Urban Ecosystems, 4: 231–251, 2000


# Subterranean termites in the urban landscape: Understanding their social structure is the key to successfully implementing population management using bait technology

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**Abstract.** Subterranean termites will likely continue to be major economic insect pests in the urban environment. Public concerns about pesticide use will not abate but continue to drive development and implementation of environmentally more acceptable control strategies for subterranean termites. Termite baits are a promising alternative to soil termiticide treatments for protection of structures, but the correlation between killing termites in the landscape and protecting structures is tenuous given our current understanding of termite biology. We discuss the problems associated with defining the efficacy of termite bait products in regard to determining termite population parameters in the field. This discussion is in the form of six case histories involving measures of termite activity and relatedness taken over a three-year period. Our findings highlight the need to include control populations in studies of termite bait efficacy, the mobility of termite populations over time, the importance of defining the techniques used to describe termite colony associations, and the need to use a multidisciplinary research approach in addressing the question of termite social structure.

**Keywords:** Rhinotermitidae, termite baiting, termite social structure, termite control

## Introduction

Subterranean termites, particularly members of the genera *Reticulitermes* and *Coptotermes*, represent the most widespread and economically important structural insect pests in the urban environment (Gay, 1969; Su and Scheffrahn, 1990). In their native woodland habitats subterranean termites are a valued part of the ecosystem (Lee and Wood, 1971; Waller and LaFage, 1987). However, when they take advantage of a ready-made food resource in the form of the wood used in building construction, subterranean termites are afforded pest status (Kofoid, 1934; Forschler, 1998a). With increased urban expansion into woodland habitats this conflict of interest concerning lumber utilization will continue and likely increase in scope.

Many species of Rhinotermitid termites live in the source of their food, wood and wood products. Human commercial activities can, therefore, unwittingly transport them to nonendemic areas. This is evidenced by the appearance of *Reticulitermes* infestations in

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Hallein, Austria, Hamburg, Germany, Devon, England, and Toronto, Canada and of *Coptotermes* in the southeastern United States, South Africa, and Japan (Gay, 1969; Su and Tamashiro, 1987; Forschler *et al.*, 2000; Jenkins *et al.*, 2001). Thus subterranean termites worldwide will continue to be a problem for urban and suburban property owners. Control efforts and damage repair due to subterranean termites annually exceeds $1 billion in the United States alone (Su and Scheffrahn, 1990). More than two billion U.S. dollars were spent according to the last worldwide survey of annual termite control costs, which was conducted over 15 years ago (Edwards and Mill, 1986). That figure is certainly larger today.

In urban areas, subterranean termite control has traditionally involved use of termiticidal soil barriers (Brown *et al.*, 1934; Edwards and Mill, 1986; Beal *et al.*, 1989; Moore, 1990; Potter, 1997). The aim of a termiticidal soil barrier is to exclude termites from the structure with no regard toward impacting populations. The concept is sound. In practice, however, it is extremely difficult to obtain a continuous and uniform distribution of insecticide at the correct concentration to the soil around a structure (Gold *et al.*, 1996; Forschler and Lewis, 1997). The amount and concentration of insecticides used in soil barriers for subterranean termite control exceeds the use of similar chemistries in an agricultural setting (Gold *et al.*, 1994). These factors combined with an increased awareness of and desire to reduce pesticide use in the urban environment have fostered the reemergence of low-insecticide input control strategies against subterranean termites. The result has been an increase in the commercial use of termite baiting as a control tactic. A termite bait-control program is a radical departure from exclusionary tactics because it attempts to manage populations (Beard, 1974; Beal and Esenther, 1980; Forschler, 1998b). The assumption is that when there are few or no termites in the vicinity of a structure it is protected from infestation. Again the concept is sound. Yet the correlation between killing termites detected near a building and protection from infestation is, in practice, tenuous (Forschler, 2000).

Measuring the efficacy of a subterranean termite population-management tactic is difficult (Forschler, 1996; Forschler and Jenkins, 1999; Thorne and Forschler, 2000). Elucidating the lifestyle of a cryptic eusocial insect that lives in organized groups containing hundreds of thousands of individuals is not easy. This is especially true with *Reticulitermes* subterranean termites whose life-forms, or castes, inhabit a diffuse network of tunnels connecting two or more feeding sites and that have no central nest site—the reproductives being mobile (Forschler, 1996; Thorne, 1998; Forschler and Robinson, 1999; Forschler and Jenkins, 1999). Research designed to determine subterranean termite bait efficacy generally involves three elements.

First, several locations, essentially termite feeding sites, are established from which measures of termite activity can be recorded.
Second, the termiticidial bait is applied at (or near) one or more of those locations.
Lastly, termite activity is recorded at all locations over time.

A successful bait trial is assumed when activity is reduced or eliminated at all locations identified as being visited by the targeted termites (Su and Scheffrahn, 1996; Thorne and Forschler, 2000). Techniques available for measuring termite activity and therefore

delineating their populations are, however, limited in scope (Jones, 1990; Forschler, 1996; Thorne *et al.*, 1996; Forschler and Robinson, 1999; Forschler and Jenkins, 1999). Researchers and termite control practitioners can only observe termite activity through one or more "windows." At best, permanent observation posts, herein termed inspection ports (IPs), are established to record a time line of activity. This limited view of the diffuse network of tunnels and feeding sites occupied by a termite population is at the heart of the problem of determining their population parameters. Conclusions, therefore, are based on interpretation of data that is largely inferential and founded in the experience of the observer (Forschler and Jenkins, 1999).

Recent studies of subterranean termite field biology and bait efficacy have assigned "colony" associations using mark-release-recapture (MRR) data (Su *et al.*, 1993; Su, 1994; Forschler, 1994; Haagsma and Rust, 1995; Su *et al.*, 1995; Grace *et al.*, 1989; Grace *et al.*, 1996; Forschler and Townsend, 1996; Forschler and Ryder, 1996). This field technique involves collecting termites from a single IP, marking, and then releasing them back into the same IP. Following a prescribed time period, all of the nearby IPs are examined for the presence of marked termites. The researcher connects the dots when marked termites are found at an IP away from the release site and assumes that these termites constitute a colony. Yet MRR has come under criticism as unsuitable for determination of either termite population estimates or colony associations (Thorne *et al.*, 1996; Evans *et al.*, 1998; Forschler and Jenkins, 1999). These criticisms center around indications that termites released at one feeding site (IP) do not equitably redistribute themselves within the population, negating an assumption of MRR models, and that colony associations are more complex than can be determined by the presence/absence of a few marked termites. Other techniques employed include assignment of colony associations based on morphometric similarities and distance between feeding sites (Haverty *et al.*, 1975; Howard *et al.*, 1982). Jones (1990) suggested combining MRR and agonism bioassay for delineation of subterranean termite field colonies. Still others have used chemotaxonomic phenotypes or isoenzyme analysis in combination with morphometrics and agonism bioassays (Clement, 1986; Haverty *et al.*, 1999). Comparison of relatedness using genetic analysis and F statistics were conducted by Reilly (1987), and recent studies have used gene sequence data (Jenkins *et al.*, 1999). But the technique(s) used in a particular study and inference remain the driving forces in describing termite "colonial" relationships (Forschler and Jenkins, 1999).

We here report results from work in which six separate termite populations were delineated using MRR, morphometric characters, agonism bioassay, hydrocarbon phenotypes, and a genetic marker. This work highlights the difficulties encountered in ascribing colony attributes to subterranean termites in the field (Forschler, 1996; Jenkins *et al.*, 1999). The term "population" is therefore used to describe those groups of termites identified using a combination of techniques as visiting one or more IPs. This distinction is made because we report results that raise questions concerning the determination of subterranean termite colony parameters. Six case histories, that involve a potential bait active ingredient, fipronil, are detailed as a backdrop for discussion of the importance of termite social structure as regards commercial claims and implementation of baiting tactics as a subterranean termite management tool.

## Materials and methods

All of the termite populations involved in this study were located on Sapelo Island, Georgia. Six separate termite populations were selected because they consistently visited previously established termite IPs from 1994 to 1997. All IPs are designated by a site name and numbered in the sequence in which they were established at that site—not according to physical proximity. The termite IPs, as described in more detail in Forschler and Townsend (1996), were 16 cm lengths of 10 cm diameter PVC pipe sections buried in the ground. A feeding and aggregation substrate was placed inside and the top was capped with a plastic "knock-out plug." One type of feeding and aggregation substrate consisted of ten pieces of weathered pinewood (4 cm by 12 cm by 2 mm) separated by dowel sticks and held together with an 18 cm length of plastic cable tie (herein called a pine sandwich). The other consisted of a roll (12 cm by 295 cm) of corrugated cardboard. One roll of cardboard or two pine sandwiches were placed per IP.

### *Measures of termite activity*

Prior to this study each site was visited monthly from April of 1994 through January of 1997. During 1997, each IP was examined every four months—in January, April, August, and December. They were examined at least every third month from March of 1998 to October 1999; monthly from March through May 1998; in August, October, and November 1998, and monthly from February through October 1999. At the time of each inspection, the aggregation and feeding substrate was removed and returned to the laboratory. The number of termites, the average live weight per individual termite, caste proportions, and the amount of wood or cardboard consumed were measured for each IP on every collection date. Caste proportions were determined by separating the individual castes (soldiers, workers, larvae, nymphs) and either counted or their number estimated by taking an average weight of 5 groups, 10 termites each, and dividing this average into the total weight of termites collected. Cardboard or wood consumption was measured by drying the substrate in an oven at 70°C for 2 or 72 h, respectively, then reweighing after acclimation to room temperature in a desiccator. The oven-dried weights of the wood before and after placement within an IP were used to calculate the amount of wood consumed. For cardboard, oven-dried weights were taken after placement in the IPs.

### *Characterization of termite populations*

Related use of IPs was first determined during 1994 and 1995 using fluorescent spray paint as a topical mark (Forschler, 1994). Further MRR work was conducted in 1995, 1996, or 1997 using an internal mark, the fat-soluble dye Nile Blue A, for estimation of population numbers (Forschler and Townsend, 1996). In addition, over the two years prior to the initiation of this study, termites collected from IPs were characterized using morphometric phenotypes and hydrocarbon profiles (Scheffrahn and Su, 1994; Haverty *et al.*, 1996). Using these data in combination we were able to assign groups of two or more IPs to a particular termite population based on related use as indicated by MRR and similarities

in morphometric or chemophenotypic characters. We also attempted agonism bioassays, but none of the termites from within these populations displayed any aggression toward one another. Because agonism data obtained with *Reticulitermes* in the southeastern United States are often equivocal, we seldom rely on this information when designating populations (Polizzi and Forschler, 1998).

At the time of each collection, randomly selected termite workers and soldiers were placed in 70% ETOH as vouchers. Soldiers were used for morphometric characterization of species and worker termites for genotypic characterization (Scheffrahn and Su, 1994; Jenkins *et al.*, 1998). DNA was extracted from three to seven randomly selected individual termites from each IP for selected dates. From these individuals part of the mitochondrial cytochrome oxidase I (COI) gene was sequenced (Jenkins *et al.*, 1999). This genetic marker allows for identification of maternal lines (MLs) because the mitochondrial genome, and therefore the conserved COI gene, is inherited directly from the mother. It is generally believed that a termite colony is initiated by one adult female reproductive (queen) and that all the individuals within that colony are the offspring of that single mother (Thorne *et al.*, 1996). Based on this monogyne concept of a termite colony we selected the mitochondrial genetic marker to delineate populations.

### *DNA extraction, amplification, sequencing, and data analysis*

DNA extraction was accomplished according to Liu and Beckenbach (1992) and Jenkins *et al.* (1999) on individual whole worker termites preserved in 70% EtOH. DNA templates consisted of total nucleic acids (Jenkins *et al.*, 1999).

Oligonucleotide COI primers (C1J2195, TTGATTYTTTGGTCAYCCWGAAGT and TL2N3014, TCYAWTGCAYTAATCTGCCATATTA) were used to polymerase chain reaction (PCR) amplify as well as to prime forward and reverse sequencing reactions. COI primers are according to Liu and Beckenbach (1992) with modifications designed from Simon *et al.* (1994). Primer names and strand identification (J for majority strand and N for minority strand) are according to Simon *et al.* (1994).

PCR amplification was performed in a standard 25 $\mu$l reaction with a minimum of 10 ng of total genomic DNA, 1 pmol of each primer, 2.0 mM $MgCl_2$, 1.6 mM dNTPs, and 0.06 U/$\mu$l *Taq* DNA polymerase. Amplification was accomplished in a Perkin–Elmer Gene Amp PCR system 9600 (PE Applied Biosystems, Foster City, CA, USA). It included a precycle denaturation at 94°C for 2 min, a postcycle extension at 72°C for 7 min, and 25 cycles of a standard three-step PCR (50°C annealing). Fragments were treated with exonuclease I (10 U/$\mu$l) and shrimp alkaline phosphatase (1 U/$\mu$l). They were then incubated in a Perkin–Elmer GeneAmp PCR System 9600 first at 37°C for 15 min, then at 80°C for 15 min (Jenkins *et al.*, 1999). DNA (10–20 ng/100 bp PCR product) sequencing reaction was performed with the Dye-Terminater Cycle Sequencing Kit (PE Applied Biosystems) in a GeneAmp 9600 PCR system. Electrophoresis was accomplished on a 6% polyacrylamide gel. Reactions were fractionated and initial base assignments were made by the ABI 373A automated DNA sequencer system (PE Applied Biosystems, Foster City, CA, USA).

Sequencer 3.1.1 software (Gene Codes Corp., Ann Arbor, MI, USA) was used to edit individual electropherograms and make contigs. Multiple individuals were processed per

collection date and site. Following alignment of sequence data (forward and reverse) consensus sequence was determined. Consensus sequence was assigned for the partial COI gene when sequences from individuals were the same using Sequencer 3.1.1 software. For the purpose of this manuscript a maternal line (ML) is defined as a unique consensus sequence.

### *Treatments*

Fipronil, a phenyl pyrazole chemistry that interferes with operation of the central nervous system by blocking the GABA (gamma-aminobutyric acid) regulated chloride channel in nerve cells was the insecticide used in these bait trials (Cole *et al.*, 1993). Sections of fipronil-treated cardboard (12.7 cm by 295 cm), at one of six rates, 0, 0.1, 0.5, 1, 5, and 10 ppm, were provided by the manufacturer, Aventis Environmental Science (Montvale, NJ). Treated cardboard was introduced into one IP for each termite population on 11 March 1998. Three populations received untreated cardboard in an IP on the same date as a control to ensure the cardboard used was not repellent. The remaining IPs, for all populations, continued to receive untreated pine sandwiches. On 21 April 1998 the aggregation and feeding substrates were removed for extraction of termites and calculation of the amount of cardboard or wood removed. The IPs that received cardboard in March were provisioned with new cardboard (both treated and untreated) on 21 April 1998. On 21 May 1998 the feeding and aggregation substrates were again removed and all cardboard replaced with untreated pine sandwiches. All IPs were provided, from May 1998 onward, with untreated pine sandwiches. Therefore all termite populations were exposed to fipronil-treated or untreated cardboard for only 10 weeks (the middle of March to the end of May—71 days). A summary of treated cardboard placements and amount removed is provided in Table 1.

## Results

Results will be discussed as separate case histories for each population. To save space we do not provide all of the termite activity data such as caste proportions, numbers of termites

*Table 1.* Summary of treated cardboard placements and amount of treated cardboard consumed by population and fipronil concentration.[a]

| Designated population number | Treated cardboard placed in IP#[a] | Fipronil rate (ppm) | Amount of treated cardboard consumed (g) |
|---|---|---|---|
| 1 | NA[b] | 0 | NA |
| 2 | 6 | 0.5 | 0 |
| 3 | 20 | 0.1 | 4 |
| 4 | 2 | 1 | 23 |
| 5 | 10 | 10 | 0 |

[a]IP# = inspection port number.
[b]NA = not applicable.

collected, wood or cardboard consumption rates per inspection date—unless important to the discussion of a particular site or point of discussion. A brief synopsis of the pertinent characteristics of each of the populations follows.

### *Population 1*

This population was not provided treated cardboard and is therefore considered the control. Population 1 was identified as occupying six IPs along the south wall of the solarium at the Reynolds Mansion study site (figure 1). The species was determined to be *Reticulitermes hageni* Banks based on soldier morphometric characters and unknown based on hydrocarbon phenotype (Scheffrahn and Su, 1994; Haverty *et al.*, 1996). During the final round of MRR (1996–1997) using Nile Blue A, approximately 2000 marked termites were released into IP #21 (September 1996), and marked termites were consistently recovered over the next four months from IPs #21 and #24 (figure 2). Marked termites were recovered only once from IP #28 (October 1996) and never from IP #19 although we collected termites only once (January 1997) from IP #19 during the course of this MRR study (figure 2). Inspection port #19 was included in our MRR estimate of colony association for three reasons. First, IP #19 was positioned between IPs #28 and #21 where marked termites were recovered (figure 1). Second, the recovery rate of marked termites is usually very low, generally less than 1% (Forschler and Townsend, 1996). We assumed that given more chances (through collecting termites more often than once in eight months) a marked termite would have been found at that site. Third, the same species was recovered from all of the IPs assigned to this particular population. Inspection ports #9 and #10 were included because the morphology

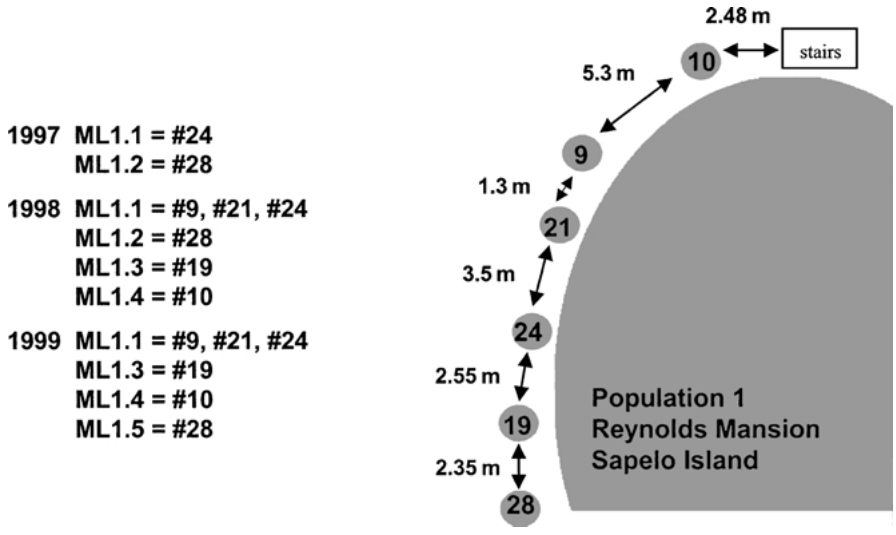


*Figure 1.* Location of inspection ports (IPs) for subterranean termite Population 1 along the south wall of the solarium at the Reynolds Mansion test site and timeline of maternal lineage (ML) associated with them.

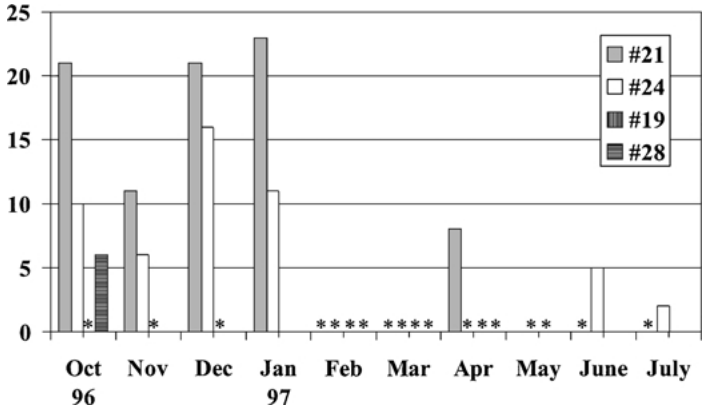


*Figure 2*. Bar graph of the number of marked termites collected by month and inspection port following release of Nile Blue A marked termites into Population 1 inspection port #24 in September 1996.—An asterisk indicates that no termites were collected from that inspection port during that month.—Bars representing each inspection port are aligned within each month on the graph coincidental with the physical proximity of the inspection ports (see figure 1).

of the termites collected from those locations changed species from *R. flavipes* (Kollar) to *R. hageni* during 1998. We assumed that termites from the nearby MRR-connected IPs simply expanded their range of feeding sites. The population estimate conducted in 1996 indicated approximately 72,000 termites. The maternal lineages (MLs) varied (figure 1). The ML (1.1) found in IPs #21 and #24, where marked termites were consistently recovered, was exactly the same for three years (1997–1999). This same ML (1.1) was found in IP #9 during 1998 and 1999 indicating our assumption that the designated population expanded their feeding sites in 1998 was justified. However, IP #10 contained a different ML (1.2) during 1998 and 1999. Inspection port #19, where we found no marked termites during the 1996–1997 MRR attempt, consistently contained yet another ML (1.3) during 1998 and 1999. Inspection port #28, where we recovered marked termites only once during the MRR study, provided a further ML (1.4). In 1999 the termites collected in IP #28 were determined, morphometrically and genotypically (ML 1.5), to be a different species, *R. flavipes* (figure 1). Thus with our control group we recorded two different species having occupied, at one time or another, three of the six IPs in the three years of this study. In addition, the *R. hageni* population, which was the focus of our population characterization, was represented by four different MLs that for at least two consecutive years were consistently collected from the same IP (figure 1).

### *Population 2*

Population 2 occupied three IPs along the southeast side of the Reynolds Mansion study site as identified by spray paint marking conducted in 1994 (figure 3). The species was determined to be *Reticulitermes hageni* based on soldier morphometric characters and unknown based on hydrocarbon phenotype (Scheffrahn and Su, 1994; Haverty *et al.*, 1996).

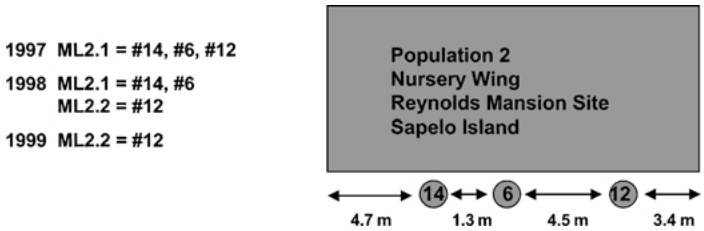


*Figure 3*. Location of inspection ports (IPs) for subterranean termite Population 2 along the southeast wall of the Nursery at the Reynolds Mansion test site and timeline of maternal lineage (ML) associated with them.

The morphology of the termites collected from these three IPs remained the same during the course of our study. We released approximately 1000 marked termites into IP #6 in 1996. However, no marked termites were recovered from any of the inspection ports. Therefore we have no population estimate. In addition, no consumption of treated cardboard was recorded during the 71 days the treatment was present in IP #6. Termites were collected from IP #6 twice during the bait study, in April and August 1998, after which no termites were collected through October 1999 when the study was terminated. Termites were consistently collected from IP #14 from April through August 1998 after which time no termites were collected from this IP. Termites were collected from IP #12 every month from April 1998 through July of 1999. Yet no termites were collected from July through October 1999 when the test was terminated. The ML data showed that during 1997 all three IPs contained a single ML (2.1) (figure 3). In 1998 IPs #6 and #14 contained the exact same ML (2.1). During 1998 and 1999 IP #12 provided a second, different ML (2.2) (figure 3). This population, identified by MRR in 1994 (yet unsuccessfully in 1996) provided consistent species characters from 1994 through 1999 and was represented by one ML (2.1) in 1997, two MLs (2.1 and 2.2) in 1998, and, following treatment, one ML (2.2) in 1999 (figure 3).

### *Population 3*

Population 3 was identified as occupying three IPs along the northeast side of the Reynolds Mansion study site as indicated by spray paint marking conducted in 1995 (figure 4). The species was determined to be *R. flavipes* based on soldier morphometric characters and hydrocarbon phenotype (Scheffrahn and Su, 1994; Haverty *et al.*, 1996). The population estimate conducted in 1995 indicated approximately 500,000 termites. Four grams of fipronil-treated cardboard were removed by termites in April 1998 (Table 1). In April and May 1998 there were thousands of dead and dying termites observed in IP #20 at the time of collection. After May of 1998, no termites were collected in any of the three IPs assigned to this population with the exception of 47 termites collected from IP #20 in October 1998. This population as delineated by MRR in 1995 provided a consistent single-species phenotype through 1998. However, immediately prior to initiation of the bait study, after January 1998, no termite activity was recorded at IPs #7 and #23 (figure 4). During the bait study (from March 1998 to October 1999) live termites were collected only twice from one IP

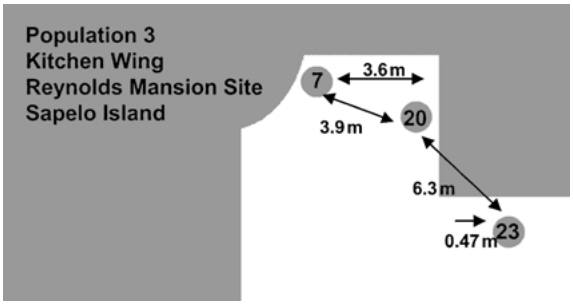


*Figure 4*. Location of inspection ports (IPs) for subterranean termite Population 3 along the northeast wall of the Kitchen at the Reynolds Mansion test site.

(#20), once in March 1998 (2750 termites) and once in October 1998 (47 termites). No ML data were determined from this population because the treatment was considered effective in eliminating all signs of termite activity. This case history represents the best-case scenario for a successful termite bait-control attempt.

### *Population 4*

Population 4 occupied 6 IPs along the south side of the Marine Institute Museum study site as identified by spray paint marking in 1994 and again in 1995 (figure 5). The species was determined to be *R. flavipes* based on soldier and alate morphometric characters, and hydrocarbon phenotype (Scheffrahn and Su, 1994; Haverty *et al.*, 1996). The population estimate conducted in 1995 indicated approximately 640,000 termites. Termite activity was consistent with no one IP displaying less than four consecutive months without evidence of termite activity between 1995 and initiation of the bait trial in March 1998. Twenty-three grams of treated cardboard were removed from IP #2 during April and May of 1998 (Table 1). During the course of the bait study termites were never collected from IP #6. However, termites were collected from IPs #1, #2, and #5 during March 1998, IPs #3 and #4 during April 1998, and IPs #3 and #5 during May 1998, after which no termites were collected for the remainder of the experiment (figure 5). Although termites were not collected from any of these six IPs after May 1998, we did have evidence of termite activity at IP #2 during October 1998. This activity was indicated by the presence of mud tubes and wood weight loss from the pine sandwiches. Because termite activity ceased at all IPs and we could claim a bait-treatment, effect no ML data were collected from this population. This population delineated using both MRR and consistent phenotypic characters is another classic termite bait success story.

### *Population 5*

Population 5 occupied 3 IPs along the southwest side of the Marine Institute Breezeway study site as indicated by spray paint marking conducted in 1996 (figure 6). The species was determined to be *R. hageni* based on soldier morphometric characters and unknown based

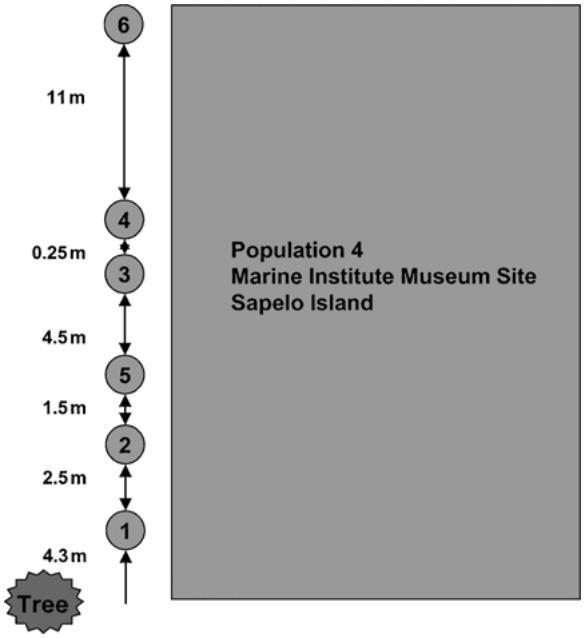


*Figure 5*. Location of inspection ports (IPs) for subterranean termite Population 4 along the south wall of the Marine Institute Museum test site.

on hydrocarbon analysis (Scheffrahn and Su, 1994; Haverty *et al.*, 1996). No population estimate was conducted. Termite activity was consistent, never more than two consecutive collection dates without termites, for all three IPs between January 1996 and January 1998. Termites were collected from all IPs in March 1998. Thereafter we collected no termites from IP #10, where the treatment was placed, for the next fourteen months, until May 1999. Termites were collected from IP #11 every month from March through May 1998, yet thereafter none were collected until June 1999. Termites were consistently collected

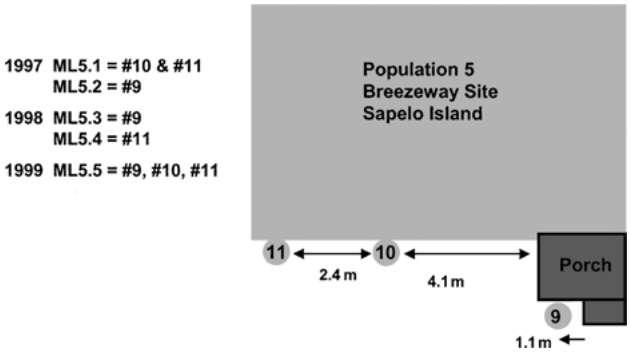


*Figure 6*. Location of inspection ports (IPs) for subterranean termite Population 5 along the southwest wall of the Breezeway test site and timeline of maternal lineage (ML) associated with them.

from IP #9 for every month from March 1998 to October 1999 with one exception, May 1998. From June 1999 to the end of the study (October 1999) termites were consistently collected from all three IPs. There was no evidence that termites removed any of the treated cardboard (Table 1). The ML data indicated that in 1997 termites collected from IPs #10 and #11 had the same ML (5.1) yet a different lineage (ML 5.2) occupied IP #9 (figure 6). In 1998, termites collected from IPs #9 and #11 represented two new MLs—5.3 and 5.4 (figure 6). In 1999 the termites in all three IPs were of the same ML (5.5) but different from the previous lines (figure 6). This population represents the most intriguing case for assigning population parameters.

Delimited by MRR in 1996, the soldier morphology consistently indicated *R. flavipes* over the next three years, and the classic assumption would be that this population represented one colony. Yet five separate MLs were present in this population from 1997 to 1999 (figure 6). Two lineages were found in 1997, one in IPs #10 and #11 (ML 5.1) and a second in IP #9 (ML 5.2). In 1998 we unfortunately did not retain any vouchers from the 651 termites collected from IP #10 on 11 March 1998, the only date when termites were collected at this IP in that year. Yet the termites collected from IPs #9 and #11 in 1998 provided two new MLs (5.3 and 5.4) (figure 6). In 1999, all three IPs provided the same ML (5.5), yet this lineage was different than all the previous MLs collected (figure 6).

### *Population 6*

Population 6 occupied 2 IPs along the northeast side of the Marine Institute Office Building study site as identified by spray paint marking conducted in 1995 (figure 7). The species was determined to be *R. flavipes* based on soldier morphometric characters and hydrocarbon phenotype (Scheffrahn and Su, 1994; Haverty *et al.*, 1996). A population estimate conducted in 1995 indicated approximately 180,000 termites. Termites were never collected from either

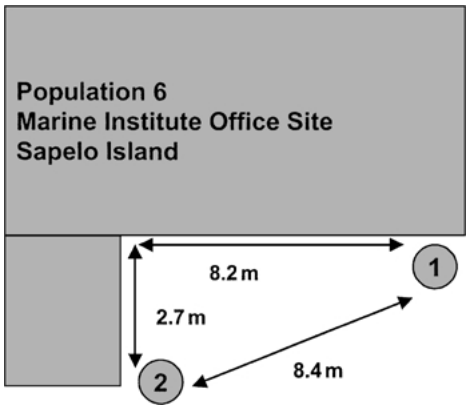


*Figure 7.* Location of inspection ports (IPs) for subterranean termite Population 6 along the northeast wall of the Marine Institute Office test site.

IP during the course of this study. The impact of our treatment on Population 6 is a moot point because we never collected termites in the 4 months prior to, or for 17 months after, bait application, although we recorded consistent termite activity at this site from 1994 to 1997 (figure 7).

## Discussion

Research on management of *Reticulitermes* populations is essentially the study of unreplicated case histories (Forschler, 1996; Forschler and Jenkins, 1999). Treatments can be replicated, but each population is unique, and finding an exact duplicate is difficult if not impossible. Such studies also require a long-term commitment of time and resources that make it difficult to manage large numbers of replicates. This study includes, for the first time, two significant approaches for better understanding the impact of bait technology on subterranean termite populations. First, we recorded data from an untreated population, the control, for monitoring "normal," seasonal or weather-related changes in activity and movement. Second, we used a genetic marker, the maternal mtDNA COI gene sequence, to identify genotypes prior to and following treatment in combination with MRR and traditional phenotypic markers. Although the case histories reported herein were conducted on an island, the data are no different from what we have collected from mainland urban sites at Aldora, Atlanta, and Athens, Georgia (Forschler, 1994; Forschler and Ryder, 1996; Forschler, unpublished data).

These case histories indicate that fipronil-treated cardboard has potential as a population management tool. Yet, as these case histories highlight, there are often difficulties in determining population impacts following implementation of a termite bait trial. We can claim success in reducing termite populations using fipronil-treated bait in as few as two or as many as four of the case histories depending on the criteria used to determine population parameters.

Population 1, the control population, illustrates many of these difficulties (figure 1). First, this case history reiterates the potential for movement and the temporal nature of the data collected during field studies. One should not assume, as we have in this study—and in the past—that an established feeding site (IP) will harbor the same termite population over time. A population shift is most apparent when there is a change of species that can be elucidated through examination of phenotypic characters (Su and Scheffrahn, 1988). In what has been considered an extreme case, we reported one IP that was occupied by three different species over a three-year period (Jenkins *et al.*, 1999). Is such movement an anomaly because it is rarely reported in the literature? Without concurrent untreated controls this question cannot be answered.

Our control group demonstrated considerable population movement between IPs. One IP (#28) connected by MRR, in 1996, to two others (#21 and #24), was occupied by a different species in 1999 compared to those found during 1996 to 1998 (figure 1). In addition, two IPs (#9 and #10) also included in Population 1 had, in 1998–1999, been occupied by a different species in 1996–1997 (figure 1). Therefore, of the six IPs included in the control group three changed populations based solely on morphometric species designations in a four year span (1996–1999).

Population 1 also demonstrates the potential for a complex and dynamic social structure. MRR techniques only connected Population 1 to three IPs (#28, #21, and #24) in 1996–1997 (figures 1 and 2). Another IP (#19) was included based on inference because of its proximity between two already MRR-connected IPs (#28 and #24), past experience with recapture probabilities, and morphometric similarities. Two additional IPs (#9 and #10) were included in 1998 due to our past experience with mobility plus morphometric similarities and lack of aggression. The genetic sequence data verified the inclusion of IP #9 within this population because the ML (1.1) was the same as that of IPs #21 and #24 (figure 1). However, the genetic data raised questions concerning termite colony composition because three of the six IP's occupied by *R. hageni* termites and designated Population 1 contained four different MLs (figure 1).

Precedence using MRR and phenotypic similarities is the sole justification for describing the termites collected from these IPs as a single population. The exact, functional relationship(s) between multiple MLs within the context of a colony as defined by MRR techniques, nonaggression, and phenotypic markers has yet to be fully elucidated. These data in conjunction with other recently published information clearly demonstrate that the social organization of *Reticulitermes* societies is dynamic and cannot be defined using a single technique like MRR or genetics (Jenkins *et al.*, 1999; Forschler and Jenkins, 1999). Obviously, more research is needed to verify the functional social dynamics of subterranean termites collected within the same general area.

We used MRR, morphometric, and chemophenotypic similarity to delimit termite Populations 3 and 4 (figures 4 and 5). These case histories provide the classic scenario sought by researchers for measuring termite bait efficacy (Su and Scheffrahn, 1996; Thorne and Forschler, 2000). Both populations consumed fipronil-treated cardboard and then disappeared from our field of view (Table 1). We did find evidence from both of these case histories that termites visited one IP five months posttreatment. This one data point of termite activity was followed by a complete lack activity for the next 12 months. Intuitively, if no activity is recorded for over 12 months following treatment we can assume a cause-and-effect relationship and claim population impacts. Compared to the control group and our past experience, 12 months of inactivity is significant and is generally accepted to be indicative of a population reduction (Grace *et al.*, 1996; Su and Scheffrahn, 1996; Forschler, 1996).

What is often not apparent in the presentation of termite activity data is that activity indices are not always consistent for all IPs (Forschler, 1996; Forschler and Jenkins, 1999). This is because data are usually combined for all IPs assigned to a given "colony" when reported in the literature (Su *et al.*, 1993; Su, 1994; Forschler and Ryder, 1996). For example, two of the three IPs (#7 and #23) occupied by Population 3 showed no activity for three months prior to baiting. Population 4 provided indices of activity from half or less of the IPs during any one month, although the IPs harboring termites changed between sample dates. We included the Population 1 MRR data in figure 2 to highlight the less-than-consistent visitation we record at many of our termite field sites (see Forschler, 1996; Forschler and Jenkins, 1999). When termite activity is reported as combined data, the reader is left with the impression that activity is constant and consistent at all IPs delineated using MRR. This ignores the potential for mobility or simple abandonment of a feeding site and could result in reporting

false positives when recording bait efficacy from only one site. In the United States, for example, termite bait practitioners often consider two consecutive months of inactivity, following bait consumption, as an indication of elimination of a termite "colony." Yet in our control group (Population 1) we recorded two consecutive months without activity from all four IPs during 1997 despite no attempt at control (figure 2). The "disappearance" of termites from a feeding site is not an uncommon phenomenon. We have reported that up to 30% of the termite populations we identify in the field "disappear" (more than 7 months of no activity) every year without any intervention other than routine collecting (Forschler, 1996; Forschler and Ryder, 1996). In practice this "normal" fluctuation in termite activity would have been considered a successful elimination of that "colony."

Two other case histories, Populations 2 and 5, present additional problems in interpretation. These case histories would represent a treatment failure using the field techniques commonly employed for the past decade (MRR and phenotypes) because termite activity was not completely eliminated from all of the IPs visited by the targeted population (Su and Scheffrahn, 1996; Thorne and Forschler, 2000). However, when the genetic sequence data are considered, because the MLs exposed to fipronil-treated bait "disappeared" after treatment, some degree of population impact can be claimed (figures 3 and 6).

Population 2, according to MRR conducted in 1994, consisted of a group of subterranean termites using three IPs (figure 3). Phenotypic similarity led us to assume that we continued to observe a single population from 1994 to 1999. In 1997 this population was represented by a single ML (2.1). In 1998 and 1999 a different ML (2.2) was collected from one of the three IPs, #12 (figure 3). Thus if we assume a single queen or monogyne colony structure and determine the populations by MLs the treatment could be deemed a success. Population impacts can be claimed because the ML (2.1) exposed to the treatment disappeared from our field of view after bait application. This claim centers on the assumption that ML 2.2 was not a functional, resource-sharing partner to ML 2.1—at least when bait was present (March–May of 1998) (figure 3).

The DNA marker data also provide insights into Population 5 that suggest a population impact following treatment (figure 6). Termites collected from these three MRR-connected IPs displayed consistent phenotypes during 1996 to 1999, reinforcing the perception of a single termite "colony." Mitochondrial sequence data, however, indicated that a more complex social organization was present prior to, during, and after bait application. The two MLs (5.1 and 5.2) identified in 1997 "disappeared" prior to bait placement. Further complicating interpretation of the population parameters for this group is that the MLs (5.3 and 5.4) recorded during baiting in 1998 were replaced by another ML (5.5) in 1999 (figure 6). Thus, by defining the targeted populations using ML data, and disregarding the probability that the MLs collected prior to treatment "disappeared" of their own volition, we could declare a successful treatment. The justification for that claim lay in that the maternal genotypes present during treatment were not collected for at least 12 months after treatment.

Case histories 2 and 5 would be considered treatment failures using MRR, hydrocarbon-phenotype, aggression bioassay, and morphometric similarities to delineate the populations, and a success if delineated by MLs. This apparent paradox is the result of two facts: our lack of understanding of subterranean termite social structure and the assumptions made in field research aimed at population management.

The question at the heart of measuring the efficacy of a termite population management control strategy remains, What is a termite colony? A simple definition of a biological colony according to *Webster's New World Dictionary* is "a group of similar animals living together." This broad definition would be acceptable to most biologists, but defining what "living together" means within the context of a group of subterranean termites collected from the field is complicated by their cryptic, mobile lifestyle and diffuse network of interconnected feeding sites. We can propose several possible explanations to describe a functional termite colony based on the data collected during these studies. The first set of explanations is based on the concept that a termite colony is organized around a single queen.

Membership in a monogyne termite colony can be defined solely on the basis of maternal lineage because all offspring of a single queen (the colony members) carry a duplicate copy of their mother's mtDNA (Birkey *et al.*, 1983). Therefore, under the monogyne termite colony scenario, our data indicate the complete inadequacy of MRR and phenotypic techniques for delineation of colonial associations. Intuitively, MRR and phenotypic similarities should identify groups of termites that are—at least—capable of sharing resources and cooperating. The appearance of termites representing two different MLs at the same IP could be indicative of several conditions given a monogyne social structure. One is that the disturbance caused by sampling moves termites to feeding sites not normally occupied by that population. Our previous work with kin recognition has demonstrated that termites of different species can coexist in artificially contrived situations—so why not, for a time, under field conditions (Polizzi and Forschler, 1999)? Given the temporal nature of the data obtained by sampling we could be recording a temporary and anomalous condition when a few marked termites appear at another IP. By assuming that the connections recorded by MRR are continuous, researchers could be overestimating subterranean termite societal relationships. This becomes especially important when using multiple MRR protocols that mark all termites from each IP where marked termites were recovered in subsequent mark–release episodes (Grace *et al.*, 1989; Su *et al.*, 1993; Haagsma and Rust, 1995; Forschler and Townsend, 1996; Forschler and Ryder, 1996). Multiple MRR techniques could therefore grossly overestimate colonial associations.

The second scenario under the monogyne colony concept is that termites from several colonies will occasionally and purposefully share resources but maintain separate colony affiliations. Maternal lines could indicate separate, functional colony units that sometimes coalesce with, and at other times remain distinct from, neighboring colonies. This is similar to the "open and closed societies" proposed by Clement (1986), which resulted from his examination of subterranean termite populations using isoenzyme analysis and agonism bioassay. The climate of the area where he worked displayed distinct wet and dry seasons, and the termite populations he studied showed seasonality corresponding to the "open" (coalescing) and "closed" (separate) phases. Our maternal gene sequence data from Populations 1, 2, and 5 raise the possibility that *Reticulitermes* societies can display "open" and/or "closed" phases yet retain a monogyne functional colony unit (figures 1, 3, and 6). No seasonality is evidenced with our data. We could, however, describe the elimination of ML 5.3 and ML 5.4 as having caught those populations in an "open" phase (figure 6). The failure of our treatment to eliminate both ML 2.1 and ML 2.2 in Population 2 could

be explained by our having placed the termiticidal bait during a "closed" phase while the MRR was conducted, in both instances, during an "open" phase (figure 3).

The site fidelity displayed by the MLs in the control group (figure 1) was unexpected given our past experience (Jenkins *et al.*, 1999). However, it corroborates the site fidelity noted in recent MRR studies with a mound-building Rhinotermitid, *Coptotermes* spp., in Australia (Evans *et al.*, 1998) and the kin-biased foraging, as reported by Kaib *et al.* (1996) in another nest-building Rhinotermitid, *Schedorhinotermes* spp., termite from Africa. We can, therefore, propose a colony organization with multiple primary queens (primary polygyny) to explain why the Rhinotermitid populations, *Reticulitermes* spp. we examined might contain multiple MLs. Under this scenario, the offspring of a single queen, would in general forage, feed, and "hang out" mostly with their brothers and sisters (same maternal line). The fact that we occasionally found marked termites in locations consistently occupied by different MLs (Populations 1, 2, and 5) would thus be explained. In a polygyne subterranean termite colony displaying kin-biased associations, the offspring of several queens would be in contact with one another through a system of underground tunnels connecting several IPs. Termites would move freely between sites but for the most part congregate at IPs with their siblings. Occasionally offspring from another queen within that "colony" would travel to sites not generally occupied by their sibling group. The proportion of those that "cross over" would be small, yet we would, on occasion, detect a marked termite at an IP not normally visited by that ML.

## Conclusions

Interpretation of the temporal data obtained from field studies of subterranean termites is, by necessity, inference and assumption driven in order to accommodate events between sampling dates. The terminology, therefore, used to define a subterranean termite population must be explained within the context of the techniques used in a particular study. Clearly, the use of a single technique, whether MRR, agonism bioassay, morphometric characters, genetic markers, or chemotaxonomic phenotypes, provides valuable information on the potential for social interactions. MRR can describe the movement of termites between feeding sites and indicates the potential for sharing resources (Grace *et al.*, 1989; Su *et al.*, 1993; Haagsma and Rust, 1995; Forschler and Townsend, 1996). Agonism bioassay provides information on kin recognition that could be indicative of the potential for subterranean termite populations to coexist (Clement, 1986; Jones, 1990; Polizzi and Forschler, 1999; Haverty *et al.*, 1999). Morphometric and chemophenotypic data delimit additional character states that can be used to infer potential connections (Haverty *et al.*, 1996; Brown *et al.*, 1996). Genetic markers provide data regarding relatedness but are dependent on the technique and region of the genome examined (Reilly, 1987; Jenkins *et al.*, 1999). The information provided by these techniques is appropriate only for the time and location of the collection and must be interpreted with caution to account for the potential mobility of termite populations. The only way, therefore, to elucidate fully the extent of termite population interactions is to collect data over time using several, if not all, of the aforementioned techniques (Forschler and Jenkins, 1999).

The term "colony" must be clearly defined when testing a population management tool like termiticidal bait. We propose, as a result of our lack of understanding of the functional social structure of subterranean termite colonies, that implementation of termite population management tactics must, at this time, be considered rudimentary (Forschler and Robinson, 1999). The science of urban entomology is in its infancy in providing realistic population management strategies for subterranean termites. Today, termite bait techniques operate under a zero tolerance action threshold that arguably is acceptable given the current technology (Forschler and Robinson, 1999; Thorne and Forschler, 2000). This study demonstrates that a *Reticulitermes* population can be composed of several matrilines that could be completely independent or associated in one form or other and highlights the potential dynamic nature of subterranean termite social interactions. It is critical that practitioners of termite baiting realize that a vacancy after bait application can mean that a true colony was eliminated, or that the lack of activity is temporary, due to movement. Regular inspections for evidence of structural infestation are currently the only method of documenting value from a termite-baiting program. Long-term, regular inspection of termite detection devices provides only limited information—the presence or absence of termites at specific locations around a structure. To implement true population management of subterranean termites, we must have realistic measures of termite population parameters. Those measures should allow assessment of the risk presented by termites detected in the vicinity of a structure and form the basis of the decision-making process prior to implementation of a management scenario. Defining what constitutes a functional termite colony should be the first step toward designing realistic, environmentally friendly subterranean termite population-management tactics.

## Acknowledgments

The authors would like to thank Monica Townsend, Sherry Ridgeway, Mark Yates, Joey Kidd, Laura Hutchings, Ryan Whitehurst, Ashley Foreman, and Fern Lovelace for technical assistance during these studies. We also acknowledge the contributions of Jennifer Blanton and Mark Hopkins for help with the genetic characterizations and Mike Haverty and Lori Nelson for the chemotaxonomic characterizations. Finally, we would like to thank Aventis Environmental Sciences and SINERR for the partial funding they provided.

## References

Beal, R.H. and Esenther, G.R. (1980) A new approach to subterranean termite control—the bait block method. *Sociobiology* **5**, 171–174.

Beal, R.H., Mauldin, J.K. and Jones, S.C. (1989) Subterranean termites—their prevention and control in buildings. *Home and Garden Bulletin 64*. USDA, Forest Service, Washington, DC.

Beard, R.L. (1974) Termite biology and bait block method of control. *Connecticut Agri. Exper. Stat. Bull. 784*.

Birkey, C.W., Maruyama, T. and Fuerst, P. (1983) An approach to population and evolutionary genetic theory for genes in mitochondria and chloroplasts and some results. *Genetics* **103**, 513–527.

Brown, A.A., Herms, W.B., Homer, A.C., Kelly, J.W., Kofoid, C.A., Light, S.F. and Randall, M. (1934) General recommendations for the control of termite damage. In *Termites and Termite Control* (C.A. Kofoid, ed.), pp. 579–591. University of California Press, Berkeley, CA.

Brown, W.V., Watson, J.A.L. and Lacey, M.J. (1996) A chemotaxonomic survey using cuticular hydrocarbons of some species of the Australian harvester termite genus Drepanotermes (Isoptera: Termitidae). *Sociobiology* **27**, 199–221.

Clement, J.-L. (1986) Open and closed societies in *Reticulitermes* termites (Isoptera: Rhinotermitidae): geographic and seasonal variations. *Sociobiology* **11**, 311–323.

Cole, L.M., Nicholson, R.A. and Casida, J.E. (1993) Action of phenyl pyrazole insecticides at the GABA-gated chloride channel. *Pestic. Biochem. Physiol.* **46**, 47–54.

Edwards, R. and Mill, A.E. (1986) *Termites in Buildings: Their Biology and Control*. The Rentokil Library, London, UK.

Evans, T.A., Lenz, M. and Gleeson, P.V. (1998) Testing assumptions of mark–recapture protocols for estimating population size using Australian mound-building, subterranean termites. *Ecol. Entomol.* **23**, 139–159.

Forschler, B.T. (1994) Fluorescent spray paint as a topical marker on subterranean termites (Isoptera: Rhinotermitidae). *Sociobiology* **24**, 27–38.

Forschler, B.T. (1996) Baiting *Reticulitermes* (Isoptera: Rhinotermitidae) field colonies with abamectin and zinc borate-treated cellulose in Georgia. *Sociobiology* **28**, 459–485.

Forschler, B.T. (1998a) Subterranean termite biology in relation to prevention and removal of structural infestation. In *NPCA Research Report on Subterranean Termites*, pp. 31–51. NPCA, Dunn Loring, VA.

Forschler, B.T. (1998b) Termite baiting: a research perspective. *Pest Control Technology* **26**(6), 90, 92, 94, 96, 98, 99.

Forschler, B.T. (2000) Baits and termite baiting technology: theory and practice. In *Proceedings of the Conference on Residential Construction Materials and Practices to Protect Homes from Formosan Subterranean Termites* (R. Smith, ed.). Louisiana Forest Products Laboratory Publications, Baton Rouge, LA, in press.

Forschler, B.T. and Jenkins, T.M. (1999) Evaluation of subterranean termite biology using genetic, chemotaxonomic, and morphometric markers and ecological data: a testimonial for multidisciplinary efforts. *Trends in Entomology* **2**, 72–80.

Forschler, B.T. and Lewis, V.R. (1997) Why termites can dodge your treatment. *Pest Control* **65**(2), 42–46, 53.

Forschler, B.T. and Robinson, W.H. (1999) Ants and subterranean termites in the urban environment: the terminology of population management control tactics. *Proc. 3rd International Conf. on Urban Pests* (W.H. Robinson, F. Rettich and G.W. Rambo, eds.), pp. 589–595. Executive Committee of the International Conference on Urban Pests. Graficke zavocy Hronov, Prague, Czech Republic.

Forschler, B.T. and Ryder, J.C., Jr. (1996) Subterranean termite, *Reticulitermes* spp. (Isoptera: Rhinotermitidae) colony response to baiting with hexaflumuron using a prototype commercial termite baiting system. *J. Entomol. Sci.* **31**, 143–151.

Forschler, B.T. and Townsend, M.L. (1996) Mark-release-recapture estimates of *Reticulitermes* spp. (Isoptera: Rhinotermitidae) colony foraging populations from Georgia, U.S.A. *Environ. Entomol.* **25**, 952–962.

Forschler, B.T., Harron, J. and Jenkins, T.M. (2000) Case histories involving the Formosan subterranean termite in Atlanta, Georgia, USA. *Sociobiology* **37**, 311–322.

Gay, F.J. (1969) Species introduced by man. In *Biology of Termites. Vol. I* (K. Krishna and F.M. Weesner, eds.), pp. 459–494. Academic Press, New York, NY.

Gold, R.E., Collins, A.A., Pawson, B.M. and Howell, H.N., Jr. (1994) Termiticide technology—the isofenphos dilemma. *Technology: J. Franklin Institute* **331**, 189–198.

Gold, R.E., Howell, H.N., Jr., Pawson, B.M., Wright M.S. and Lutz, J.C. (1996) Persistence and bioavailability of termiticides to subterranean termites (Isoptera: Rhinotermitidae) from five soil types and locations in Texas. *Sociobiology* **28**, 337–364.

Grace, J.K., Abdallay, A. and Farr, K.R. (1989) Eastern subterranean termite (Isoptera: Rhinotermitidae) foraging territories and populations in Toronto. *Can. Entomol.* **121**, 551–556.

Grace, J.K., Tome, C.H.M., Shelton, T.G., Oshiro, R.J. and Yates, J.R., Jr. (1996) Baiting studies and considerations with *Coptotermes formosanus* (Isoptera: Rhinotermitidae) in Hawaii. *Sociobiology* **28**, 511–520.

Haagsma, K.A. and Rust, M.K. (1995) Colony size estimates, foraging trends, and physiological characteristics of the western subterranean termite (Isoptera: Rhinotermitidae). *Environ. Entomol.* **24**, 1520–1528.

Haverty, M.I., Nutting, W.L. and La Fage, J.P. (1975) Density of colonies and spatial distribution of foraging territories of the desert subterranean termite *Heterotermes aureus* (Snyder). *Environ. Entomol.* **4**, 105–109.

Haverty, M.I., Forschler, B.T. and Nelson, L.J. (1996) An assessment of the taxonomy of *Reticulitermes* (Isoptera: Rhinotermitidae) from the southeastern United States based on cuticular hydrocarbons. *Sociobiology* **28**, 287–318.

Haverty, M.I., Getty, G.M., Copren, K.A. and Lewis, V.R. (1999) Seasonal foraging and feeding behavior of *Reticulitermes* spp. (Isoptera: Rhinotermitidae) in a wildland and a residential location in northern California. *Environ. Entomol.* **28**, 1077–1084.

Howard, R.W., Jones, S.C., Mauldin, J.K. and Beal, R. (1982) Abundance, distribution, and colony size estimates of *Reticulitermes* spp. (Isoptera: Rhinotermitidae) in southern Mississippi. *Environ. Entomol.* **11**, 1290–1293.

Jenkins, T.M., Basten, C.J., Dean, R., Mitchell, S.E., Krescovich, S. and Forschler, B.T. (1998) Matriarchal genetic structure of *Reticulitermes* (Isoptera: Rhinotermitidae) populations. *Sociobiology* **33**, 239–263.

Jenkins, T.M., Basten, C.J., Krescovich, S. and Forschler, B.T. (1999) Mitochondrial gene sequence questions Reticulitermes sp. social structure (Isoptera: Rhinotermitidae). *Sociobiology* **34**, 161–172.

Jenkins, T.M., Verkerk, R., Dean, R. and Forschler, B.T. (2001) Phylogenetic analyses of two mitochondrial and one nuclear intron region illuminate European subterranean termite (Isoptera: Rhinotermitidae) taxonomy and gene flow. *Mol. Phylgenet. Evol.* **20**, 286–293.

Jones, S.C. (1990) Delineation of *Heterotermes aureus* (Isoptera: Rhinotermitidae) foraging territories in a Sonoran desert grassland. *Environ. Entomol.* **19**, 1047–1054.

Kaib, M., Husseneder, C., Epplen, C., Epplen, J.T. and Brandl, R. (1996) Kin-biased foraging in a termite. *Proc. R. Soc. Lond.* **263**, 1527–1532.

Kofoid, C.A. (1934) Biological backgrounds of the termite problem. In *Termites and Termite Control* (C.A. Kofoid, ed.), pp. 1–13. University of California Press, Berkeley, CA.

Lee, K.E. and Wood, T.G. (1971) *Termites and Soils*. Academic Press, New York.

Liu, H. and Beckenbach, A.T. (1992) Evolution of the mitochondrial cytochrome oxidase II gene among 10 orders of insects. *Mol. Phylgenet. Evol.* **41**, 41–52.

Moore, H.B. (1990) Termites: their prevention and control. In *Wood Destroying Insects*, pp. 34–71. Pest Control Magazine. Cleveland, OH.

Polizzi, J.M. and Forschler, B.T. (1998) Intra- and interspecific agonism in *Reticulitermes falvipes* (Kollar) and *R. virginicus* (Banks) and effects of arena and group size in laboratory bioassay. *Insectes Soc.* **45**, 43–49.

Polizzi, J.M. and Forschler, B.T. (1999) Factors that affect aggression among the worker caste of Reticulitermes spp. subterranean termites. (Isoptera: Rhinotermitidae). *Journal of Insect Behavior* **12**, 133–146.

Potter, M.F. (1997) Termites. In *Mallis Handbook of Pest Control. 8th ed.*, pp. 233–333. Mallis Handbook & Technical Training Co., Cleveland, OH.

Reilly, L.M. (1987) Measurements of inbreeding and average relatedness in a termite population. *Am. Nat.* **130**, 339–349.

Scheffrahn, R.H. and Su, N.-Y. (1994) Keys to soldier and winged adult termites (Isoptera) of Florida. *Florida Entomologist* **77**, 460–474.

Simon, C., Frati, F., Beckenbach, A., Crespi, B., Liu, H. and Flook, P. (1994) Evolution, weighting, and phylogenetic utility of mitochondrial gene sequences and a compilation of conserved polymerase chain reaction primers. *Annals Entomol. Soc. Am.* **87**, 651–701.

Su, N.-Y. (1994) Field evaluation of a hexaflumuron bait for population suppression of subterranean termites (Isoptera: Rhinotermitidae). *J. Econ. Entomol.* **87**, 389–397.

Su, N.-Y. and Scheffrahn, R.H. (1988) Intra- and interspecific competition of the Formosan and eastern subterranean termites: evidence from field observations (Isoptera: Rhinotermitidae). *Sociobiology* **14**, 157–164.

Su, N.-Y. and Scheffrahn, R.H. (1990) Economically important termites in the United States and their control. *Sociobiology* **17**, 77–94.

Su, N.-Y. and Scheffrahn, R.H. (1996) A review of the evaluation criteria for bait-toxicant efficacy against field colonies of subterranean termites (Isoptera). *Sociobiology* **28**, 521–530.

Su, N.-Y. and Tamashiro, M. (1987) An overview of the Formosan subterranean termite, *Coptotermes formosanus* (Isoptera: Rhinotermitidae) in the world. In *Proceedings of the International Symposium on the Formosan Subterranean Termite* (M. Tamashiro and N.-Y. Su, eds.), pp. 3–15. College of Tropical Agriculture and Human Resources, University of Hawaii, Research Extension Series 083, Honolulu, Hawaii.

Su, N.-Y., Ban, P.M. and Scheffrahn, R.H. (1993) Foraging populations and territories of the eastern subterranean termite (Isoptera: Rhinotermitidae) in southeastern Florida. *Environ. Entomol.* **22**, 1113–1117.

Su, N.-Y., Thoms, E.M., Ban, P.M. and Scheffrahn, R.H. (1995) Monitoring/baiting stations to detect and eliminate foraging populations of subterranean termites (Isoptera: Rhinotermitidae) near structures. *J. Econ. Entomol.* **88**, 932–936.

Thorne, B.L. (1998) Biology of subterranean termites of the genus *Reticulitermes*. In *NPCA Research Report on Subterranean Termites*, pp. 1–30. NPCA, Dunn Loring, VA.

Thorne, B.L. and Forschler, B.T. (2000) Criteria for measuring the efficacy of stand alone termite baits. *Sociobiology* **36**, 245–255.

Thorne, B.L., Russek-Cohen, E., Forschler, B.T., Breisch, N.L. and Traniello, J.F.A. (1996) Evaluation of mark-release-recapture methods for estimating forager population size of subterranean termite (Isoptera: Rhinotermitidae) colonies. *Environ. Entomol.* **25**, 938–951.

Waller, D.A. and La Fage, J.P. (1987) Nutritional ecology of termites. In *Nutritional Ecology of Insects, Mites and Spiders* (F. Slansky and J.G. Rodriguez, eds.), pp. 487–532. John Wiley, New York, NY.