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Abid Farid
Department of Agricultural Sciences, the University of Haripur, Khyber Pakhtunkhwa, Pakistan.

Maid Zaman
Department of Agricultural Sciences, the University of Haripur, Khyber Pakhtunkhwa, Pakistan.

Muhammad Saeed
Department of Agricultural Sciences, the University of Haripur, Khyber Pakhtunkhwa, Pakistan.

Maazullah Khan
Nuclear Institute for Food & Agriculture (NIFA) Peshawar, Pakistan.

Tahir Bad shah
Department of Agricultural Sciences, the University of Haripur, Khyber Pakhtunkhwa, Pakistan

Correspondence:
Abid Farid
Department of Agricultural Sciences, the University of Haripur, Khyber Pakhtunkhwa, Pakistan.

Evaluation of boric acid as a slow-acting toxicant against subterranean termites (*Heterotermes* and *Odontotermes*)

Abid Farid, Maid Zaman, Muhammad Saeed, Maazullah Khan and Tahir Bad shah

Abstract
Various concentrations (100, 500, 1000, 5000 and 10000 ppm) of boric acid were tested in laboratory to check its potential as a slow-acting toxicant against subterranean termite, *Heterotermes indicola* (Rhinotermitidae: Isoptera). Under the tested concentrations, ELT 50 (Effective Lethal Dose to kill 50% of the exposed termites) values ranged from 2.7 to 5.4 days while ELT 90 were 4.5 and 9.1 days for the lowest and highest concentrations used in the lab trial. Choice feeding test showed that bait matrix treated with all the above concentrations was acceptable to termites. At a concentration of 10000 ppm, boric acid resulted in 91% mortality within two weeks when both the treated and untreated substrates were offered to *H. indicola*. Field testing of boric acid bait showed that it was readily fed upon by *Odontotermes* sp. while *H. indicola* avoided the bait after initial feeding exposure. However, it led to significant population suppression for both the genera of subterranean termites.

Keywords: Boric acid, slow-acting toxicant, *Heterotermes indicola*, *Odontotermes* sp

1. Introduction
Termite is a serious pest of agricultural crops and buildings in Pakistan inflicting huge losses throughout the country. According to surveys conducted in Khyber Pakhtunkhwa province, 12-100% termite infestation has been observed in orchards, cereals, and tobacco and sugar crops especially in areas with low water table [1]. Fifty termite species have been reported from Pakistan. Out of these, termites belonging to *Heterotermes*, *Coptotermes* (Rhinotermitidae); *Odontotermes*, *Microtermes* (Termitidae) cause economic damage to crops, buildings and orchards in Pakistan [2]. *Heterotermes indicola* is one of the economically important species damaging crops, building and orchards in Pakistan. Most of the termites causing economic damage (including *H. indicola*) in Pakistan are subterranean in nature and live as highly organized colonies comprising of nesting system of earthen tunnels and satellites present underground and may spread over an area of more than 100 m^2 [1].

Use of chemical barriers is not very effective as the termites seal off their tunnels, divert to other feeding sites and continue the damage in the nearby areas [3]. Successful management of subterranean termite populations requires the application of control measures that can have a major impact on reducing colony population. Use of slow-acting toxicant bait is such a method that uses very little quantities of a toxicant which are applied on established foraging points. Lethal doses of these slow-acting toxicants are picked up by the termite workers and distributed within the colonies. The toxicant slowly poisons the termites and eventually reduces or eliminates the entire colony [4]. Mirex impregnated in wooden blocks was used for the first time to suppress populations of subterranean termites [5]. Since then, different metabolic inhibitors and growth regulators have been tried as bait toxicants.

Boric acid is widely used for controlling household pests and boron-based compounds are used as wood preservative treatments to inhibit termites, beetles and some decay fungi [6]. Usta [7] found that when boric acid-treated medium-density fiberboards were offered to *Coptotermes formosanus* Shiraki, it caused delayed mortality in the termites. No mortality was observed after the first week while the mortality ranged from 20-100% after 2-3 weeks at a concentration range of 1%. The present studies were conducted to evaluate the potential of using boric acid as a slow-acting toxicant against subterranean termites both in orchards and buildings.

2. Materials and Methods

The present work was carried out at Nuclear Institute for Food and Agriculture (NIFA) Peshawar, Khyber Pakhtunkhwa Pakistan, during the year 2013-2014.

2.1 Insect Collection

The termites were collected by stake-survey and trapping method followed by Farid *et al.* [7] at Nuclear Institute for Food and Agriculture (NIFA), Khyber Pakhtunkhwa (KPK) Pakistan.

2.2. Characterization of Boric Acid as a Slow-Acting Toxicant

Fifty termite workers (plus five soldiers) were placed in Petri dishes (9.0 cm diameter by 1.5 cm high) provisioned with two layers of 9 cm circular blotting papers (Millat paper art, Karachi, Pakistan) treated with different concentrations of aqueous solution of boric acid (Merck) to yield 100. 500, 1000, 5000 or 10,000 ppm (w/w). The treated blotting paper was dried at room temperature and moistened with 2 ml of deionized water before using in the experiment. All treatments were replicated four times. Dead or moribund (when test individual could not walk after probing) workers were counted and removed daily until all the termites were found dead. ELT 50 and ELT 90 (the time required for a fixed dosage to kill 50 or 90% of the test insects) were estimated using days (within each dose) as independent variable in probit analysis (Using SPSS 16).

2.3 Free-Choice Feeding Studies

Free-choice studies were conducted to evaluate acceptability of different concentrations of boric acid for feeding by *H. indicola*. The experimental arena used consisted of plastic petri dishes (9 cm diameter by 1.5 cm high). Two strips (3 cm × 2 cm) of blotting papers were placed 3 cm apart horizontally at bottom of the dish and covered with 25 g of sand moistened with deionized water (20% w/v). One piece of blotting paper was treated by dipping in boric acid solution to get required concentration of 100, 500, 1000, 5000 or 10,000 ppm (wt of ai /wt of blotting paper). The second strip was dipped in deionized water only. Dry weights of the paper strips were determined before using them in the trial. The bottoms of the petri dishes were already brushed with sandpaper to provide traction for termites. Each concentration was considered as a treatment and was replicated eight times. Fifty workers and three soldiers of *H. indicola* were introduced into each unit. All the units were kept in round glass chambers (30 cm diameter, 25 cm high) with an air-tight lid, having water at the bottom to maintain 90 ± 5% RH, and kept in the laboratory at 25 ± 2 °C. The units were disassembled after two weeks and data were recorded on the number of termites still alive. The blotting paper strips were cleaned off the sand particles and their dry weights were recorded. Consumption was determined by subtracting the initial dry weight of the strip from the final dry weight. Paired t-test was used to compare the consumption of treated and untreated strip within each unit [9].

2.4. Testing Boric Acid Bait against *Heterotermes Indicola* in the Field

Mark-release-recapture method Crosland and Su [10] was used for colony delineation in a building which was severely infested by *H. indicola*. Major infestation was observed in a hall measuring 15× 10 m. Termite damage was observed in four wooden closets along with earthen galleries. Active galleries (the ones showing termite activity) were traced to the points of their contact with the ground. A bundle of five wooden slices (*Populus* sp., 15 cm high by 8 cm wide by 1 cm thick) wrapped in a blotting paper and held together by a rubber band was placed on the point of termites' entry into the ground.

Wooden bundles were collected and replaced with the newer ones after two weeks. In the area of the highest termite activity, the bundel was replaced with a new bundle with the middle wooden slice having a 5 mm hole across its length for releasing marked termites. The termites collected from this foraging point were force-fed on blotting paper (Millat paper art, Karachi) soaked in aqueous solution of Nile blue to yield a 0.2% concentration (w/w) of the dye in the paper in laboratory. Well-stained and active workers were counted and released back to the point of their collection. To release the stained termites, a glass funnel was placed over the hole already present in the middle slice of the wooden bundle and dyed termites (present in the glass Petri dish) were poured into it and watched to make sure that all the termites disappeared into the ground. All the foraging points were again inspected after two weeks and the wooden bundles were collected. The foraging points having a capture of dyed termites were considered to belong to the same colony. The results of mark-release-recapture studies showed that all the ten foraging points were interconnected and hence formed a single colony spread over an area of 45 sq. m. After delineation, all the foraging points were again replaced with fresh slice bundles.

Boric acid bait was prepared by impregnating pieces of blotting paper, 15 × 8 cm (3.65 g) each, with 1000 ppm aqueous solution of boric acid. Four of these treated blotting paper pieces were sandwiched in three poplar slices (15 × 8×2 cm) and held together with a rubber band. These baited traps were replaced with two wooden slice-bundles showing the highest termite activity. Termite activity was checked at the treated spots as well as on the other foraging points by collecting the bundles to estimate the number of termites within each bundle after four weeks. T-test was used to compare consumption and worker population with the help of SPSS 16.

2.5. Testing Boric Acid Bait against *Odontotermes* Sp.

A house lawn infested with *Odontotermes* sp. was selected for testing boric acid bait. Three active forging points were established using stake-survey method. Monitoring stations (as mentioned earlier) containing pre-weighed poplar slices were placed at these foraging points. Data on wood consumption were recorded after one month. All the monitoring stations were then replaced with boric acid bait. Baited traps were prepared as mentioned earlier the data on bait consumption and termite activity was recorded after one month by checking the bait on all the foraging points. The points were replaced with monitoring stations to check the termite activity. Data were recorded on the number of termite workers present in each trap and the amount of wood+ paper consumed. T-test was used to compare consumption and worker population with the help of SPSS 16.

3. Results and Discussions

Probit analysis (Table 1) indicated that estimated time to kill half of the exposed workers was around 5 days for 100 ppm and 2.5 days for 10000 ppm. Estimated time to kill 90% of the termite worker (ELT 90) exposed to different concentrations of boric acid was around 8–9 days at the lowest concentration used (100 ppm) while it was 4–5 days at a concentration of 10000 ppm. Under a choice condition, when boric acid was offered in the presence of untreated food in sand, the termites fed equally on both the treated and untreated substrates for all the concentrations indicated by a non-significant value of paired t-test (p>0.05) for consumption rate of treated and untreated food (Table 2). This shows that termites were unable to distinguish between the treated and untreated substrates at all the concentrations used in our experiment. Under a choice condition, *H. indicola* did not suffer from a significant mortality unless the highest concentration of boric acid (10000 ppm) was used wherein 91% mortality was observed. Although *H. indicola* was not deterred by any concentration of boric acid used in our experiments but concentration lesser than 10000 ppm did not cause a significant mortality (> 90%) indicating that this concentration can be tested in the field.

Boric acid has traditionally been used as wood preservative for a long time. At higher concentrations, it deters termite feeding but it has been reported to have slow acting properties also. Usta [7] found that when medium density fiberboard treated with 1% boric acid were offered to Co*ptotermes formosanus* Shiraki, delayed mortality was caused in the termites. No mortality was observed after the first week while the mortality ranged from 20 to 100% within 2-3 weeks. Su *et al.* [11] estimated oral and contact toxicity of boric acid against the subterranean termites *Coptotermes* and *Reticulitermes*. They observed that ELT 90 for *Reticulitermes* ranged between 7-12 days at different concentrations (1000-2000 ug / g) of boric acid used although it failed to produce >90% mortality in *Coptotermes*. Kard [6] conducted laboratory bioassays on boric Acid mixed with soil and caused significant subterranean termite mortality. Termites were not repelled by boric acid and continued tunneling activity even at higher concentrations of boric acid (4%).

Table 1: Estimated lethal time (days) required for 50 and 90% mortality (along with 95% confidence interval) of *H.* indicola after exposure to various concentrations of boric acid

Dose ppm	ELT 50 (days)	95% CI	ELT 90 (days)	95% CI
100	5.4	5.2-5.6	8.7	8.3-9.1
500	5.0	4.6-5.3	7.5	7.0-8.2
1000	5.2	4.8-5.5	8.0	7.4-8.8
5000	3.1	3.0-3.2	4.5	4.3-4.7
10000	2.7	2.4-2.9	4.5	4.1-4.9

Table 2: Difference in blotting paper consumption by *H. indicola* between untreated blotting paper and those treated with different concentrations of boric acid after two weeks

Dose ppm		Consumption mean ±SE	t-value (p value)	Percent mortality ± SE
100	Control	0.20±0.052	-0.599(0.591)	7.50 +0.64 a
	Treated	0.27±0.071		
500	Control	0.22±0.051	-0.568(0.610)	18.0+1.22 b
	Treated	0.27±0.035		
1000	Control	0.27±0.028	1.303(0.284)	14.75±1.84 ab
	Treated	0.18±0.055		
5000	Control	0.26±0.091	0.750(0.508)	57.0±2.6 c
	Treated	0.17±0.034		
10000	Control	0.04±0.024	-2.513(0.128)	91.0±3.0 d
	Treated	0.09±0.013		

Boric acid bait was tested against *Odontotermes* sp., and *H. indicola* (Table 3). The bait when offered as a sandwich in poplar slices was fully consumed by *Odontotermes* sp leading to suppression of the colony. *Odontotermes* sp. is a voracious pest of agriculture, forestry and buildings in Pakistan and causes much more damage than the other subterranean species due to their larger size and higher feeding rates. Application of boric acid bait can be simple and cost effective method for its control. Jones [12] tested a borate based bait in the field at 2500 to 5000 ppm concentration in Arizona and found that it was not repellent and successfully destroyed a colony of the desert subterranean termite, *Heterotermes aureus*.

When boric acid bait was tested against *H. indicola*, relatively lower bait consumption was observed. At the treated spot (where bait was applied), 1.74 g of the bait was consumed which led to an overall decrease in colony population by 60% but the bait application did not lead to a complete elimination or suppression of the colony. After the initial feeding, termites did not come back to the bait station. We found *H. indicola* to be more sensitive to the presence of bait toxicant after the initial feeding experience and did not continue to feed on the bait for a long period of time. A way to deal with this issue is to apply the bait on all the detected foraging points allowing initial feeding exposure of toxicant to a larger proportion of termites. This will lead to greater consumption of the toxicant before the workers learn to avoid it. This initial toxicant consumption can lead to a significant suppression of termite population.

Due to its very low mammalian toxicity, boric acid is considered as a safe chemical. The reported acute oral LD_{50} for boric acid in mice is 3450 mg/kg. Studies in rats report LD_{50} values for boric acid ranging from 2660-5140 mg boric acid/kg body weight depending on the duration of exposure [13, 14]. Boric acid is a low cost and easily available chemical even from the drug stores. Thus due to its effectiveness as a slow-acting toxicant, low toxicity and easy availability, boric acid can be used for suppression of colonies of subterranean termites. We recommend using boric acid impregnated (1000 ppm w/w) blotting papers sandwiched in poplar slices and placed on active foraging points (determined by stake survey) until termite activity is ceased.

Table 3: Field evaluation of boric acid bait against *Odontotermes* sp. and *Heterotermes indicola*

	wood consumption *Odontotermes* (g)	Population *Odontotermes* (Number of workers)	wood consumption *Heterotermes* (g)	Population *Heterotermes* (Number of workers)
Pre- bait	276 a	3240 a	73 a	7920 a
Post-bait	0 b	0 b	17.6 b	3187 b

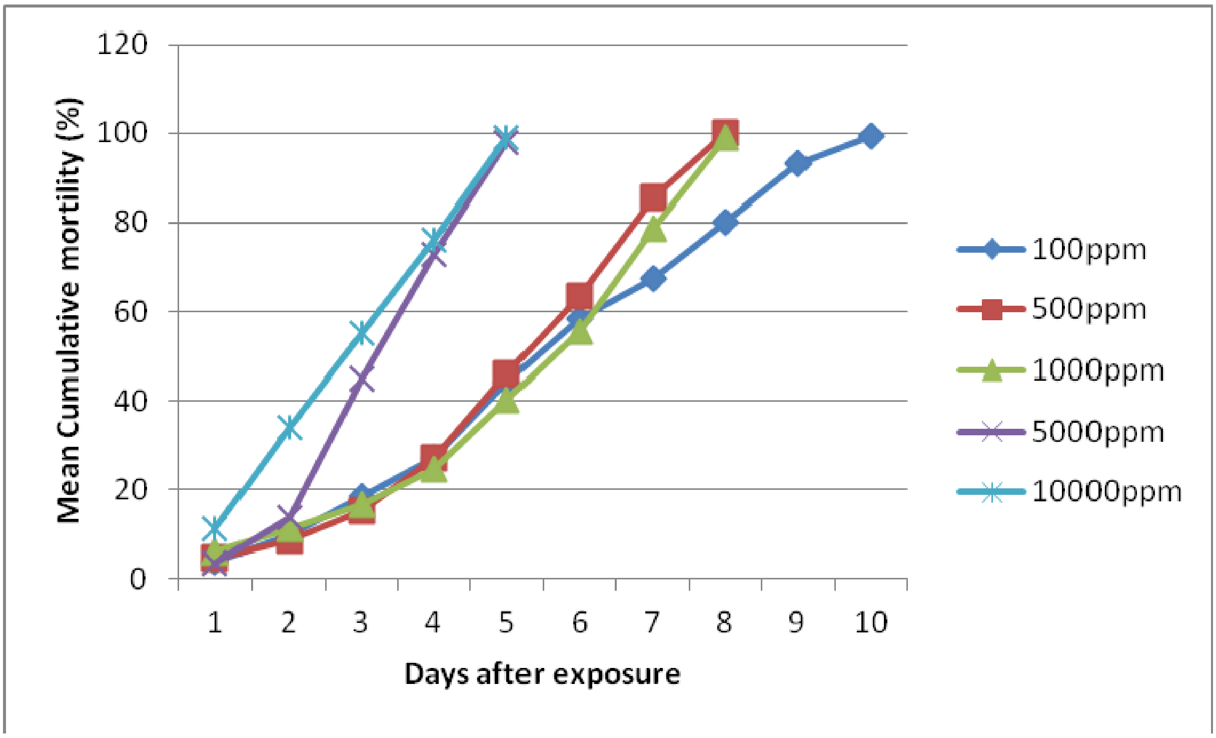

Fig1: Cumulative mortality (adjusted) of *H. indicola* at various intervals after exposure to lower concentrations of boric acid

4. Conclusions and Recommendations

Boric acid can be used as an effective slow-acting toxicant against subterranean termites when used at a concentration of 1000 ppm (w/w) in a blotting paper matrix.

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6. References

1. Salihah Z, Sattar A, Farid A, Shakoori AR. Termites of Pakistan and their control: Zoological Society of Pakistan, University of the Punjab, Lahore, Pakistan, 2012.
2. Chaudhry MI, Ahmed M, Malik KN, Akhtar SM, Arshad M. Termites of Pakistan. Final Technical Report, Pakistan Forest Institute Peshawar - Pakistan. Project No. A17-FS-12, Grant No.FG-Pa-131 (Under Pl-480 Programme of U.S.A).PP.1-70. 1972.
3. Su NY, Tamashiro M, Yates JR, Haverty M. Effect of behavior on the evaluation of insecticide for prevention or remedial control of the Formosan subterranean. Journal of Economic Entomology 1982; 75:188-193.
4. Jones SC. Termite Baits. The Ohio State Extension Fact Sheet, HYG-2092A-03 1991.
5. Esenther GR, Beal: RH. Attractant-mirex bait suppresses
6. Activity of *Reticulitermes* spp. Journal of Economic Entomology 1974; 67:85-88.
7. Kard BM. Detrimental effects of boric acid treated soil against foraging subterranean termites (Isoptera: Rhinotermitidae). Sociobiology 2001; 37:363-378
8. Usta M. Termite resistance of MDF panels treated with various boron compounds. Int J Mol Sci 2009, 2789-2797.
9. Farid A, Haq MM, Ullah M, Sattar A. Potential of fipronil as a feeding toxicant against the subterranean termite Heterotermes indicola (Rhinotermitidae: Isoptera). Philippine Agricultural Scientist 2014; 97:73-78.
10. Su NY, Scheffrahn RH. Laboratory Evaluation of Two Chitin Synthesis Inhibitors, Hexaflumuron and Diflubenzuron, as Bait Toxicants against Formosan and astern Subterranean Termites (Isoptera: Rhinotermitidae). Journal of Economic Entomology 1993; 86:1453-1457.
11. Crosland MWJ, Su NY. Mark-recapture without estimating population sizes: A tool to evaluate termite baits. Bulletin of entomological research 2006; 96:99-103.
12. Su NY, Tokoro M, Scheffrahn RH. Estimating oral toxicity of slow-acting toxicants against subterranean termites (Isoptera: Rhinotermitidae). Journal of Economic Entomology 1994; 87:398-401.
13. Jones SC. Borate baiting systems for subterranean termite (Isoptera: Rhinotermitidae) control. In: Hamel, 128, 1990.
14. Weir RJ, Fisher RS. Toxicologic studies on borax and boric acid. Toxicol. Appl Pharmacol 1972; 23:351-364.
15. Pfeifer CC, Hallman LF, Gersh I. Boric acid ointment: A study of possible intoxication in the treatment of burns J Am Med Assoc 1945; 128:266-274.