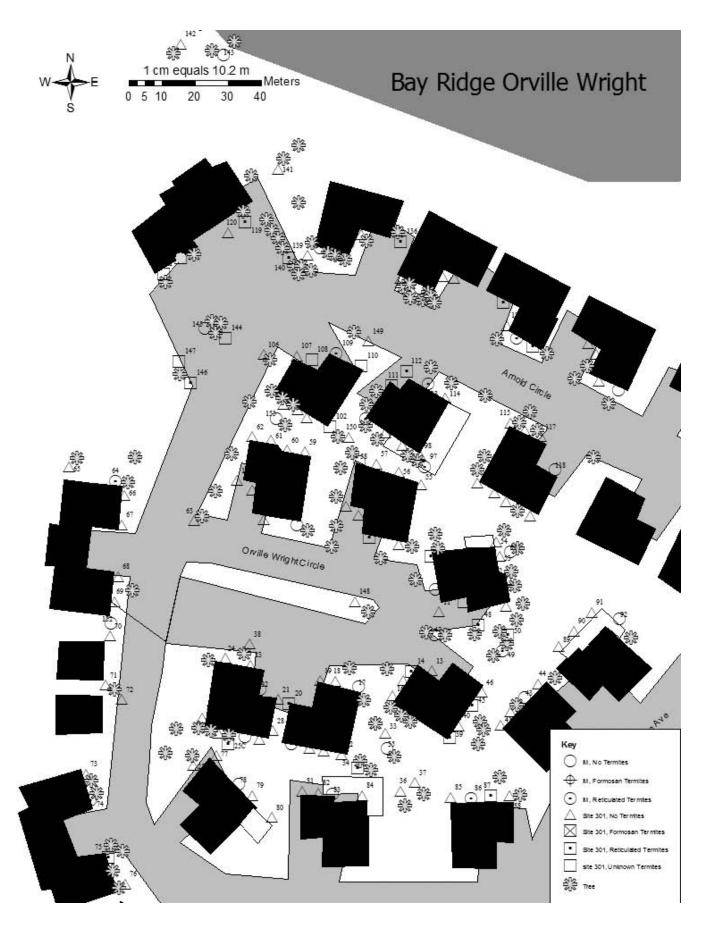
### **Baited neighborhoods**

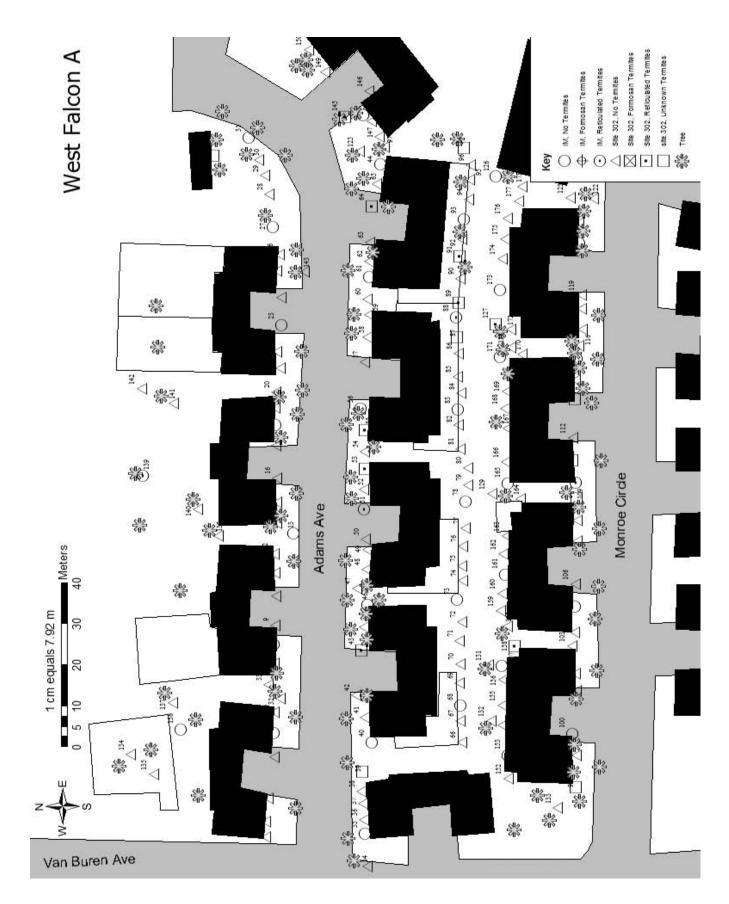
Bay Ridge Fechet	
Bay Ridge Orville Wright	
West Falcon A	
West Falcon B	
Shadylawn	
Sun Coast Villa A	

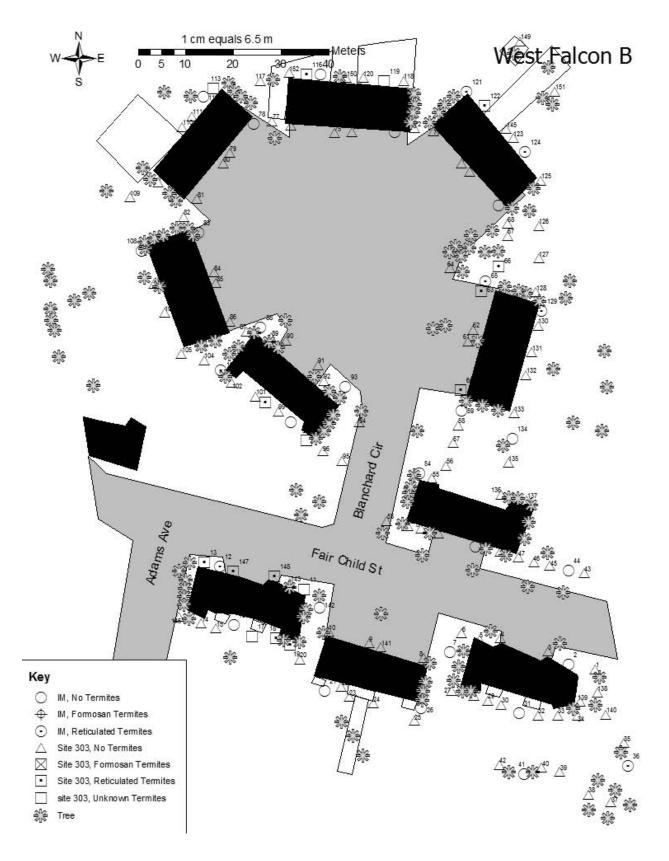
### Nonbaited (control) neighborhoods

East Falcon A	
East Falcon B	
Oak Park	
Pinehaven A	
Pinehaven B	43
Sun Coast Villa B	















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