

Laboratory Evaluation of High Temperatures to Control *Cryptotermes brevis* (Isoptera: Kalotermitidae)

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ABSTRACT Rates of thermal increase as low as 0.04°C/min were measured in large wooden members during high-temperature termite control treatments. It was hypothesized that slow rates of thermal increase might promote termite acclimation to high-treatment temperatures. However, in laboratory studies, 46 and 49°C core temperatures were 100% lethal to *Cryptotermes brevis* (Walker) nymphs in wooden blocks (13.5 by 13.5 cm) in both 30- and 60-min exposures, whereas equal durations at 46°C were between 30 and 70% effective in 8.5 by 8.5-cm blocks. Rates of temperature increase were slower in larger blocks and were correlated positively with observed thermal tolerance. The negative effect of low rates of increase may be the result of the cumulative effects of sublethal stresses. It was concluded that the commercially recommended target wood-core temperature of 54.4°C is sufficient for controlling *C. brevis* in large timbers. However, priority should be given to achieving lethal temperatures in worst-case areas as opposed to increasing treatment temperatures because higher temperatures may not result in better control and may increase the risk of heat damage to property within the treated structure.

KEY WORDS *Cryptotermes brevis*, temperature, thermotolerance, termite control, drywood termite

THE USE OF heat to control drywood termites is not new to Hawaii or to pest control in general. Ehrhorn (1934) reported that drywood termites in railway coaches on the island of Oahu were controlled effectively with a 48-h exposure to 65.5°C. Randall and Doody (1934) reported that temperatures from 51.5 to 52°C killed *Incisitermes minor* (Hagen), whereas Horner and Bowe (1934) reported that temperatures from 50 to 65°C were effective in controlling drywood termites in infested lumber <5.1 cm thick.

Forbes and Ebeling (1987) developed a high-temperature process for drywood termite control and suggested the thermal end point of 49°C over a 30-min duration for controlling *I. minor*. This heat-control process involves defining and isolating the area of infestation and then raising the ambient air temperature to increase the temperature of the infested wood to the suggested end point and duration. This recommendation has now been increased to 54.4°C for 1 h (Ebeling 1997).

Lewis and Haverty (1996) evaluated high-temperature control by using simulated infestations of *I. minor* in a specially fabricated structure. Their results demonstrated that 1-h exposures to 50°C were 96 and 98% effective at 3 d and 3 wk after treatment, respectively. The lack of 100% control was caused by a reduction in efficacy in simulated infested wood in the subarea of the structure that was in contact with the concrete foundation. Because concrete can serve as a heat sink, wooden members in contact with foundation slabs are difficult to heat. In addition, high am-

bient relative humidity in substructures, compared with the upper portion of the structure, can result in a high wood-moisture content that could decrease the heating rate. Rust and Reiersen (1997) observed as much as a 10°C difference in core temperatures when 8.3-cm Douglas-fir cubes with wood-moisture contents between 2.6 and 19.2% were heated for 1 h at 49.8°C.

The drywood termite *Cryptotermes brevis* (Walker) has a worldwide distribution and is a pest wherever it is found (Gay 1969). It is also well-known for its desiccation tolerance (Minnick et al. 1973; Steward 1981, 1982; Williams 1977). Scheffrahn et al. (1997) investigated the effects of rate of temperature increase and relative humidity on the thermal tolerance of *C. brevis*. They achieved complete mortality of *C. brevis* nymphs at temperatures as low as 48°C and did not observe any overall effects of thermal rate of increase or relative humidity on mortality. The lowest rate of increase they used was 0.5°C/min. However, field observations (Woodrow 1997) indicate that heating rates during commercial heat treatments are frequently lower than 0.5°C/min and can sometimes be as low as 0.04°C/min.

Acclimation to high-temperature extremes has been observed in a number of insects (Cohet et al. 1980, Berger and Woodward 1983, Dean and Atkinson 1983, Kimura and Beppu 1993, Reisen 1995). Within the Isoptera, Mitchell et al. (1993) found that previous thermal history affected the high-temperature tolerance of *Hodotermes mossambicus* (Hagen). It also has

been suggested that slow rates of temperature increase may allow enough time for the synthesis of stress proteins that could contribute to high-temperature tolerance (Lindquist 1986). We hypothesized that thermal tolerance of *C. brevis* would increase with increasing acclimation temperature and that a steep thermal gradient would have a negative effect on thermal tolerance because higher rates would allow less time for acclimation. Thus, typical rates of $<0.5^{\circ}\text{C}/\text{min}$ could allow for a greater degree of acclimation in commercial high-temperature control sessions (Woodrow and Grace 1997). The goal of this study was to evaluate the effects of various rates of temperature increase on the efficacy of high-temperature control of *C. brevis*.

Materials and Methods

Cryptotermes brevis nymphs were extracted from infested lumber collected from a warehouse at the State of Hawaii, Foreign Trade Zone No. 9, Honolulu, HI. Lumber was carefully split using a hand ax and hammer and the termites were placed into plastic containers with hardwood tongue depressors as a food source. The plastic containers were placed in an unlighted incubator (Precision model 815, Precision Scientific, Chicago, IL) set to $28 \pm 0.5^{\circ}\text{C}$ for at least 1 wk before to testing. Ten 3rd instars or older nymphs (pseudoworkers) as determined by size were obtained from the laboratory colonies and placed into wooden test blocks of various sizes.

The blocks were constructed from untreated 8.5 by 8.5-cm (nominal 4 by 4 in) and 13.5 by 13.5-cm (nominal 6 by 6 in) Douglas-fir, (*Pseudotsuga menziesii* (Mirb.) Franco, stock. Two lengths of each stock size were tested, and the following test block volumes were used: 0.614 liter (8.5 by 8.5 by 8.5 cm), 1.84 liter (8.5 by 8.5 by 25.4 cm), 2.46 liter (13.5 by 13.5 by 13.5 cm), and 4.65 liter (13.5 by 13.5 by 25.4 cm). Three replicates of each block size were prepared. Each block was cut to the appropriate length and then cut into 2 sections (one 2 cm longer than the other) by using a band saw. An 8-cm³ chamber (2 by 2 by 2 cm) was chiseled out of the center of the upper surface of the larger (bottom) block (Fig. 1). Polyethylene foam weather stripping (0.48 cm thick) was used to form an airtight seal between the blocks. In each block, two 3.6-mm-diameter holes were drilled for thermocouple insertion into the test chamber; 1 hole was in the center of the top block, and the 2nd hole at a depth of 1 cm into the top block (Fig. 1). Once the termites were placed into the central chamber, the blocks were combined and held together with 27.94 cm cable ties (GB Electrical, Milwaukee, WI), attached in series (number varied from 2 to 4 according to block size). Next, 24-gauge type-t thermocouples were inserted into the predrilled holes, sealed in place with pieces (2 by 2 cm) of duct tape, and a 4th thermocouple was attached to the outside of the block to monitor the air temperature inside the oven.

The test blocks were placed upright and singly on the bottom shelf of an oven (Salvis Thermocenter Top

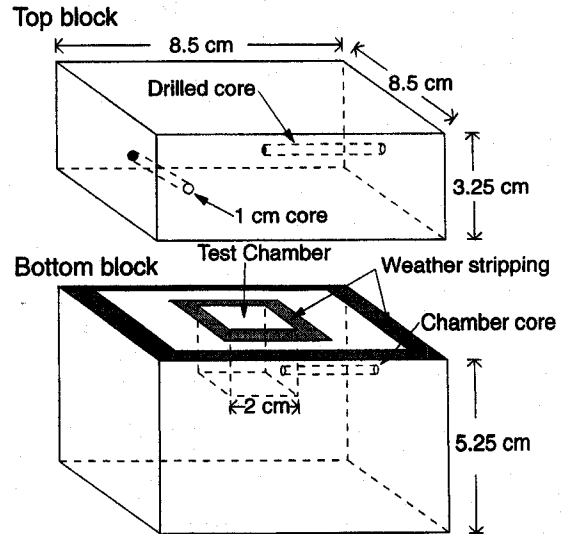


Fig. 1. The 0.614-liter test block used to hold *C. brevis* nymphs during laboratory thermal treatments.

programmable oven, Cole-Parmer, Vernon Hills, IL) and the temperatures were monitored and recorded with a data logger (Omnidata EL925-128, Omnidata, Logan, UT). The oven was programmed to increase the internal air temperature at $5^{\circ}\text{C}/\text{min}$ ($\pm 1.0^{\circ}\text{C}$ in space; $\pm 0.1^{\circ}\text{C}$ in time) up to 70°C . This rate was maintained until temperatures of 30 (control), 46, and 49°C were reached within the test chamber of the block (30°C was chosen as the control temperature because it was the lowest that could be achieved in the oven). The 1st program was stopped and another started to maintain the treatment temperatures. These temperatures were maintained for either 30 or 60 min and temperature-time treatments were completed in 3 separate replicates for each block size. Mortality was assessed immediately after treatment and then at 1 d, 1 wk, and 1 mo after treatment to determine latent effects of heat treatment. The treatment regime also was performed for blocks without termites to a maximum temperature of 49°C to measure the heating rates of the various block sizes. Rates were calculated using least-squared lines plotted using linear regression (SAS Institute 1985).

Before hypothesis testing, model residuals were analyzed to determine if model assumptions were met. Temperature rates were subjected to analysis of variance (ANOVA) (SAS Institute 1985) to test the significance of block volume and probe location (test chamber, cover block core, and 1-cm-deep core) on rate of temperature increase in added-last *F* tests. Mortality data from the blocks were analyzed using ANOVA to test the significance of the following factors: volume (0.614, 1.84, 2.46, and 4.65 liter), treatment duration (30 and 60 min), temperature (30°C [control], 46°C , and 49°C), and posttreatment duration (immediately after exposure, 1 d, 1 wk, and 1 mo) in added-last *F* tests. A Tukey-Kramer honestly sig-

Table 1. Rates of temperature increase in test chambers and mean times required to reach 49°C within various-sized Douglas-fir blocks during high temperature exposures

| Block size, liter | Location | Mean rate, °C/min ^a | Time to 49°C, min ^b |
|-------------------|--------------|--------------------------------|--------------------------------|
| 0.614 | Test chamber | 0.745 ± 0.025a | 32.2 |
| 1.84 | | 0.449 ± 0.016b | 53.4 |
| 2.46 | | 0.232 ± 0.003c | 103.4 |
| 4.65 | Drilled core | 0.156 ± 0.009c | 153.8 |
| 0.614 | | 0.855 ± 0.031a | 28.1 |
| 1.84 | | 0.481 ± 0.015b | 49.9 |
| 2.46 | | 0.249 ± 0.008c | 96.4 |
| 4.65 | | 0.173 ± 0.011c | 138.7 |

^a Mean ± SEM; like letters denote least-square means that were not significantly different ($P = 0.05$ Tukey-Kramer HSD test).

^b Time required for the temperature to reach 49°C from a starting temperature of 25°C.

nificant difference (HSD) test was used to group least-squares of percentage mortality (SAS Institute 1985).

Results and Discussion

An overall model containing probe location, block size, and their interaction was highly significant ($F = 205.60$; $df = 11, 24$; $P < 0.0001$). Rate of temperature increase was highly correlated with wood-block volume and location of the probe ($r^2 = 0.989$). As expected, larger blocks had slower rates of increase ($F = 700.57$; $df = 3, 24$; $P < 0.0001$) (Table 1). The slowest rate of 0.156°C/min was observed within the chamber of 4.65-liter blocks, whereas the 0.614-liter blocks had a mean rate of increase of 0.745°C/min.

The difference between the time required to reach 49°C at the 2 locations in the 4.65-liter blocks was 15.1 min, and at the slower rate of 0.156°C/min the temperature differential would be 2.35°C (0.156°C/min × 15.1 min) (Table 1). In the 0.614-, 1.84-, and 2.46-liter blocks, the temperature differences between the drilled cores and test chambers were 3.05, 1.57, and 1.63°C, respectively. The location of probes also had significant effects on observed temperature rates within blocks. The rates of increase were different among the 3 locations across the blocks ($F = 52.75$; $df = 2, 24$; $P < 0.0001$) (Table 1). Mean rates of increase for the different probe locations were 0.396, 0.440, and 0.542°C/min for the test chamber, cover block core, and the 1-cm-deep core, respectively. These results suggest that the drilled-core thermocouple readings overestimated the temperature of the test chambers within the blocks. These differences could have been the result of the thermocouples in the core holes being in contact with wood whereas those in the chamber were in contact with air. Air has a much lower conductivity ($\approx 0.06 \text{ kcal} \cdot \text{cm} \cdot \text{cm}^{-2} \cdot \text{s}^{-1} \cdot \text{C}^{-1}$) as compared with wood ($\approx 4 \text{ kcal} \cdot \text{cm} \cdot \text{cm}^{-2} \cdot \text{s}^{-1} \cdot \text{C}^{-1}$).

Immediately after exposure to the thermal treatment, a number of termites in the 0.614-liter and 1.84-liter blocks treated at 46°C and in the 4.65-liter blocks treated at 49°C were knocked down but recovered within 24 h (Table 2). After initial knockdown, the percentage of mortality in the 46°C treatment stabilized, whereas all individuals treated at 49°C gradually

Table 2. Mean mortality of *C. brevis* nymphs immediately after treatment and 1 d, 1 wk, and 1 mo after extended exposures to high temperatures within 4 sizes of Douglas-fir blocks

| Temp; °C | Block size, liter | Mean posttreatment mortality ^{a,b} (n = 6) | | | |
|----------|-------------------|---|--------|--------|---------|
| | | Time 0 | 24 h | 1 wk | 1 mo |
| 30° | 0.614 | 0a | 0a | 10.0a | 13.3a |
| | 1.84 | 0a | 0a | 16.7a | 36.7abc |
| | 2.46 | 5.00a | 5.00ab | 5.00a | 5.00a |
| | 4.65 | 0a | 2.50ab | 5.00a | 5.00a |
| 46 | 0.614 | 55.0b | 25.0b | 30.0ab | 33.3ab |
| | 1.84 | 88.3c | 68.3c | 68.3bc | 70.0cd |
| | 2.46 | 100c | 100d | 100c | 100d |
| | 4.65 | 100c | 100d | 100c | 100d |
| 49 | 0.614 | 100c | 100d | 100c | 100d |
| | 1.84 | 100c | 100d | 100c | 100d |
| | 2.46 | 100c | 100d | 100c | 100d |
| | 4.65 | 100c | 95.0cd | 95.0c | 100d |

^a Exposure time (30 and 60 min) did not significantly affect percentage of mortality and was pooled in the model.

^b Like letters denote least-square means that were not significantly different ($P = 0.05$ Tukey, Kramer HSD test).

^c Control group.

expired during the 1 mo after treatment. As control mortality also increased slightly over this same period, it is possible that an overall stress effect, as opposed to a treatment effect, could have led to delayed mortality. This observation of delayed mortality, however, is consistent with Scheffrahn et al. (1997) and Lewis and Haverty (1996) who reported that mortality resulting from heat treatment was not fully expressed until 1 wk and 1 mo after treatment, respectively.

Overall, 46 and the 49°C treatments were more effective than the 30°C controls. At 49°C, >95% control was observed in 30-min exposures and 100% control in 60-min exposures (Table 2). Treatment temperature and block size had significant effects on posttreatment mortality ($F = 21.47$ – 33.32 ; $df = 11, 48$; $P < 0.0001$), whereas the duration of temperature exposure was not significant. The ANOVA model containing treatment temperature, block size, and their interaction was highly significant ($F = 24.32$; $df = 11, 48$; $P < 0.0001$). Treatment temperature and block size significantly effected mortality (added-last F tests; $F = 8.11$ and 98.14 ; $df = 3, 48$; $P = 0.0079$ and 0.0001 , respectively) as did the interaction between size and temperature ($F = 7.82$; $df = 6, 48$; $P < 0.0001$) at 1 mo after treatment. The Tukey-Kramer HSD test indicated that the mortality in 46 and the 49°C treatments was greater than the 30°C controls, except for the 0.614-liter blocks treated at 46°C (Table 2).

The significant interaction between block size and treatment temperature was due to the variation among block sizes treated at 46°C (Table 2). Contrary to our initial hypothesis, lower rates of thermal increase in large wooden blocks had a negative effect on termite survival. Slower rates and greater mortality were observed at 46°C in the 0.614- and 1.84-liter blocks than in the 2.46- and 4.65-liter blocks. After 1 mo, mean mortality was 33.3 and 70% in 0.614- and 1.84-liter blocks, respectively, and 100% in and both the 2.46- and 4.65-liter blocks.

The negative effect of low rates of temperature increase on the percentage of mortality negated our original hypothesis of acclimation. However, it poses some interesting questions relative to the physiology of heat stress inside the wooden chambers. Decreased thermotolerance at lower rates could be partly attributable to desiccation because nymphs in larger blocks spent the most time under potentially stressful conditions. This hypothesis is supported by Rust et al. (1979) who found that low humidity had a negative effect on thermotolerance in the drywood termites *I. minor* and *Incisitermes fruticavus* Rust during exposures >4 h.

Our results revealed that heating rates were higher in drilled wood cores than within the inner test chamber. For this reason, it may be important in commercial treatments to consider temperatures in drywood termite galleries in addition to drilled cores. Large temperature differentials also can occur between similar core locations even under controlled conditions. Proper placement of thermal probes is essential to achieving lethal temperatures in areas that are particularly difficult to heat because these areas could serve as refuges for termites during thermal treatments (Cabrera and Rust 1996).

Although our initial hypothesis of acclimation at low temperature was rejected, a more interesting result was achieved. Low rates of temperature increase appear to be more detrimental to survival at high temperatures than do high rates. This indicates that higher ambient treatment temperatures resulting in rapid rates of temperature increase may not be advisable as a means of increasing the efficacy of heat treatments. Rather, given the lack of acclimation at low rates of temperature increase, it may be advisable to decrease ambient treatment temperatures and increase the length of the treatment, which also would reduce the possibility of thermal damage to property. The recent increase in the commercial treatment recommendation from 49°C for 30 min (Ebeling 1994) to 54.4°C for 1 h (Ebeling 1997) may not be entirely warranted for *C. brevis* given the observations on thermal rate and the effectiveness of 49°C in our study. The efficacy of high-temperature control of drywood termites can be ensured while minimizing the potential for thermal damage if care is taken to locate drywood termite infestations and place thermocouples in worst-case locations without necessarily resorting to higher treatment temperatures.

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