# EFFECTS OF DISODIUM OCTABORATE TETRAHYDRATE IN ETHYLENE GLYCOL ON CONSUMPTION AND MORTALITY OF THE EASTERN SUBTERRANEAN TERMITE

By

COLIN DOLAN HICKEY

A THESIS PRESENTED TO THE GRADUATE SCHOOL
OF THE UNIVERSITY OF FLORIDA IN PARTIAL FULFILLMENT
OF THE REQUIREMENTS FOR THE DEGREE OF
MASTER OF SCIENCE

UNIVERSITY OF FLORIDA

2006

This thesis is dedicated to my parents, Charles and Janice Hickey.

ACKNOWLEDGMENTS

I would like to thank my friends of the Urban lab, especially Dave “Face” Melius, Justin “Dursban” Sanders and Ryan “Tarzan” Welch, for the good times and for the support and motivation of Pili Paz, who put me back on track to finish writing my thesis. I thank my family for their patience and advice through the ups and downs of grad school and life in Florida.

Special recognition goes to Gil S. Marshall and Tiny Willis. If not for their help with supplies, advice and friendship, I would still be searching for the pipette and trying to figure out how to get research supplies. I greatly appreciate the assistance of Debbie Hall, her assistant Josh Crews, and Nancy Sanders for helping me negotiate the labyrinth of administrative details necessary for completing my degree.

I thank Dr. Faith Oi, whose workspace I used in my usual messy way (of course in the name of good science) and whose helpful scientific guidance and practical advice and recommendations were sincerely appreciated. I thank Cindy Tucker for her generous assistance with termite colonies for my research and reading and discussing termite research with me, especially mine.

My deepest thanks go to Dr. Philip Koehler for giving me this unique opportunity and putting up with my unorthodox methods. His help and guidance were essential for me to complete my degree. I also thank the rest of my graduate committee, Drs. Simon Yu and Brian Cabrera.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

Figure page

Abstract of Thesis Presented to the Graduate School
of the University of Florida in Partial Fulfillment of the
Requirements for the Degree of Master of Science

EFFECTS OF DISODIUM OCTABORATE TETRAHYDRATE IN ETHYLENE GLYCOL ON CONSUMPTION AND MORTALITY OF THE EASTERN SUBTERRANEAN TERMITE

By

Colin Dolan Hickey

May 2006

Chair: Philip Koehler
Major Department: Entomology and Nematology

The economic impact of termites on a yearly basis is staggering. From pre- and post-construction treatments, re-treatments, and repair costs, termite control climbs into billions annually. Termites and humans have developed a conflict of interest between finished wood products for construction and aesthetics. Recent interest in boron as a potential wood preservative has been spurned by the search for environmentally friendly and cost-effective replacements to existing wood preservation strategies. Disodium octaborate tetrahydrate (DOT), a borate salt, is a broad spectrum toxicant that acts against fungi and insects with a low mammalian toxicity and has been proven particularly effective against termites. Borates diffuse through wood because they dissolve in water. The loading capacity of DOT is increased when ethylene glycol is used as a solvent. Rate of mortality and deterrence of feeding in *Reticulitermes flavipes* were evaluated with treatment of filter paper using DOT in ethylene glycol.

A lethal time bioassay was conducted to determine how quickly contact with DOT/glycol killed termites. DOT killed termites rapidly. At DOT/glycol concentrations ≥7,774 ppm, termite mortality was >85% within 192 h. Although ethylene glycol, a contact desiccant, accelerated mortality because it contacted the termites in the DOT/glycol treatment, aqueous treatments of ≥ 7,774 ppm DOT caused >85% mortality within 192 h. DOT/glycol treatments exhibited relatively high $LT_{50}$s. The $LT_{50}$ of 303,209 ppm DOT/glycol was 49.69 h.

*R. flavipes* consumption of filter paper treated with different concentrations of DOT was conducted to determine deterrence of feeding. Termites began feeding on the filter papers placed in each container in 24 h. Termite consumption of treated filter papers decreased as concentrations of DOT increased. At 783 ppm, DOT reduced cellulose ingestion by ~10%. At 303,209 ppm DOT, ingestion was reduced by ~84%. Despite reduction in consumption of filter paper, DOT consumption increased with higher concentration of treatment. Filter paper treated with DOT did not deter feeding.

When mortality of termites was observed at 192 h, greater mortality had occurred in treatments at the highest concentration of DOT (81.3% for 303,209 ppm DOT). Termites were ingesting greater quantities of DOT with higher concentration of treatment. High mortality was caused by ingestion of lethal doses of DOT. My study determined that DOT kills termites rapidly by ingestion, consequently limiting damage to wood in the structure. DOT/glycol treatments were not found to be deterrents of feeding except at the highest concentrations. As a result, untreated wood in the structure can be protected because treated wood would be a more convenient food source and the treatment would probably not cause feeding deterrence.

CHAPTER 1
LITERATURE REVIEW

## Termite Biology

Termites (Isopterans) are medium sized, social insects that consume cellulose as their main source of nourishment. Isoptera have similar sized fore and hind wings and antennae are monoliform. Specific morphological characteristics that differentiate termites into different families are divided between the soldier and alate forms of each species (Snyder 1948). Wing characters, presence of a fontanelle and ocelli, pronotum shape and forewing scale size in relation to pronotum are physical keys to determine taxonomy in alates (Scheffrahn and Su 1994). For the soldier caste, head size and mandibles are the keys to determine taxonomy of termites.

Subterranean termites are distributed virtually across the entire United States. The endemic *Reticulitermes spp.* are most prolific, but genera *Coptotermes, Heterotermes* and *Prorhinotermes* are also present (Light 1934). In urban areas, human structures provide termites with a host of advantageous conditions. Because Americans have favored construction with wood, termites have become a constant threat to structures (Forschler 1999). Structures also provide harborage and moisture, allowing termites' access to the most important conditions for their survival.

Isoptera is believed to have evolved during the Permian period (200 mya) from a line that branched from Blattaria (Krishna 1969). Similarities between the primitive termite species in Australia, *Mastotermes darwiniensis* Froggart, and primitive Blattaria are evidence of this common ancestry (Thorne 1997). However, Isopterans have evolved

a dynamic social structure unlike other insects. Unlike Hymenoptera, the social structure of Isoptera does not function on the basis of a haplo-diploid reproduction (Wilson 1971). All termites are diploid. There are many factors that have influenced the evolution of termites and predisposed them to eusocial organization. Dense familial habitats with a common food source, the slow development and overlap of generations, a high mortality risk for individuals outside of the familial habitat and associated advantages of a mutual and community defense, and the obligate dependence on recycled flagellated protozoans for the digestion of material containing cellulose, are plausible conducive elements that contributed to the social behavior that has evolved in Isoptera (Bartz 1979; Thorne 1997).

Subterranean termites are aptly named by their cryptobiotic behavior associated with soil. Rhinotermitidae tunnel through soil with an objective of locating food sources. Instances in which wood and soil are in contact are conducive to subterranean termite infestation (Potter 2004). Where non-organic materials impede termites from reaching wood, termites frequently build shelter tubes over the material to reach on the wood. Once a food source is located, the tunnel is reinforced with anal cement (Stuart 1967). Various factors, including the species of termite and the size and the quality of the food source, influence the intensity of termite feeding after a food source has been discovered.

Communication in termites is a successful adaptation that has enabled termites to maintain and defend efficient, well-organized colonies. Because termite soldiers and workers are blind, pheromones are the most important method of communication in termites (Clement and Bagneres 1998). Termites use pheromones to mark trails for more efficient foraging. Although (Z,Z,E) 3,6,8 dodecatrien-1-ol has been found as a principal pheromone marker in termites, extracts from *C. formosanus* trails provide evidence for

other trail following compounds (Matsumura et al. 1968; Tokoro et al. 1994). Pheromones are used to differentiate caste members during development. Species-specific behaviors are determined by pheromones and likewise pheromones elicit other specific responses, including recruitment to food sources and for defense, as well as functioning as a sex pheromone for alates. However, experiments by Cornelius and Bland (2001) failed to detect any species-specific pheromone trail following behavior. Colony specific cuticular hydrocarbons prevent intruders from other colonies of the same species from infiltrating the nest, although agonism studies have determined that colony behavior to inter-colony influences remain open and closed at different times depending on season and weather.

Termite workers groom other members of the colony to remove potentially parasitic fungi and bacteria with their mouthparts (Thorne 1996). Termites participate in stomadeal and proctadeal trophallaxis, the sharing of regurgitated and partially digested food for nutrient and symbiotic exchange (McMahan 1969). Soldiers cannot feed in the manner of worker termites because of their elongate mandibles and therefore soldiers receive nutrients from their nestmate workers via trophallaxis. Furthermore, because the obligate symbionts in the midgut are shed with the midgut lining after each larval instar, the flagellated protozoans that are lost need to be replaced for termites to feed. Thus, trophallaxis among nest-mates maintains that developing termites receive the symbionts that enable them to be productive colony members (McMahan, 1969).

The termite life cycle is hemimetabolous and the development follows three distinct pathways: reproductive, soldier and worker. The queen or secondary reproductive lays eggs in the nest. These eggs hatch into larvae that in turn can develop into the three

castes. Immature larvae follow two pathways of development. Soldiers and workers branch from the developing imaginal reproductive track due to conditions present before their first molt, not from a fate determined at birth or from the egg (Krishna 1969). Termite larvae following the reproductive pathway become nymphs. The nymphal stage is a precursor to the alate reproductive or the brachypterous reproductive. Alate or brachypterous reproductives mate and the female renews the cycle by laying eggs. Termites show an amazing amount of developmental plasticity; nymphs can regress from becoming reproductives to workers and under different conditions, such as the loss of the nested queen or male, workers can re-develop into functional reproductives (Lee and Wood 1971).

The reproductive caste can be further differentiated into primary and secondary reproductives. Primary reproductives, alates or swarmers, have functional wings and are important in the dispersal and the foundation of new colonies. Mature colonies of subterranean termites produce massive numbers of alates of which their timely dispersal leads to numerous potential infestations. Secondary reproductives or neotenics develop as a result of changing conditions in the colony (Lee and Wood 1971). When a colony becomes well established and sufficiently dispersed, or something happens to the queen (death or infertility), neotenics develop. In some instances, the secondary reproductives, due to their large numbers in the termite colonies rather than high fecundity, replace the queen as the main source of eggs. There are two forms of secondary reproductives that occur in subterranean colonies. Brachypterous neotenics develop from nymphs and retain wing buds (not functional). Apterous neotenics derive from workers and have no wings

or wing pads and have the smallest potential fecundity. In either case, secondary reproductives mate without the possibility of a swarming flight (Thorne et al. 1999).

A dark, enlarged, scleroticized head and the presence of large, obtuse mandibles distinguish the soldier caste. The sclerotized head capsule protects the soldiers from frontal attacks but their soft, white body is defenseless from the rear. Soldiers comprise only 1-2% of the individuals of a *R. flavipes* termite colony (Howard and Haverty 1980). Their purpose in the colony has been traditionally been thought of as defensive, using the large mandibles to slice and cut invaders. They also have some function in colony scouting and foraging, but depend on workers for nourishment because their specialized mouthparts prevent normal feeding (Weesner 1965). These members of the colony do not participate in reproduction.

Workers are the driving force of each colony. They are the most numerous and damaging form and the only caste that actually feeds on wood (Thorne et al. 1999). A "true worker" is a non-soldier, non-reproductive individual that differentiated early and usually irreversibly from the imaginal line. They are blind, have soft white bodies, and control the tasks and chores of a successful colony (Thorne 1996). Workers tend the king and queen, care for the brood, and feed soldiers through trophallaxis. In defense, workers sacrifice their bodies to block incoming predators from invading the nest (Snyder 1948). With chewing mouthparts, workers also use their feeding mandibles for a proactive defense. In other termite species, fantastic mechanisms have been discovered for the role of workers in defense of the colony (Thorne 1982, Thorne et al. 1999).

With the ability to digest cellulose as a food source, termites have become a pest to humans because of the widespread use of wood as a building material. Any cellulose

material that comes into contact with the ground is an economic liability. Wood is at risk for infestation even when elevated because of the termite ability to make shelter tubes and alate swarmers' ability to infest aerially. Subterranean termite foraging is conducted primarily with the construction of tunnels and underground galleries (Hedlund and Henderson 1999).

Termites excavate tunnels for foraging in a generally even manner until either a food source is located, or a termite tunnel reaches a guideline (Potter 2004). A guideline is a natural or artificial edge or pathway that allows termites to easily navigate through the soil with the least possible energy wasted on tunnel excavation. Root systems from plants, pipes or a crack in concrete provide termites with access to simple unobstructed pathways. Forms of termite treatment methods, particularly baiting, can undermine this termite behavior.

## Control Methods

The economic impact of termites on a yearly basis is staggering. From pre- and post-construction treatments, re-treatments, and repair costs, termite control climbs into billions annually (Thorne et al. 1999). Termites and humans have developed a conflict of interest between finished wood products for construction and aesthetics. Control measures probably began in ancient times with the Latin "termes," and control still remains a difficult task with this pest today.

Termite barriers and shields are designed to block termites from underground access to cracks and voids that are mistakenly left in the construction, by using a full structure treatment of physical barriers that prevent termites from passing through to the structure. There are several different technologies that have been developed (Potter 2004). Some metal shields do not prevent infestation, but rather force termites to tube

around the shield and become openly visible. Thus, the tubes can be mechanically removed to prevent termites from reaching parts of the structure that have not been shielded.

Biological control of subterranean termites has been promising in the laboratory but has suffered shortcomings in field trials. Termite predators are abundant in nature, termites are easy prey for many organisms including man, however only a few species of ants specialize in predation of termites. Nematodes and fungi have been studied in their effectiveness against termites in the field. Nematode efficacy is precluded by a lack of parasitization and the termites overwhelming avoidance (Epsky and Capinera 1988). Fungi treated in field studies caused significant mortality but became less effective over time. Fungi offer the best classical control method, but the limitations of rearing Fungi in a cost-effective manner and their erratic performance in field studies, limits the plausibility of extensive fungi use (Delate et al. 1995).

Liquid termiticide use can be divided into new construction preventative treatments, post-construction preventative treatments and control treatments of infestations. New construction termiticides are applied to the soil underneath the area of the future slab foundation. Post-construction treatments are applied using a drill injection of the termiticide under foundation or drenching a trench dug around the foundation.

Termiticides can be divided between repellents and non-repellents. Historically, repellent termiticides have been used with a design of making a liquid barrier to prevent structural attack. In the wake of organophosphate (OP) phase out, several termiticides have been marketed to replace the use of OPs for effective control of termites. Pyrethroids have practically replaced OPs for repellent barrier treatments because they

function in a similar way to OP and repel termites away from the treatment zone (Potter 2004).

A novel understanding of termite foraging patterns and social behavior has aided in the development of non-repellent termiticides. Non-repellent termiticides are invisible and undetectable to foraging termites. Termites unknowingly come into contact with the chemical and then spread the lethal chemical via grooming and voluntary trophallaxis to other members of the colony. Non-repellent termiticides have proven remarkably effective and have become favored for prevention and control of subterranean termites.

Termite baiting strategies have been developed in recent years, but there has been a historical precedent set by termite baiting (Potter 2004). Baiting stations are set in the soil flush with ground level and 'baited' with wood to monitor termite activity. As a prerequisite for effectiveness, sanitary methods must be undertaken to prevent termites from alternate food sources and guidelines removed that will allow termites from evading bait detection stations. Another key element in baiting effectiveness is placement. Placement of bait stations should coincide with infestation or likely entry points of structures. Once termite activity has been established, the station is set with active ingredient, and termites are returned into the bait station for self-recruitment. The logic behind bait stations is that termites will consume active ingredient and then pass the chemical directly and indirectly to other nest-mates by trophallaxis. Baiting has become a popular alternative to liquid treatments because of the minimal pesticide residual and its external application.

## Wood Treatment and Preservation

Wood has been favored as a construction material in the United States and around the world. Hagen (1876) warned of the deleterious effects that wood-destroying

organisms can inflict upon buildings and homes. Insomuch as wood is a food source for a number of different organisms, particularly termites; preservation of wood has been a subject of concern for many years. Initially, developers of wood preservatives were concerned with wood that was in direct contact with the ground (McNamara 1990). In the 19th century, log homes, railroad cross ties and wood beams to support mine shafts, were the primary target of wood preservation strategies.

There are a number of wood species that confer various physical and chemical properties that protect them from termite and other wood-destroying organisms. The density of wood is a factor that predisposes wood to termite attack. Hardwoods are known to be more termite resistant than softwoods because of a greater density. Specific chemicals found to be produced by wood resistant to termite attack have been isolated and identified. These chemicals, such as chlorophorin from *Chlorophora excelsa* and pinosylvin from *Pinus sylvestris* have been observed to be repellent to termite attack. Hickin (1971) reviewed and listed wood species known to be resistant to termites. It has also been observed, however, that natural repellents are not indefinitely reliable as termites exposed to these chemicals for long periods of time become conditioned and the chemical loses it repellency.

Chemicals were used to form a protective shield around the wood, which coated the wood with a toxic chemical to provide a barrier from wood destroying organisms. Creosote, an amalgamation of two oils from coal tar, was developed by Moll in 1836 (Murphy 1990). The use of creosote in wood preservation was researched and implemented until around the turn of the century. At this time, Wolman and Malenkovic developed water soluble preservatives that used fluorides, dinitrophenol, chromates and

arsenic, known as “Wolman salts”. The awareness of potential leaching of water based preservatives became apparent and copper chromate arsenate (CCA), which could be fixed into wood, was developed in 1933. CCA has become the preservative of choice for wood protection (Webb 1999). Because of the tedious process of the application of wood preservatives to lumber, the use of liquid termiticides took favor in the protection of homes and buildings from termite attack (Potter 2004).

In the United States, Randall and Doody (1934) noted the effective chemical properties of using boron as a potential pesticide, but was largely ignored in the United States as a potential wood preservative because of its potential to leach from the treated wood (Williams 1990). The application of boron-based chemicals as wood preservatives did, however, find practical application in Australia and New Zealand in the 1930s and 40s. Boric acid was applied to lumber using a technique that involved immersion of the wood in 1.24% boric acid at 200°F (93°C) (Cummins 1939). In the late 1940s, legislation in Australia was enacted to guarantee that all structural timber on homes be chemically treated (Greaves 1990). Boron-based preservatives found some interest in Europe and Canada during the 1960s but were competing against the application of the successful CCA preservative already widely used.

Recent interest in boron as a potential wood preservative has been spurned by the search for environmentally friendly and cost-effective replacements to existing wood preservation strategies. Boron exists in nature bound to oxygen, called borates, and have been noted to be toxic to wood-destroying organisms and diffusible through wood with moisture (Williams and Amburgey 1987; Williams and Mitchoff 1990; Becker 1976). Borates are especially diffusible in wood containing >15% moisture content (Schoeman

1998). This has led borates to be considered a promising strategy for the protection of borate-treated wood from wood-destroying organisms.

A borate salt, disodium octaborate tetrahydrate (DOT), has been marketed as a wood preservative and is found in many existing products labeled for wood protection. The mode of action of boron-based insecticides remains unresolved. Ebeling (1995) suggested that boric acid destroys the digestive tract cell wall of cockroaches. Cochran (1995) confirmed the destruction of the cockroach foregut epithelium, suggesting that ingested boric acid leads to starvation. Williams and Mitchoff (1990) and Lloyd et al. (1999) suggest that DOT interferes with chemicals of metabolic importance, such as the NAD+ and NADP+ coenzymes, because of their chemical reaction with the borate anion. Bennett et al. (1988) was in agreement with Williams and Mitchoff (1990) and Lloyd et al. (1999), determining that that the slow mortality of cockroaches from boric acid occurred because of the interference with energy conversion inside the insect's cells. Borates have been asserted to be inhibitors of hindgut protozoan symbiont activity associated with termite digestion of cellulose. Starvation seems unlikely because the rate of mortality that occurs when termites are exposed to large concentrations of borates. Therefore, mortality occurs more quickly than can reasonably explained by starvation (Grace 1991; Su and Scheffrahn 1991b).

## Disodium Octaborate Tetrahydrate in Ethylene Glycol

Disodium octaborate tetrahydrate has several advantages as a wood preservative. It is a broad spectrum toxicant that acts against fungi and insects with a low mammalian toxicity (Krieger et al. 1996). DOT has been proven particularly effective against termites (Grace 1997). Applications of DOT are colorless and odorless (non-volatile) and because of the natural occurrence of boron in nature, are accepted as being more environmentally

friendly than other wood preservatives. Borates diffuse through wood because they dissolve in water. This allows the borates to be carried by wood moisture from the wood's surface into the interior of the wood (Barnes et al. 1989). The advantages of diffusibility into wood have also been historically viewed as disadvantages and borates have been limited to treatment on sheltered, interior wood. Williams and Mitchoff (1990) demonstrated the susceptibility of boron leaching when exposed to weathering, but also demonstrated the effectiveness of the residual, protecting the treated wood from termite consumption. Through observations of termite survival, the lethal effects of DOT were demonstrated, even at drastically reduced concentrations.

The loading capacity of DOT is increased when ethylene glycol is used as a solvent. The toxicity of ethylene glycol is hard to predict due to its chemical nature. It is an odorless, colorless liquid that is greatly hygroscopic, absorbing twice its weight in water in 100% humidity (Budavari 1996). When applied directly to wood block, ethylene glycol caused significant mortality on termites (Grace and Yamamoto 1992). However, ethylene glycol applied to sawdust particles fed directly to termites caused elevated but not significant differences from untreated controls which led Tokoro and Su (1993) to conjecture that ethylene glycol appeared to synergize DOT toxicity on termites.

Based on $LD_{50}$ values, disodium octaborate tetrahydrate in ethylene glycol (DOT/glycol) appears to be 1.5 times more toxic than aqueous DOT on both *R. flavipes* and *Coptotermes formosanus* (Tokoro and Su 1993). Grace and Yamamoto (1994) observed that ethylene glycol did not aid in diffusion (into Douglas-fir wood) but one application of DOT (20%) in glycol was found to obtain more than twice the amount of DOT than two applications of DOT (10%) in water on the surface of the treatment.

Instead of aiding in diffusion into the wood, ethylene glycol was believed to limit DOT from running-off the surface of the wood because of its greater viscosity as compared with water.

The most important limitations of *in situ* applications of structural lumber with DOT/glycol and aqueous DOT are the accessibility of the wood for treatment and the penetration of DOT into the wood. Structural wood that is in place has many inaccessible surfaces. Grace and Yamamoto (1994) noted significant wood weight loss to surfaces that were not exposed to treatment. Su and Scheffrahn (1991a) determined DOT/glycol to diffuse into a wood at a relatively slow pace. After eight months at 13 ± 2% relative humidity, only 40% of the treated wood contained greater than 2,500 ppm DOT. Concentrations of less than 2,500 ppm could be expected in the wood, although according to $LD_{50}$ statistics of DOT, those concentrations present would provide a lethal dose at 95.5 μg/g AI (DOT), well below the colorimetric test (Tokoro and Su 1993).

## Statement of Purpose

The first objective of my research focused on determining the deterrence of termite feeding on cellulose treated with decreasing concentrations of DOT in ethylene glycol and in water. Prior research of DOT's effects on termites concentrated on observations of termite mortality after an extended period of time. Although borates have been purported to deter feeding, termites were experiencing high mortality within a short period of time, leading to the possibility that termites are not deterred from feeding but are prevented by mortal effects. Mortality, as a result of termite's ingestion of DOT and as a function of time after direct contact with DOT, becomes critical in deciphering whether DOT can be considered a deterrence of termite feeding. Thus, a second objective was to determine mortality as a function of borate consumption. Termites began to die more quickly than

was expected, therefore, as a third objective of my research, I conducted tests to observe termite mortality over many time intervals to determine how rapid termite mortality occurs as a result of termites being in direct contact with DOT. Comparing the rate of mortality with the amount of consumption of treatment will show the degree of feeding deterrence of DOT-treatments to the eastern subterranean termite.

# CHAPTER 2
# MATERIALS AND METHODS

## Insects

*R. flavipes* were harvested from widely separated collection sites on the University of Florida campus. Collection sites consisted of buckets (Venture Packaging, Inc. Monroeville, OH. 811192-2) inserted about 15 cm into the soil with the lid flush with the ground. Six holes measuring 5 cm diameter were drilled into the sides and bottom of the bucket for termite access and water drainage. Two rolls of moist corrugated cardboard (236 by 20 cm) were placed vertically in the bottom of the bucket. A wood block (Pinus spp) was also included to establish termite permanence in the collection bucket. Termites were collected from the cardboard and stored at 24ºC in plastic sweater boxes (30 by 19 by 10 cm) with moist corrugated cardboard. Colonies were stored for no longer than two weeks in the sweater boxes.

## Lethal Time Bioassay

### Chemicals

BoraCare™ (40% Disodium octaborate tetrahydrate, 60% mono- and polyethylene glycol; Nisus Co. Rockford, TN.) Tim-Bor™ (98% Disodium octaborate tetrahydrate powder, Nisus Co. Rockford, TN.) Ethylene glycol (99%). Distilled Water.

### Application of Treatments

Four treatments of BoraCare™ were applied at four concentrations (1:1, 1:10, 1:100 and 1:1000, BoraCare™ product: water, by volume) to filter papers. The disodium

octaborate tetrahydrate (DOT) concentration in the four treatments was 303,209 ppm, 73,317 ppm, 7,774 ppm and 782 ppm. Ethylene glycol was applied to filter papers in concentrations of 30.0%, 5.45%, 0.594% and 0.0599%, equivalent to the percentages applied in the BoraCare™ treatments. Ethylene glycol was also applied as a solvent control at stock solution (99%). Tim-Bor™ was applied at the same DOT concentrations as were done in the BoraCare™ applications for the lowest three concentrations. However, because DOT cannot dissolve in the rate it does in ethylene glycol, only half the concentration of DOT could be dissolved for use in the highest concentration of treatment. 4.899 g, 1.182 g, 0.1256 g and 0.01265 g were mixed with water to make a total volume of 25 ml for each application solution of Tim-Bor™. Therefore, the highest concentration of aqueous DOT treatment was 151,605 ppm. Distilled water was applied as a control. The application was done using an adjustable Eppendorf 1 ml volume pipette. Applications of 300 µl were applied to the filter paper achieving complete saturation.

**Bioassay Procedure**

Petri dishes (100 x 15 ml, Fisher Scientific, Ocklawaha, FL) were sealed with parafilm (4 in., American Can Company, Greenwich, CT) around the edges to reduce moisture loss. A hundred termite workers and one termite soldier were placed on top of each treated filter paper (Whatman International Ltd., Maidstone, England, #1, 55 mm) in the Petri dish. After termite workers were placed on top of the treated filter papers, termite mortality observations were made at 20, 45, 50, 57, 65, 70, 80, 96, 115, 135, 140, 165, 192 h, by counting the live termites in the Petri dish. At 192 h, the test was concluded.

### Data Analysis

The experiment was designed as a complete block design with 3 colonies (replicates) for six treatments. Percent mortality data were analyzed by an arcsine square root transformation and means were separated using Student Newman Keuls test in a one-way analysis of variance. $LT_{50}$ and $LT_{95}$ were estimated for each concentration using a probit analysis (SAS, 2001) and the error range was determined by the non-overlapping of 95% confidence intervals.

## Consumption and Mortality Bioassay

### Chemicals

BoraCare™ (40% Disodium octaborate tetrahydrate, 60% mono- and polyethylene glycol Nisus Co. Rockford, TN.). Tim-Bor™ (98% Disodium octaborate tetrahydrate powder, Nisus Co. Rockford, TN.). Ethylene glycol (99%). Propylene glycol (98%). Distilled Water.

### Application of Treatment

Circular filter papers (Whatman International Ltd., Maidstone, England, #1, 55 mm) were oven dryed for 15 min at 150°C and were pre-weighed. Four treatments of BoraCare™ were applied at four concentrations (1:1, 1:10, 1:100 and 1:1000, BoraCare™ product: water, by volume) to filter papers. The disodium octaborate tetrahydrate (DOT) concentration in the four treatments was 303,209, 73,317, 7,774 and 782 ppm. Distilled water was applied as a control and ethylene glycol (99%) was applied as a solvent control. Tim-Bor™ was applied at the same DOT concentrations as were done in the BoraCare™ applications for the lowest three concentrations. However, because DOT cannot dissolve in the rate it does in ethylene glycol, only half the concentration of DOT could be dissolved for use in the highest concentration of

treatment. 4.899 g, 1.182 g, 0.1256 g and 0.01265 g were mixed with water to make a total volume of 25 ml for each application solution of Tim-Bor™. Therefore, the highest concentration of aqueous DOT treatment was 151,605 ppm. DOT was applied as a 20% mixture with propylene glycol was applied at a DOT-propylene glycol rate with water at 1:1. Propylene glycol was applied as a solvent control (98%). The application was done using an adjustable Eppendorf 1 ml volume pipette. Applications of 300 µl were applied to the filter paper for complete saturation.

**Bioassay Procedure**

Glad containers (Glad Products Co. Oakland, CA., 739 ml) were filled with 250 g of builder's sand with 25 ml of water (10% w:w) and uniformly moistened in sealed plastic bags. Termites were aspirated from each colony and sorted into cohorts of 200. Each cohort was introduced into a container and allowed 24 h to burrow from the surface and excavate tunnels in the sand, without the presence of a food source. Hardware cloth (0.64 cm mesh, 23 gauge, LG sourcing, North Wilkesboro, NC) was cut into squares (6 x 6 cm) and centered in the container on the surface of sand. After insecticide treatment, filter papers were placed as a food source on top of the hardware cloth square in each container. After 96 h, the treated filter papers were removed from the containers, cleaned, triple-rinsed with tap water, oven dried at 150°C for 15 min and re-weighed to determine termite consumption. The removed filter papers were replaced by new pre-weighed filter papers of the same concentrations. The containers were left again for 96 h at which time the filter papers were then removed, using the same procedure as above. Survivorship was recorded after 192 h in the container.

**Data Analysis**

The DOT/glycol experiment was designed as a complete block design with eight colonies (replicates) for six treatments. Consumption data (mg) were determined by subtracting the post-treatment weight from the pre-treatment weight and analyzed using a one-way Analysis of Variance ($p = 0.05$) using SAS (SAS Inst. Release 8.1, 2001). Means were separated using Student-Neuman-Keuls method. Mortality data were recorded by counting live termites, Arc sine transformation and means were separated using the Student-Neuman Keuls method. There was 48 experimental units with a total of 9600 termites used in this test.

The aqueous DOT and propylene glycol experiment was designed as a complete block design with four colonies (replicates) for seven treatments. Consumption and Mortality data were determined and analyzed in the same form as mentioned for the DOT/glycol experiment.

CHAPTER 3
RESULTS

## Lethal Time of DOT/glycol.

Termites placed in a Petri dish with treated filter paper aggregated on the paper surface and began feeding within hours. At 20 hours, mortality in the water treatment, and all DOT/glycol treatments did not significantly differ, ranging from 0.67 to 8.33% mortality (Table 1). However, ethylene glycol treatment killed significantly more termites (80.67%) than DOT/glycol treatments. At 45 to 80 hours, 303,209 ppm DOT/glycol treatment increased mortality from 45 to 87%, which was significantly greater than the water treatment. Lower concentrations of borate did not provide significant kill (<33% mortality). At 96 hours, only DOT/glycol treatments ≥73,217 ppm provided significant kill (54 to 94%). After 115 hours all concentrations of borate provided significant mortality. By the end of the study at 192 hours all concentrations of DOT/glycol killed 89 to 100% of termites; whereas, mortality in the water treatment was 21% (Table 1). The $LT_{50}$s of termites exposed to DOT/glycol treatments show relatively rapid mortality (Table 2).

## Lethal Time of Aqueous DOT and Ethylene Glycol

Termites placed in a Petri dish with treated filter paper aggregated on the paper surface and began feeding within hours. At 20 hours, mortality in all treatments did not significantly differ, ranging from 1.67 to 5.67% mortality (Table 3). At 45 to 50 hours, mortality in the water control and all aqueous DOT treatments did not significantly differ, ranging from 6.33 to 34.00% mortality. However, at 40 to 192 hours, ethylene glycol at

30% concentration provided significantly greater kill than all other treatments (Table 3). At 70 hours, mortality from aqueous DOT at 151,605 and 7,774 ppm (49.67 and 40.67% kill) were significantly greater than all other treatments except 73,217 ppm DOT (29.33% kill) and 2.727% Ethylene glycol (20.33%) which were both not significantly greater than the distilled water control (8.33%) and 30% ethylene glycol (94.33%), which was significantly greater. At 80 hours, aqueous DOT at 73,217 ppm increased mortality from 29.33 to 42%, which was significantly greater than the water treatment. At 96 to 115 hours, mortality in 30% ethylene glycol and the three highest concentrated aqueous DOT treatments were significantly greater than the water control (Table 3). All other concentrations of ethylene glycol did not significantly differ from the water control with a range of 11% (water) to 37.33% (2.727% ethylene glycol) mortality. At 96 hours, 30% ethylene glycol caused 100% mortality. At 135 hours, 2.727% ethylene glycol provided significantly greater kill (39.67%) than the water control (11.67%). However, 2.727% ethylene glycol did not provide significantly greater mortality than the two lower concentrations of ethylene glycol or from the lowest concentration of aqueous DOT and significantly less than the higher concentrations of aqueous DOT treatments. From 140 to 192 h, 2.727% ethylene glycol remained significantly less than 7,774 to 151,605 ppm DOT but significantly greater than the two less concentrated ethylene glycol solutions and water. Ethylene glycol treatments at 0.297 and 0.029% and aqueous DOT at 783 ppm did not significantly differ from the water controls for the whole test. At 192 hours, mortality in the water treatment was 13%. Calculated $LT_{50}$s of aqueous DOT treatments show similar results to DOT/glycol as aqueous DOT treatments caused rapid mortality of termites (Table 4).

## DOT/glycol Consumption

Termites began feeding on the filter papers placed in each container in 24 h. In some cases termites excavated soil underneath, while in other containers, termites fed directly on top of the filter paper. Results of the ANOVA for termite 96 h consumption indicated significantly less consumption as the concentrations of DOT/glycol increased. However, consumption of the lowest concentration of DOT-treated filter paper tested, 783 ppm, was not significantly different (Table 5) at 26.13 mg. At 96 h, 303,209 ppm, 73,217 ppm, and 7,774 ppm DOT/glycol consumption were significantly lower than controls. Treatments at 7,774 ppm had significantly greater consumption by termites than treatments of 303,209 ppm DOT/glycol. Consumption was 4.85 mg of filter paper treated with 303,209 ppm, 6.91 mg of filter paper treated with 73,217 ppm, and 14.51 mg of filter paper treated with 7,774 ppm DOT/glycol, while the distilled water control was measured at 31.213 mg. Ethylene glycol treated filter paper consumption, 26.913 mg, did not significantly differ in comparison with the distilled water control.

When the filter papers were removed and replaced after 96 h, termites were less voracious because termite consumption decreased in all treatments. Results of the ANOVA from the consumption of filter papers measured from 96-192 h indicated similar significance as consumption after 96 h. (Table 5) Results indicated significant difference for concentrations above 783 ppm DOT/glycol.

Consumption was combined for both periods (0-96 h and 96-192 h) for a total consumption mass. Consumption totals at 192 h produced similar results as results from 0-96 h and 96-192 h; there was significant difference in filter paper consumption in applications of DOT concentrations above 783 ppm compared with filter papers treated with distilled water. (Table 5) Termites consumed a total of 5.51 mg filter paper treated

with 303,209 ppm, 8.88 mg filter paper treated with 73,217 ppm and 19.16 mg filter paper treated with 7,774 ppm DOT/glycol.

At 303,209 ppm DOT, two of the replicates appeared to avoid the treated filter paper after initial contact. This resulted in increased survivorship for both replicates and considerable reduction in consumption of filter paper compared with the average mortality and consumption at 303,209 ppm DOT. Deterrence of feeding had occurred because termites were actively avoiding the treated cellulose and refraining from feeding.

Termites fed upon the distilled water treated filter papers at an average of 0.156 mg/termite over 0-96 h. In comparison with DOT/glycol treatment at the label rate, termites fed on the 303,209 ppm treated filter papers at an average of 0.024 mg/termite over the 0-96 h period. At 96 h, termite consumption of DOT/glycol-treated filter papers is inversely related to treatment concentration. Although termites consumed significantly less filter paper from 7,774 to 303,209 ppm DOT, they ingested more μg of DOT (Fig. 1). Therefore, the highest concentration of treatment resulted in the largest ingestion of DOT.

## DOT/glycol Mortality

Mortality in the containers was observed within 96 h. Results of the ANOVA for mortality resulted in significant differences between the distilled water control and DOT concentrations above 783 ppm. Mortality at 7,774 ppm resulted in 44.4% kill. Termites in the highest concentrations of DOT, 73,217 and 303,209 ppm, were recorded at 73.1 and 81.3% mortality after eight days compared with the distilled water control at 13.9%. The ethylene glycol treatment did not result in significant mortality from the control (Table

5). Treatments of DOT/glycol caused more mortality in concentrations ≥7,774 ppm DOT. Mortality increased as ingestion of µg of DOT increased (Fig. 2).

**Aqueous DOT/Propylene Glycol Consumption**

Filter papers were placed inside the each container and the termites contacted the paper within 24 hours. Results of the ANOVA at 96 h indicate significantly less consumption than on the filter paper treated with the distilled water control at 24.78 mg, except for the lowest concentration of aqueous DOT (783 ppm) at 25.33 mg. At 96 h, termite consumption with aqueous DOT-treated filter papers at the highest concentrations (151,605 and 73,217 ppm) and the mixture of 20% DOT (303,209 ppm), 30% propylene glycol and 50% water by volume were not significantly different from each other ( all ≤0.10 mg). Termite consumption at 7,774 ppm DOT (8.68 mg) and in the propylene glycol solvent control (98%) (5.75 mg) were significantly greater than both the aforementioned higher concentrations of DOT, but significantly less than the least concentrated DOT treatment at 783 ppm (25.33 mg) and the distilled water control (24.78 mg) (Table 6).

Results from 96-192 h indicate no significant difference between the consumption of DOT treatments at 151,605 ppm, 73,217 ppm, 7,774 ppm, the DOT/propylene glycol mixture, and propylene glycol as a solvent control. DOT treated filter paper consumption at 783 ppm was significantly less at 17.45 mg, than the distilled water control at 25.60 mg.

Total termite consumption of filter paper, from 0-192 h, indicates every treatment is significantly less than the distilled water control at 50.38 mg. Aqueous DOT treated at 783 ppm had the least change in consumption at 42.78 mg. DOT treated at 7,774 ppm was not significantly different at 10.35 mg than consumption of filter papers treated with

the propylene glycol solvent control at 7.55 mg. Consumption of the filter papers treated with propylene glycol was not significantly different than the remaining treatments; DOT treated at 151,605 and 73,317 and the DOT/propylene glycol mixture were consumed at 1.53 mg, 0.55 mg, and 1.08 mg, respectively (Table 6).

There is significantly less consumption of treated filter paper as concentrations of DOT on the filter papers are increased. There was no significant difference of consumption of filter papers treated at >73,217 ppm aqueous DOT-treated filter papers or DOT in propylene glycol.

**Aqueous DOT/Propylene Glycol Mortality**

Mortality in the containers was observed within 96 h. Results of the ANOVA for mortality indicate significant differences between the distilled water control and DOT concentrations above 783 ppm. Mortality in the higher concentrations of DOT and the mixture of DOT/propylene glycol were not significantly different at 86.0% (151,605 ppm DOT), 94.9% (73,317 ppm DOT), 76.1% (7,774 ppm DOT) and 88.4% (20% DOT, 30% propylene glycol and 50% distilled water). Mortality from the propylene glycol solvent control was significant from all other treatments at 99.9%. Mortality caused by aqueous DOT treatments did not statistically differ at concentrations $\geq$7,774 ppm (Table 6).

Table 3-1. Lethal effects of DOT/glycol-treated filter papers on *R flavipes* workers (n=100)

| Treatment | Mortality (% ± SE) at time (h) | | | | | | |
|---|---|---|---|---|---|---|---|
| | 20 | 45 | 50 | 57 | 70 | 80 | 96 |
| Control | 0.67 ± 0.33b | 8.33 ± 1.86c | 9.00 ± 1.53c | 9.67 ± 2.19c | 10.00 ± 2.52b | 10.67 ± 3.18b | 11.67 ± 2.63c |
| Ethylene glycol | 80.67 ± 8.95a | 97.00 ± 1.53a | 99.00 ± 1.00a | 99.33 ± 0.67a | 100.0 ± 0.00a | --- | --- |
| DOT/glycol[1] | | | | | | | |
| 783 | 0.67 ± 0.67b | 4.67 ± 1.76c | 6.33 ± 2.19c | 8.67 ± 2.91c | 10.00 ± 4.16b | 13.33 ± 6.38b | 18.33 ± 6.84bc |
| 7,774 | 1.00 ± 0.58b | 8.00 ± 4.51c | 9.00 ± 5.03c | 10.33 ± 6.36c | 15.00 ± 8.50b | 22.67 ± 14.7b | 34.00 ± 13.7bc |
| 73,217 | 1.67 ± 1.20b | 16.67 ± 9.94c | 17.67 ± 10.5c | 21.00 ± 9.17c | 28.67 ± 9.82b | 33.00 ± 10.6b | 54.33 ± 18.7b |
| 303,209 | 8.33 ± 5.36b | 45.33 ± 10.2b | 55.00 ± 11.2b | 66.33 ± 9.39b | 82.00 ± 10.7a | 87.67 ± 6.89a | 94.33 ± 4.18a |

Means followed by the same letter are not significantly different ($\alpha = 0.05$ Student Newman Keuls [SAS, 2001]).

[1] Disodium octaborate tetrahydrate/ethylene glycol (ppm of DOT on filter paper)

Table 3-1. Continued

| Treatment | Mortality (% ± SE) at time (h) | | | | | |
|---|---|---|---|---|---|---|
| | 96 | 115 | 135 | 140 | 165 | 192 |
| Control | 11.67 ± 2.73b | 13.00 ± 3.00d | 15.00 ± 2.65e | 15.33 ± 2.96c | 16.00 ± 3.06b | 21.33 ± 3.76b |
| Ethylene glycol | --- | --- | --- | --- | --- | --- |
| DOT/glycol[1] | | | | | | |
| 783 | 18.33 ± 6.84b | 41.67 ± 9.17c | 62.33 ± 2.03d | 77.33 ± 9.26b | 81.67 ± 8.97a | 89.33 ± 6.12a |
| 7,774 | 34.00 ± 13.7b | 49.67 ± 14.3c | 75.33 ± 1.45c | 87.67 ± 5.46b | 91.67 ± 5.61a | 94.67 ± 3.18a |
| 73,217 | 54.33 ± 18.7b | 78.33 ± 9.68b | 88.00 ± 2.08b | 94.33 ± 1.20b | 97.00 ± 1.15a | 99.00 ± 0.58a |
| 303,209 | 94.33 ± 4.18a | 98.00 ± 1.53a | 99.67 ± 0.33a | 100.0 ± 0.00a | --- | --- |

Means followed by the same letter are not significantly different ($\alpha = 0.05$ Student Newman Keuls [SAS, 2001]).

[1] Disodium octaborate tetrahydrate/ethylene glycol (ppm of DOT on filter paper)

Table 3-2. Toxicity of disodium octaborate tetrahydrate in ethylene glycol to 100 *R. flavipes* workers.

| Treatment | | Model Parameters[c] | | Lethal time (hour)[d] | | Model fit | | |
|---|---|---|---|---|---|---|---|---|
| DOT/glycol[a] | *n*[b] | Intercept ±SE | Slope ± SE | $LT_{50}$ (95% FL) | $LT_{95}$ (95% FL) | $\chi^2$ | df | P |
| 783 | 1200 | -24.0 ± 2.5 | 11.4 ± 1.2 | 127.0 (123.5-130.8) | 177.0 (165.7-195.2) | 2.57 | 2 | 0.28 |
| 7,774 | 1800 | -15.2 ± 1.7 | 7.3 ± 0.8 | 117.5 (112.5-123.9) | 197.1 (175.8-237.1) | 2.24 | 2 | 0.32 |
| 73,217 | 1500 | -14.1 ± 1.3 | 7.1 ± 0.6 | 95.24 (91.54-99.08) | 162.0 (148.6-182.6) | 2.87 | 3 | 0.41 |
| 303,209 | 1500 | -9.2 ± 1.2 | 5.4 ± 0.7 | 49.69 (46.21-52.46) | 99.93 (88.66-121.2) | 0.16 | 3 | 0.98 |

[a] Disodium octaborate tetrahydrate/ethylene glycol (ppm of DOT on filter paper)
[b] The number of trials with 300 termites at each observation
[c] The intercept and slope parameters are for models in which the independent variable is the natural logarithm of the exposure time (hour).
[d] Abbot's correction was performed to adjust the data with control mortality

Table 3-3. Lethal effects of borate and ethylene glycol treated filter papers on *R flavipes* workers (n=100)

| Treatment | Mortality (% ± SE) at time (h) | | | | | |
|---|---|---|---|---|---|---|
| | 20 | 45 | 50 | 70 | 80 | 96 |
| Control | 4.33 ± 1.20a | 6.33 ± 0.33b | 6.33 ± 0.33b | 8.33 ± 0.33d | 10.33 ± 0.88d | 10.67 ± 0.88c |
| Ethylene glycol %[a] | | | | | | |
| 30.000 | 4.67 ± 2.19a | 78.33 ± 5.93a | 82.33 ± 6.12a | 94.33 ± 0.33a | 99.67 ± 0.33a | 100.0 ± 0.00a |
| 2.727 | 1.67 ± 0.88a | 11.67 ± 0.67b | 12.33 ± 0.88b | 20.33 ± 4.48bcd | 26.00 ± 5.69cd | 32.00 ± 7.55c |
| 0.297 | 1.33 ± 0.33a | 8.00 ± 1.53b | 8.33 ± 1.20b | 13.00 ± 2.52cd | 15.00 ± 2.08d | 17.00 ± 2.65c |
| 0.029 | 3.00 ± 0.58a | 12.33 ± 0.88b | 12.33 ± 0.88b | 15.33 ± 1.20cd | 18.00 ± 2.08d | 21.00 ± 2.08c |
| Aqueous DOT[b] | | | | | | |
| 783 | 2.33 ± 1.45a | 8.00 ± 2.65b | 8.00 ± 2.65b | 17.00 ± 2.08cd | 20.67 ± 2.73d | 22.00 ± 2.08c |
| 7,774 | 2.67 ± 1.76a | 8.67 ± 0.33b | 9.00 ± 2.52b | 40.67 ± 7.69b | 56.33 ± 3.28b | 64.33 ± 3.28b |
| 73,217 | 4.00 ± 0.58a | 8.00 ± 1.53b | 8.33 ± 1.45b | 29.33 ± 4.84bcd | 42.00 ± 5.13bc | 59.33 ± 4.26b |
| 151,605 | 5.67 ± 1.76a | 11.00 ± 1.00b | 34.00 ± 11.5b | 49.67 ± 13.3bc | 61.33 ± 14.2b | 75.33 ± 15.8b |

Means followed by the same letter are not significantly different ($\alpha = 0.05$ Student Newman Keuls [SAS, 2001]).

[a] Solutions of ethylene glycol and water. Percentages are ethylene glycol content

[b] Aqueous disodium octaborate tetrahydrate (ppm of DOT on filter paper)

Table 3-3. Continued

| Treatment | Mortality (% ± SE) at time (h) | | | | |
|---|---|---|---|---|---|
| | 115 | 135 | 140 | 165 | 192 |
| Control | 11.00 ± 0.58b | 11.67 ± 0.67c | 12.00 ± 0.58c | 12.67 ± 0.88c | 13.00 ± 0.58c |
| Ethylene glycol %[a] | | | | | |
| 30.000 | --- | --- | --- | --- | --- |
| 2.727 | 37.33 ± 8.01b | 39.67 ± 6.74b | 42.00 ± 6.08b | 44.67 ± 5.81b | 48.00 ± 4.36b |
| 0.297 | 21.33 ± 4.10b | 22.67 ± 3.84bc | 23.00 ± 3.79c | 23.33 ± 4.10c | 24.00 ± 3.79c |
| 0.029 | 21.67 ± 1.45b | 22.00 ± 1.73bc | 22.00 ± 1.73c | 22.67 ± 1.45c | 23.00 ± 1.53c |
| Aqueous DOT[b] | | | | | |
| 783 | 22.33 ± 2.40b | 22.33 ± 2.40bc | 22.33 ± 2.40c | 23.00 ± 2.08c | 24.00 ± 2.31c |
| 7,774 | 71.67 ± 6.12a | 79.00 ± 4.93a | 79.67 ± 4.63a | 84.33 ± 3.76a | 91.33 ± 3.93a |
| 73,217 | 67.00 ± 3.06a | 74.33 ± 5.24a | 76.00 ± 5.57a | 83.67 ± 1.45a | 93.67 ± 2.33a |
| 151,605 | 75.33 ± 15.8a | 81.00 ± 11.3a | 83.67 ± 10.4a | 89.67 ± 8.09a | 95.67 ± 6.12a |

Means followed by the same letter are not significantly different ($\alpha = 0.05$ Student Newman Keuls [SAS, 2001]).

[a] Solutions of ethylene glycol and water. Percentages are ethylene glycol content

[b] Aqueous disodium octaborate tetrahydrate (ppm of DOT on filter paper)

Table 3-4. Toxicity of disodium octaborate tetrahydrate and ethylene glycol to 100 *R. flavipes* workers.

| Treatment | | Model Parameters[d] | | Lethal time (hour)[e] | | Model fit | | |
|---|---|---|---|---|---|---|---|---|
| | *n*[c] | Intercept ±SE | Slope ± SE | $LT_{50}$ (95% FL) | $LT_{95}$ (95% FL) | $\chi^2$ | df | P |
| Ethylene glycol %[a] | | | | | | | | |
| 30.000 | 900 | -6.2 ± 1.3 | 4.2 ± 0.7 | 29.61 (22.01-34.33) | 72.90 (65.79-88.71) | 0.18 | 1 | 0.67 |
| 2.727 | 3300 | -4.5 ± 0.2 | 2.0 ± 0.1 | 181.9 (168.3-199.4) | 1228 (956.4-1674) | 7.04 | 9 | 0.63 |
| 0.297 | 3300 | -3.7 ± 0.2 | 1.3 ± 0.1 | 519.3 (402.9-739.7) | 8667 (4482-22023) | 7.80 | 9 | 0.55 |
| 0.029 | 3300 | -2.1 ± 0.2 | 1.0 ± 0.1 | 857.7 (571.7-1599) | 44182 (14712-2.43e5) | 10.8 | 9 | 0.29 |
| Aqueous DOT[b] | | | | | | | | |
| 783 | 900 | -4.9 ± 0.6 | 2.2 ± 0.4 | 193.5 (137.7-368.1) | 1098 (518.1-4684) | 0.08 | 1 | 0.78 |
| 7,774 | 1500 | -6.4 ± 0.7 | 3.4 ± 0.3 | 76.93 (71.82-81.14) | 232.9 (200.4-289.8) | 4.15 | 3 | 0.25 |
| 73,217 | 2100 | -7.7 ± 0.5 | 3.9 ± 0.2 | 90.56 (86.89-93.96) | 237.9 (217.4-266.2) | 6.11 | 5 | 0.29 |
| 151,205 | 2100 | -9.2 ± 0.4 | 4.8 ± 0.2 | 84.48 (81.95-86.97) | 187.4 (175.2-202.1) | 4.73 | 5 | 0.45 |

[a] Solutions of ethylene glycol and water. Percentages are ethylene glycol content
[b] Disodium octaborate tetrahydrate/ethylene glycol (ppm of DOT on filter paper)
[c] The number of trials with 300 termites at each observation
[d] The intercept and slope parameters are for models in which the independent variable is the natural log of the exposure time (hour)
[e] Abbot's correction was performed to adjust the data with control mortality

Table 3-5. Consumption (mg) of DOT/glycol treated filter paper by *R. flavipes* workers (n = 200) and resultant mortality

| Treatment | Mean consumption (mg) ± SE | | | % Mortality ± SE |
|---|---|---|---|---|
| | 0-96 h | 96-192 h | Total | 192 h |
| Control | 31.21 ± 3.91a | 20.00 ± 3.12a | 51.21 ± 4.80a | 13.9 ± 2.2a |
| Ethylene glycol | 26.91 ± 2.39a | 15.13 ± 2.11a | 42.04 ± 3.83a | 29.9 ± 5.5ab |
| DOT/glycol[1] | | | | |
| 783 | 26.13 ± 2.50a | 13.86 ± 3.26a | 39.99 ± 4.28a | 22.8 ± 4.4ab |
| 7,774 | 14.51 ± 4.15b | 4.65 ± 0.82b | 19.16 ± 4.00b | 44.4 ± 8.5b |
| 73,217 | 6.91 ± 1.35bc | 1.96 ± 0.80b | 8.88 ± 1.64b | 73.1 ± 10c |
| 303,209 | 4.85 ± 0.88c | 0.66 ± 0.17b | 5.51 ± 0.89b | 81.3 ± 7.8c |

Means followed by same letter are not significantly different ($\alpha = 0.05$, Student Newman Keuls [SAS, 2001]).

[1] Disodium octaborate tetrahydrate/ethylene glycol solution (ppm of DOT on filter paper)

Table 3-6. Consumption (mg) of aqueous DOT and DOT/propylene glycol treated filter paper by *R. flavipes* workers (n = 200) and resultant mortality

| Treatment | Mean consumption (mg) ± SE | | | % Mortality ± SE |
|---|---|---|---|---|
| | 0-96 h | 96-192 h | Total | 192 h |
| Control | 24.78 ± 2.35a | 25.60 ± 0.84a | 50.38 ± 3.03a | 12.4 ± 1.1a |
| Propylene glycol | 5.75 ± 1.65b | 1.80 ± 0.18c | 7.55 ± 1.63cd | 99.9 ± 0.1c |
| Aqueous DOT[1] | | | | |
| 783 | 25.33 ± 2.19a | 17.45 ± 3.37b | 42.78 ± 4.77b | 24.6 ± 3.4a |
| 7,774 | 8.68 ± 0.97b | 1.68 ± 0.46c | 10.35 ± 1.25c | 76.1 ± 2.7b |
| 73,217 | 0.10 ± 0.10c | 0.45 ± 0.18c | 0.55 ± 0.27d | 94.9 ± 4.0bc |
| 151,605 | 0.00 ± 0.00c | 1.53 ± 0.51c | 1.53 ± 0.51d | 86.0 ± 10.3bc |
| DOT/propylene glycol[2] | | | | |
| 303,209 | 0.00 ± 0.00c | 1.80 ± 0.38c | 1.80 ± 0.38d | 88.4 ± 4.0bc |

Means followed by same letter are not significantly different ($\alpha = 0.05$, Student Newman Keuls [SAS, 2001]).
[1] Disodium octaborate tetrahydrate applied in water solution.
[2] Disodium octaborate tetrahydrate/propylene glycol solution (ppm of DOT on filter paper)

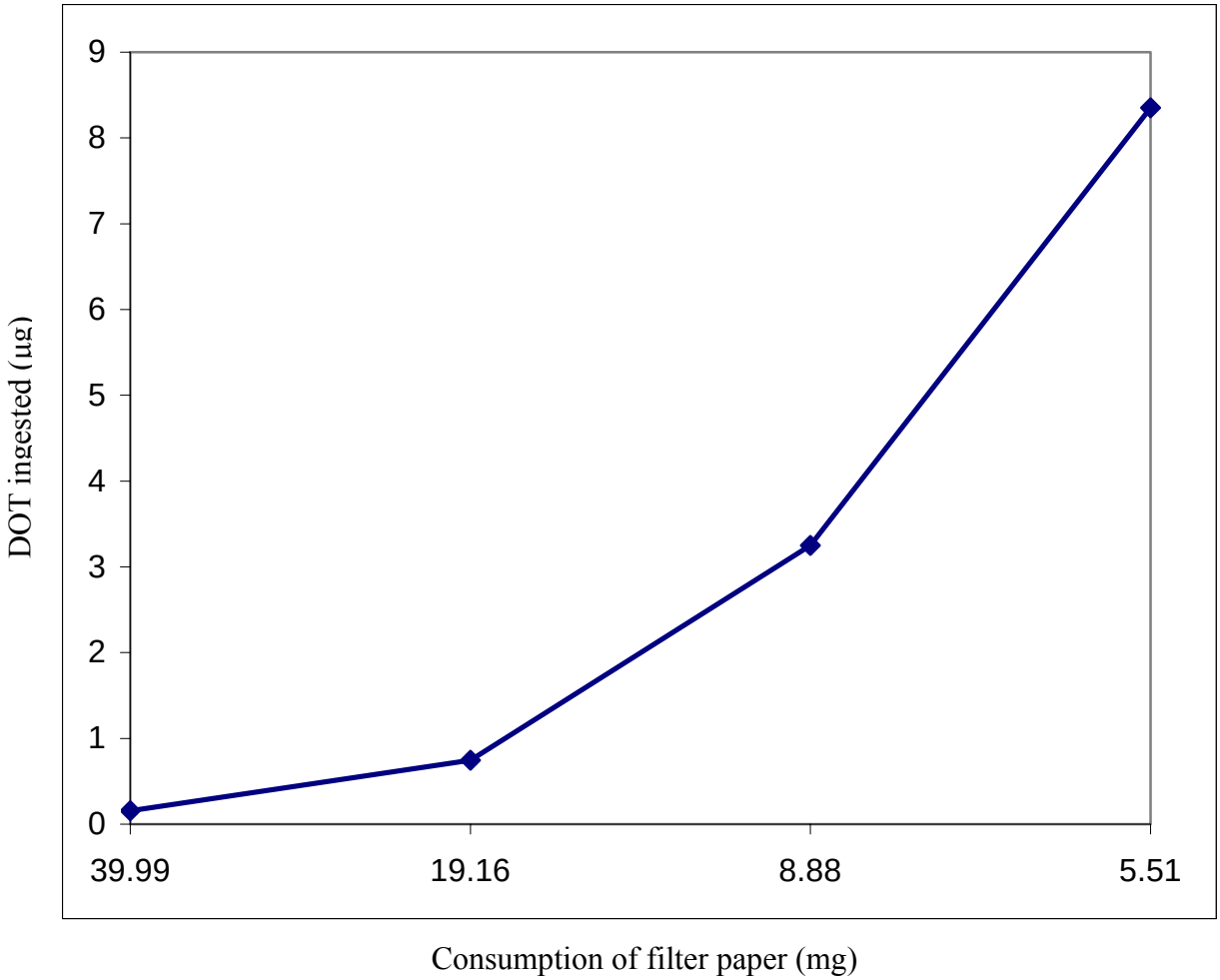


Figure. 3-1. Consumption of filter paper (mg) by termites as a function of DOT ingested (µg). Consumption was observed at 192 h.The graph was charted using the consumption data from the DOT/glycol consumption/mortality bioassay.

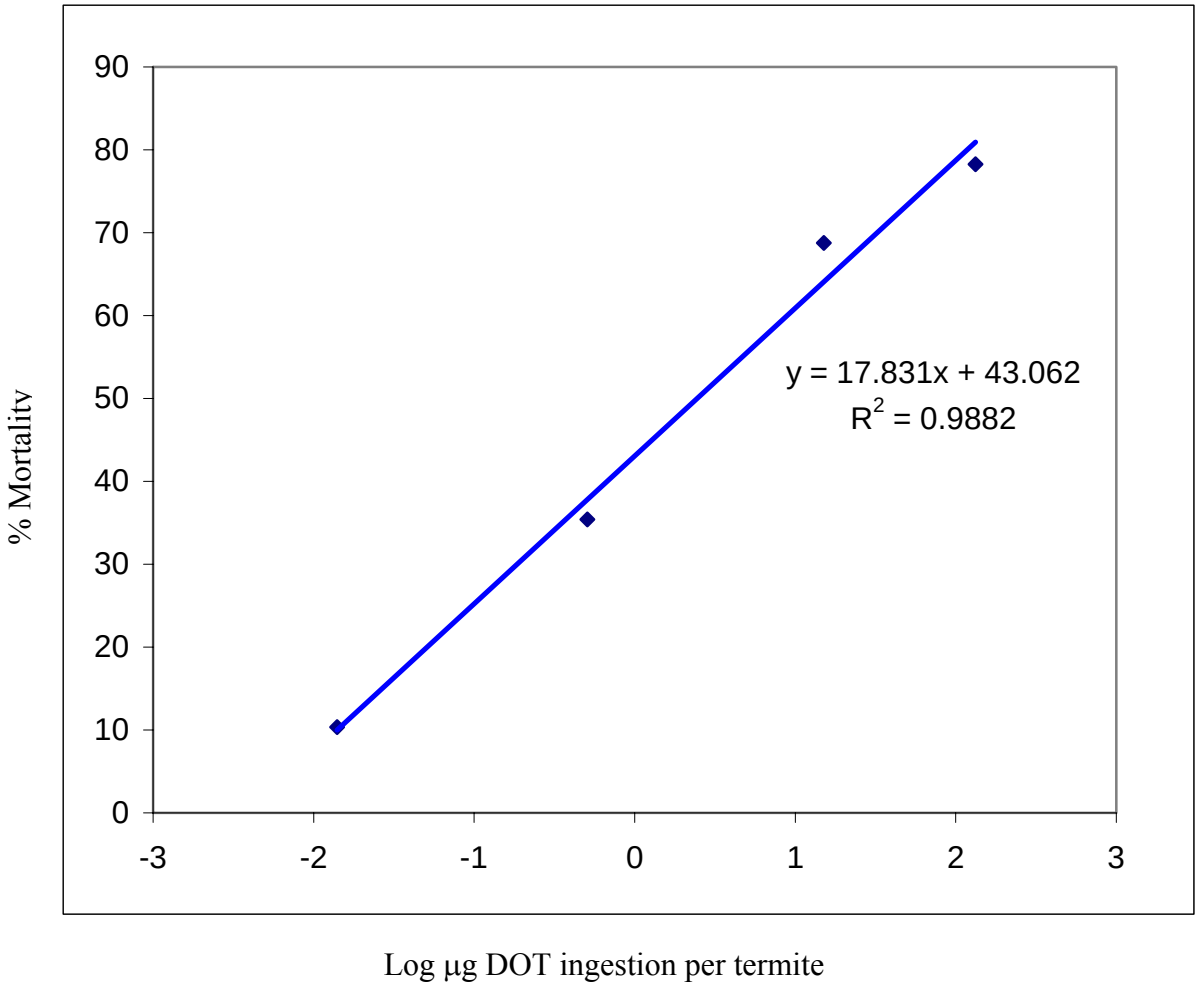


Figure 3-2. Log µg ingestion of DOT per termite as a function of mortality (%). Mortality was recorded at 192 h after treatment and corrected by Abbott's formula (SAS 2001).The graph was charted using the consumption and mortality data from the DOT/glycol consumption/mortality bioassay.

# CHAPTER 4
# DISCUSSION

Contact with ethylene glycol can cause rapid termite mortality. Surprisingly, the 30% ethylene glycol solvent caused the most rapid termite mortality ($LT_{50}$ of 30% Ethylene glycol <30 h). Ethylene glycol is a potential desiccant and probably dehydrated the termites, which were forced into contact with the treated filter papers. Desiccation with ethylene glycol causes termites to become sluggish and appear shriveled and smaller than healthy termites. These effects were specifically noted when termites came into direct contact with the liquid solvent on filter papers. In a study where contact with ethylene glycol was reduced by feeding termites treated saw-dust, Tokoro and Su (1993) found no significant mortality. When ethylene glycol was treated to wood blocks, mortality of termites was only slightly significant when compared with control mortality. In this study, ethylene glycol was consumed at the same mass as the distilled water control, but similar to Tokoro and Su (1993), I observed elevated but not significant mortality. Feeding on cellulose containing ethylene glycol did not cause desiccation and subsequent mortality.

DOT killed termites rapidly. At concentrations $\geq$7,774 ppm of DOT/glycol, termite mortality was >85% within 192 h. Although ethylene glycol accelerated mortality because it contacted the termites in the DOT/glycol treatment, aqueous treatments of DOT $\geq$ 7,774 ppm caused >85% mortality within 192 h. Therefore, aqueous DOT treated filter papers proved the effectiveness of DOT as a potent termiticide without ethylene

glycol as a solvent. DOT dissolved in ethylene glycol accelerated mortality of termites, probably due to the combination of contact and ingestion poisons.

Termite consumption of treated filter papers decreased as concentrations of DOT increased. Similarly, Su and Scheffrahn (1991a, 1991b) found that termite consumption of cellulose was severely deterred at concentrations >1000 ppm. In my study, 783 ppm DOT reduced cellulose ingestion by ~10%. However, at 7,774 ppm DOT, ingestion of treated cellulose was reduced by ~54%. At 303,209 ppm DOT, feeding was reduced by ~84%. Even at the highest concentrations, most termites fed and subsequently died.

Although effective concentration levels of DOT have been found to severely limit termite consumption of cellulose, whether DOT is a termite deterrent of feeding cannot be determined by measures of consumption alone. Other studies (Su and Scheffrahn 1991a, Tokoro and Su 1993, Grace and Yamamoto 1994) recorded termite mortality 7, 14 or 28 days after treatment. Su and Scheffrahn (1991a) specifically noted >85% mortality in 7 d. Obviously consumption amounts recorded after 7, 14 and 28 d will be affected by mortality among feeding termites and reductions of consumption may not be due to feeding deterrence. Even in my study where consumption was recorded after 96 h, mortality effects on consumption were limited but not completely eliminated.

In all concentrations of DOT/glycol <303,209 ppm, termites fed on the treated filter paper. Only at the highest concentration of DOT/glycol treatment (303,209 ppm) was complete feeding deterrence observed in 2 of the 8 treatment replicates. Feeding deterrence was observed as termites initially contacted and fed on the treated filter papers, but subsequently preferred to cease feeding. At treatments <303,209 ppm,

DOT/glycol is not a feeding deterrent and reductions in consumption are primarily due to mortality effects.

Aqueous treated DOT applied to filter papers caused greatest reduction in termite consumption but also caused greatest mortality for each treatment >783 ppm DOT. A possible example of this is the evaporation rate of the DOT's solvents. Ethylene glycol has a low vapor pressure (0.06 mm Hg at 20°C) and is slow to evaporate compared with water (17.54 mm Hg at 20°C), which evaporates quickly (Budavari 1996). As the solvent evaporates, DOT precipitates. Solid DOT particles blocked the termite gut, similar to findings of Ebeling (1995) that borate ingestion blocked cockroach digestion. As a result, solid DOT limited termite ingestion but still was capable of causing mortality probably by blocking passage through the gut and subsequently poisoning the stomach.

When mortality of termites was observed at 192 h, greater mortality had occurred in treatments at the highest concentration of DOT. Termite mortality over the 192 h period of the test confirmed the efficacy of DOT/glycol and aqueous DOT treated cellulose as effective means to prevent termite feeding and cause termite mortality. Termite mortality was significantly greater than the distilled water control in treatments $\geq$7,774 ppm DOT. Mortality could be expected to be greater for increasing concentrations of DOT. As mentioned prior, termites, although consuming less filter paper, were ingesting greater quantities of DOT with higher concentration of treatment. Therefore, high mortality was caused by ingestion of lethal doses of DOT. Analysis of the DOT/glycol mortality data as a function of DOT consumed per termite shows a logarithmic correlation ($r^2 = 0.9882$). (Fig.1) Termites consumed more active ingredient with the higher concentrations of DOT/glycol application even though the termites

consumed far less filter paper. The largest increase in mortality was associated with an increase of termite consumption from 0.745 to 3.251 μg DOT, which resulted in an increase of mortality from 35.42 to 68.75%.

From conclusions drawn from results of this study, borates cannot be assumed or proved to be feeding deterrents of treated cellulose. Rapid mortality of termites caused by borates, whether visible or even quantifiable does not matter, the amount of cellulose consumption is irrelevant at the highest concentration of borate treated-filter paper if such concentrations of borates kill termites so quickly. Concentrations of active ingredient are so high, contact with treatment would probably lead to enough borates deposited on the termite cuticle that grooming would lead to the acquisition of a lethal dose. As the treatment concentration decreases, increased consumption occurs while ingestion of DOT decreases. Therefore, termites are not deterred from feeding at higher concentrations because higher concentrations of DOT are being ingested at the higher concentrations of DOT treatments. Ingestion of higher concentrations of DOT causes greater termite mortality. Rapid time to mortality, especially with concentrations ≥7,774 ppm DOT and observed mortality as a result of mg DOT ingested, confirm the likelihood of mortality, rather than borate feeding deterrence as the reason for a decrease in consumption of cellulose treated with >7,774 ppm DOT compared with distilled water treated controls.

Although consumption of filter paper treated with 783 ppm DOT did not cause significantly greater termite mortality compared with distilled water treatments, it is logical to assume that continued feeding on cellulose at that concentration of treatment would eventually lead to termite mortality. As the amount of DOT ingested increases, termites would acquire a lethal dose of DOT.

Even with a lack of termite feeding deterrence at low concentrations (<1000 ppm, Su and Scheffrahn 1991a), the relative quick termite mortality as a result of exposure to 783 ppm DOT has implications for baiting. The $LT_{50}$ of aqueous-treated DOT and DOT/glycol at 783 ppm at <200 h does not lend itself to an effective time for transfer of the bait throughout the colony. Therefore, the rapid mortality of DOT with and without ethylene glycol, even at low concentrations does not support DOT use as a potential bait.

The similar chemical properties of ethylene glycol and propylene glycol enticed the experimentation of propylene glycol as a substitute for ethylene glycol to carry DOT into solution. Propylene glycol has a considerably lower mammalian toxicity compared with ethylene glycol (Budavari 1996). Propylene glycol was also able to dissolve DOT into a 40% solution. The 20% DOT/ 30% propylene glycol (DOT/ propylene glycol) treated filter papers caused >88% termite mortality in 192 h. Propylene glycol applied to filter papers at 98% caused 100% mortality in the same period of time. Propylene glycol, similar to ethylene glycol, probably desiccated the termites, yet when ethylene glycol was treated at 99% and fed to termites with the sand buffer in the consumption/mortality bioassay, mortality was non-significant compared to distilled water controls. Therefore, propylene glycol may infer subsequent toxic properties to termite ingestion. The effectiveness of borates in propylene glycol, in terms of reducing termite consumption, causing termite mortality and being less toxic to humans than ethylene glycol leads to the possibility of the development of this combination in lieu of ethylene glycol based borate treatments.

The eastern subterranean termite consumes cellulose for nourishment. Houses that contain structural wood components are potential targets of termite attack if methods to

prevent access to termites are not taken. Treatment of wood with DOT can be an effective preventative measure to avoid termite attack on wood and is being applied as a stand-alone new construction treatment. Wood near the ground and close to termite entry is treated, whereas wood higher in the structure is not usually treated.

My study determined that DOT kills termites rapidly by ingestion, consequently limiting damage to wood in the structure. DOT/glycol treatments were not found to be deterrents of feeding except at the highest concentrations. As a result, untreated wood in the structure can be protected because treated wood would be a more convenient food source and the treatment would probably not cause feeding deterrence. DOT/glycol treatments appear to have promise to prevent damage from new construction.

LIST OF REFERENCES

**Barnes, H.M., Amburgey, T.L., Williams, L.H., and J.J. Morrell. 1989.** Borates as wood preserving compounds. The status of research in the United States. Int'l. Res. Group on Wood Preservation. Doc. No. IRG/WP/3542. 16 pp.

**Bartz S. 1979.** Evolution of eusociality in termites. *Proc. Natl. Acad. Sci.* 76(11): 5764-5768.

**Becker, G. 1976.** Treatment of wood by diffusible salts. *J. Inst. Wood Sci.* 7: 30-36.

**Bennett, G.W., Owens, J.M., and R.M. Corrigan. 1988.** Truman's scientific guide to pest control operations. Edgell Communications, Duluth, MN. 495 pp.

**Budavari, E. 1996.** The Merck index: an encyclopedia of chemicals drugs and biologicals. 12th ed. Merck & Co. Inc. Whitehouse Station, NJ.

**Clement, J., and A. Bagneres. 1998.** Nestmate recognition in termites, pp 126-155 in Pheromone communication in social insects: Ants, wasps, bees and termites. (Vander Meer, R.K., Breed, M.D., Winston, M.L. and K.L. Espelie, eds.) Westview Press, Boulder, CO.

**Cochran, D.G. 1995.** Toxic effects of boric acid on the German cockroach: *Experientia*. 51: 561–563.

**Cornelius, M.J., and J.M. Bland. 2001.** Trail-following behavior of *Coptotermes formosanus* and *Reticulitermes flavipes* (Isoptera: Rhinotermitidae): Is there a species-specific response? *Environ. Entomol.* 30(3): 457-465.

**Cummins, J.E. 1939.** The preservative treatment of timber against the attack of the powder post borer (*Lyctus brunneus* Stephens) by impregnation with boric acid. *J. Council Sci. Ind. Res.* 12: 30-49

**Delate, K.M., Grace, J.K., and C.H.M. Tome. 1995.** Potential use of pathogenic fungi in baits to control the Formosan subterranean termite (Isoptera:Rhinotermitidae). *J. Appl. Entomol.*, 119: 429-433.

**Ebeling, W. 1995.** Inorganic insecticides and dusts pp193-230 *in* Understanding and controlling the German cockroach (M. K. Rust J. M. Owens D. A. Reierson ed.). Oxford University Press, New York.

**Epsky, N.D., and J.L. Capinera. 1988.** Efficacy of the entomagenous nematode Steinernema feltiae against a subterranean termite, Reticulitermes tibialis (Isoptera:Rhinotermitidae) *J. Econ. Entomol.* 81: 1313-1317.

**Forschler, B.T. 1999.** Biology of subterranean termites of the genus Reticulitermes. Part II, pp 31-50. National Pest Control Association research report on subterranean termites. National Pest Control Associations. Dunn Loring, VA.

**Grace, J.K. 1991.** Response of eastern and Formosan subterranean termites (Isoptera: Rhinotermitidae) to borate dust and soil treatments. *J. Econ. Entomol.* 84: 1753-1757.

**Grace, J.K. 1997.** Review of recent research on the use of borates for termite prevention. The Second International Conference on Wood Preservation with Diffusible Preservatives and Pesticides. Madison. USA. pp 85-91.

**Grace, J.K., and R.T. Yamamoto. 1992.** Termiticidal effects of a glycol borate wood surface treatment. *Forest Products J.* 42(11/12): 46-48.

**Grace, J.K., and R.T. Yamamoto. 1994.** Simulation of remedial borate treatments intended to reduce attack on Douglas-fir lumber by the Formosan subterranean termite (Isoptera: Rhinotermitidae). *J. Econ. Entomol.* 87(6):1547-1554.

**Grace, J.K., Yamamoto, R.T., and M. Tamashiro. 2000.** Toxicity of sulfuramid to *Coptotermes formosanus* (Isoptera:Rhinotermitidae) *Sociobiol.* 35 (3): 457-466.

**Greaves, H. 1990.** Wood protection with diffusible preservatives: historical perspectives in Australia. Proceedings of the First International Conference on Wood Protection with Diffusible Preservatives. Nashville, TN. pp.14-18.

**Hagen, H. A. 1876.** The probable danger from white ants. *Amer. Naturalist* 10(7): 401-410.

**Hedlund, J.C., and G. Henderson. 1999.** Effect of available food size on search tunnel formation by the Formosan subterranean termite (Isoptera: Rhinotermitidae). *J. Econ. Entomol.* 92(3): 610-616.

**Hickin, N.E. 1971.** Termites: a world problem. Hutchinson & Co. Ltd., London.

**Howard, R.W., and M.I. Haverty. 1980.** Reproductives in mature colonies of *Reticulitermes flavipes*: abundance, sex-ratio, and association with soldiers. *Environ. Entomol.* 9: 458-460.

**Krieger, R.I., Dinoff, T.M., and J. Peterson. 1996.** Human disodium octaborate tetrahydrate exposure following carpet flea treatment is not associated with significant dermal absorption. *J. Expo. Anal. Environ. Epidemiol.* 6: 279-288.

**Krishna, K. 1969.** Introduction, pp 1-17 *in* Biology of termites. Vol. 1. (Krishna, K. and F. Weesner, eds.) Academic Press, New York, NY.

**Lee, K.E., and T.C. Wood. 1971.** Termites and soils. Academic Press, New York, NY.

**Light, S.F. 1934.** The constitution and development of the termite colony pp 22-41. *In* Termites and termite control. (Kofoid, C.A. ed.) University of California Press, Berkeley, CA.

**Lloyd, J.D., Schoeman, M.W., and R. Stanley. 1999.** Remedial timber treatment with borates. Paper prepared for the 3rd Int'l. Conf. on Urban Pests. Czech University of Agriculture, Prague.

**Matsumura, F., Coppel, H.C., and A. Tai. 1968.** Isolation and identification of termite trail-following pheromone. *Nature (Lond.)* 219: 963-964.

**McMahan E.A. 1969.** Feeding relationships and radioistope techniques. *In* Biology of termites. Vol. 1. (Krishna, K. & F. Weesner, Eds.) Academic Press, NewYork & London.

**McNamara, W.S. 1990.** Historical uses of diffusible wood preservatives in North America. Proceedings of the First International Conference on Wood Protection with Diffusible Preservatives. Nashville, TN. pp.19-21.

**Murphy, R.J. 1990.** Historical perspective in Europe. Proceedings of the First International Conference on Wood Protection with Diffusible Preservatives. Nashville, TN. pp 9-13.

**Potter, M. 2004.** Termites, pp 216-31 *in* Handbook of pest control: the behavior, life history and control of household pests (Mallis A., and S. Hedges, eds.) 9th ed. GIE Media, Inc. Cleveland, OH.

**Randall, M., and T.C. Doody. 1934.** Poison Dusts, 463-476 *In* Termites and termite control. (Kofoid, C.A. ed.) University of California Press, Berkeley, CA.

**Statistical Analysis Software Institute (SAS) 2002.** Statistical analysis software computer program, version 8.01. Institute, S.A.S., Cary, NC.

**Scheffrahn, R.H., and N.-Y. Su. 1994.** Keys to soldier and winged adult termites (Isoptera) of Florida. *Florida Entomol.* 77(4): 460-474.

**Schoeman, M.W., Lloyd, J.D., and M.J. Manning. 1998.** Movement of borates in a range of timber species at various moisture contents. Paper prepared for the 29th Annual Meeting of the Int'l. Res. Group on Wood Preservation, Maastricht, Netherlands.

**Snyder, T.E. 1948.** Our enemy, the termite. [rev. ed.] Comstock Publ. Co., Inc., Ithaca, NY.

**Stuart, A. 1967.** Social behavior and communication, pp 193-232 *in* Biology of termites. Vol. 1. (Krishna K. and F. Weesner, eds.)Academic Press, NewYork & London.

**Su, N.-Y., and R.H. Scheffrahn. 1991a.** Remedial wood preservative efficacy of Bora-Care against the Formosan subterranean termite and eastern subterranean termite (Isoptera: Rhinotermitidae. The Int'l Res. Group on Wood Preservation. Doc.No. IRG/WP/1504.

**Su, N.-Y., and R.H. Scheffrahn. 1991b.** Laboratory evaluation of disodium octaborate tetrahydrate (Tim-Bor) as a wood preservative or a bait-toxicant against the Formosan and Eastern subterranean termites (Isoptera:Rhinotermitidae) Int'l. Res. Group on Wood Preservation. Doc No.:IRG/WP/1513.

**Su, N.-Y., and R.H. Scheffrahn. 1993.** Laboratory evaluation of two chitin synthesis inhibitors, hexaflumuron and diflubenzuron, as bait toxicants against the Formosan subterranean termite (Isoptera:Rhinotermitidae), *J. Econ. Entomol.* 86: 1453-1457.

**Su, N.-Y., Tokoro, M., and R.H. Scheffrahn. 1994.** Estimating oral toxicity of slow-acting toxicants against subterranean termites (Isoptera: Rhinotermitidae). *J. Econ. Entomol.* 87: 398-401.

**Thorne, B. 1982.** Termite-termite interactions: workers as an agonistic caste. *Psyche* 89: 133-150.

**Thorne, B. 1996.** Termite terminology. *Sociobiol*. 28: 253-263.

**Thorne, B. 1997.** Evolution of eusociality in termites. *Ann. Rev. Ecol. Syst.* 28: 27-54.

**Thorne, B., Breisch, N.L., and J.F.A. Traniello, 1997.** Incipient colony development in the subterranean termite Reticulitermes flavipes (Isoptera:Rhinotermitidae). *Sociobiol*. 30(2): 145-159.

**Thorne, B., Traniello, J.F.A, Adams, E.S., and M. Bulmer, 1999.** Reproductive dynamics and colony structure of subterranean termites of the genus *Reticulitermes* (Isoptera: Rhinotermitidae): a review of the evidence from behavioral, ecological, and genetic studies. *Ethol. Ecol. Evol.* 11: 149-169.

**Tokoro, M., and N.-Y. Su. 1993.** Oral toxicity of Tim-bor®, Bora-Care™, boric acid and ethylene glycol against the Formosan subterranean termite and the eastern subterranean termite. Int'l Res. Group on Wood Preservation. Doc. No. IRG/WP/93-10045.

**Tokoro, M., Takahashi, M., and R. Yamaoka. 1994.** Dodecatrien-1-ol: a minor component of trail pheromone of termite *Coptotermes formosanus* Shiraki, *J. Chem. Ecol.* 20: 199-215.

**Webb, D. 1999.** Creosote, its use as a wood preservative in the railroad industry with environmental considerations. Railway Tie Association Research and Development Committee. Fayetteville, GA.12 pp.

**Weesner, F. 1965.** Termites of the US: a handbook. National Pest Control Association. Elizabeth, NY.

**Williams, L.H. 1990.** Potential benefits of diffusible preservatives for wood protection: an emphasis on building protection. Proceedings of the First International Conference on Wood Protection with Diffusible Preservatives. Nashville, TN. pp.29-34.

**Williams, L.H., and T.L.Amburgey. 1987.** Integrated protection against lyctid beetle infestations. IV. Resistance of boron-treated wood (*Virola spp*) to insect and fungal attack. *Forest Prod. J.* 37(2): 10-17.

**Williams, L.H., and M. Mitchoff. 1990.** Termite feeding on borate-treated wood after 30 months' exposure to 145 inches of rainfall. USDA Forest Serv., Southern Forest Expt. Sta, New Orleans, LA.

**Wilson, E.O. 1971.** The insect societies. Belknap press of Harvard University Press, Cambridge, MA.

## BIOGRAPHICAL SKETCH

Colin Dolan Hickey was born on March 13, 1980, to Charles and Janice Hickey. He has one older brother, Michael Hickey. Colin was born and raised in Newton, MA. After graduating from Newton South High School in 1998, Colin attended Gettysburg College from fall of 1998 until the spring of 2000, at which time he transferred to Providence College in Rhode Island, to earn a Bachelor of Science in December of 2002.

Between spring and fall semesters at Providence College, Colin worked at the State Laboratory Institute in Jamaica Plain, MA, for the Massachusetts Department of Public Health as a laboratory technician tasked with the surveillance of mosquito populations. Mosquitoes captured Colin's interest in entomology and he applied to the University of Florida to work on a graduate degree. Upon acceptance, Colin moved to Gainesville, FL, where he earned a Master of Science degree from the University of Florida researching subterranean termites.