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Insecticidal studies on heterotermis indicola wasmann (Isoptera: Heterotermitidae) under laboratory conditions

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Abstract
Insecticidal studies on *Heterotermes indicola* Wasmann (Isoptera: Heterotermitidae) were carried out under laboratory conditions at the Nuclear Institute for Food and Agriculture (NIFA), Peshawar, Pakistan during 2010. Chlorpyrifos, Cadusafos and Agenda each were tested in five concentrations, i.e. 100, 50, 25, 12.5 and 6.25 ppm against *H. indicola* along with a control. Chlorpyrifos yielded highest mortalities of > 80% against *H. indicola* at 100 and 50 ppm after 8 h and all concentrations after 24 and 48 hours. Cadusafos was second in toxicity against *H. indicola* where it caused more than 80% mortalities at 100 ppm after 8h and at 100, 50 and 25 ppm after 24 and 48 hours. Agenda caused no mortalities at all concentrations after 8 h, but it yielded 14 to 33% mortalities after 24 and 48 h. After 15 days, however, toxicity of Chlorpyrifos and Cadusafos against *H. indicola* was reduced with all the concentrations. On the other hand, toxicity of Agenda increased with time, where it ranged from 25-50% after 8 h, 42-65% after 24 h and 80-92% after 48 h with all concentrations after 15 days. Toxicity of all the insecticides increased in a dose dependent manner. Generally, Chlorpyrifos, and Cadusafos showed quick while Agenda resulted slow toxicity against *H. indicola* under laboratory conditions.

Keywords: *H. indicola, insecticides, toxicity, termites*

1. Introduction
Subterranean termite species (Order: Isoptera) are serious pests to agricultural crops. They cause 90-100% damage to sugarcane crop. They also cause 45% damage to maize and 10-12% damage to wheat crop and 80-90% to fruit orchards [1]. *Heterotermes indicola* Wasmann (Isoptera: Heterotermitidae) is a widely distributed termite species of the genus Heterotermes found in different parts of Pakistan [2]. Two species including *H. indicola* and *Coptotermes heimi* have been reported that infest buildings in Peshawar [3].
Termites' control is based mainly on chemicals especially synthetic insecticides [4]. Insecticides are categorized as repellent, toxic and non repellent with delayed toxicity [5]. Repellent insecticides protect structures typically by repelling foraging termites inhibit termite tunneling [6]. The toxic and non repellent insecticides kill termites by contact. With slow acting insecticides, the level of mortality and the speed of kill are dependent on pesticide concentrations [7].
The use of non- repellent insecticides has increased over the last many years, due to the fact that termites cannot detect the treated soil and continue foraging in the soil. These compounds have a delayed mode of action [8, 9]. All insecticides formulations are able to stop termite soil penetration, either by killing or repelling them. The termiticide properties (repellent or non-repellent) may be dependent on the dose of the insecticides used [10]. Keeping in view the importance of *H. indicola* and the damages caused by it to various agricultural crops and buildings, the present study was conducted to determine toxicity of three insecticides in five concentrations under laboratory conditions.

2. Materials and Methods
2.1 *H. indicola* Stock Culture
The present research work was conducted in the Termite Research Laboratory Entomology Division, NIFA Peshawar during 2010. *H. indicola* were collected from the infested orchards and buildings by using NIFA–TERMAP [3]. The foraging points of termites were detected by using stake method as well as visually observing termite's galleries [11].

2.2 Insecticide Formulations and Testing Procedure

The insecticides tested in the experiments were Chlorpyrifos, Cadusafos and Agenda. The tested insecticides were purchased from the local market. Stock solutions of 100 ppm of each of Chlorpyrifos, Cadusafos and Agenda were prepared. Five concentrations, i.e. 100, 50, 25, 12.50 and 6.25 ppm. of each of these insecticides were prepared by serial dilutions of the stock solutions. In control, the blotting paper was treated with distilled water only. Three replicates of each concentration were conducted in the experiments.

2.3 Insecticides Toxicity Test

The required numbers of Petri dishes were washed and sterilized at 121 °C for 20 minutes prior to the experiments. Two pieces of blotting paper were cut at the size of the Petri dish measuring 90 mm×15 mm. The blotting papers were treated with five different concentrations of Chlorpyrifos, Cadusafos and Agenda and placed at the bottom of the Petri dishes. Twenty five termites (twenty workers and five soldiers) were gently released into each Petri dish using camel hair brush. The experiment was replicated three times for each insecticides concentration. In control, the blotting papers were treated with distilled water only. The test individuals were examined after every hour up to 8 h, 24 h and 48 h and then after fifteen days the blotting papers were diluted with distilled water and the same procedure was repeated as mentioned above.

2.4 Statistical Analysis of Data

The experiments were laid out in Completely Randomized Design (CRD) and the data were analyzed using one way ANOVA and means were separated using LSD test at 5% level of significance using STATISTIX 8.1® [12] package.

3. Results and Discussion

3.1 Insecticides Toxicity Test

The results showed significant differences in mortality caused by the three insecticides and its various concentrations (Table 1). Toxicity by Chlorpyrifos and Cadusafos at 100 ppm was significantly higher with 99.0 and 89.33% after 8 h, respectively. At 50, 25, 12.5 and 6.25 ppm, Chlorpyrifos caused significantly higher mortality of 85.44, 78.22, 72.00 and 65.11%, respectively, than the other insecticides. No mortality was recorded with all concentrations of Agenda and control after 8 h treatment.

Table 1: Toxicity of Chlorpyrifos, Cadusafos and Agenda against *H. indicola* after 8, 24 and 48h during 2010

Concentration (ppm)	Mortality (%)											
	8h				24h				48h			
	Chl	Cad	Age	Cont	Chl	Cad	Age	Cont	Chl	Cad	Age	Cont
100	88.00a	89.33a	0.00b	0.00b	94.33a	92.44a	33.11b	0.00c	100.00a	100.00a	33.11b	0.00c
50	85.44a	78.44b	0.00c	0.00c	90.55a	85.44b	26.55c	0.00d	100.00a	95.44b	29.55c	0.00d
25	78.22a	70.22b	0.00c	0.00c	88.00a	80.00b	23.44c	0.00d	95.00a	87.22b	25.33c	0.00d
12.5	72.00a	60.00b	0.00c	0.00c	85.11a	69.22b	21.00c	0.00d	90.00a	78.00b	22.00c	0.00d
6.25	65.11a	41.22b	0.00c	0.00c	80.00a	60.99b	14.33c	0.00d	86.22a	72.22b	20.55c	0.00d

Means within a row followed by different letters are significantly different at $p < 0.05$ (LSD-test)

After 24h of treatment at 100 ppm, Chlorpyrifos (94.33%) and Cadusafos (92.44%) caused significantly higher mortality than agenda (33.11%). Chlorpyrifos at 50, 25, 12.5 and 6.25 ppm yielded significantly higher mortality of 90.55, 88.00, 85.11 and 80.00%, respectively than the other treatments. After 24 h no mortality was observed in control.

Forty eight hours after treatment at 100 ppm, Chlorpyrifos and Cadusafos each gave 100% mortality, which was significantly higher than with agenda and control. Chlorpyrifos yielded significantly higher mortality of 100.00, 95.00, 90.00 and 86.22% at 50, 25, 12.5 and 6.25 ppm, respectively than the other treatments and control. In control no mortality occurred after 48 h. Generally, *H. indicola* mortality increased with increase in concentrations of all the insecticides.

Toxicity of the five insecticides against *H. indicola* after 15 days of treatment showed significantly different results of mortality (Table 2). Among the treatments, chlorpyrifos at 100 ppm yielded significantly higher mortality (53.33%) after 8 h, whereas agenda at 50, 25, 12.5 and 6.25 ppm resulted in significantly higher mortality of 48.00, 36.33, 33.33 and 25.33%, respectively. In control no mortality of the insect were recorded after 8h.

Table 2: Toxicity of Chlorpyrifos, Cadusafos and Agenda against *H. indicola* after 8, 24 and 48h after 15 days during 2010

Concentration (ppm)	Mortality (%)											
	8h				24h				48h			
	Chl	Cad	Age	Cont	Chl	Cad	Age	Cont	Chl	Cad	Age	Cont
100	53.33a	30.11c	50.44b	0.00d	73.77a	25.55c	65.00b	0.00d	80.00b	23.55c	92.55a	0.00d
50	36.00b	27.33c	48.00a	0.00d	62.89a	23.44b	60.22a	0.00c	73.44b	20.22c	88.22a	0.00d
25	26.67b	22.22bc	36.33a	0.00d	48.00b	17.55c	55.11a	0.00d	55.33b	16.44c	84.22a	0.00d
12.5	14.67b	17.11b	33.33a	0.00d	41.11b	15.55c	49.44a	0.00d	50.00b	12.33c	82.33a	0.00d
6.25	5.33c	12.00b	25.33a	0.00d	26.33b	11.10c	42.11a	0.00d	38.22b	10.44c	80.99a	0.00d

Means within a row followed by different letters are significantly different at $p < 0.05$ (LSD-test)

After 24h of treatment at 100 ppm, chlorpyrifos gave significantly higher mortality (73.33%) than the other treatments and control. At 50 ppm, chlorpyrifos (62.89%) and agenda (60.22%) yielded significantly higher mortality than cadusafos and control. Agenda at 25, 12.5 and 6.25 ppm resulted in significantly higher mortality with 55.11, 49.44 and 42.11%, respectively. No mortality occurred in control after 24h.

After 48 h of treatment, cadusafos at all concentrations resulted in significantly higher mortality of 92.55, 88.22, 84.22, 82.33 and 80.99% at 100, 50, 25, 12.5 and 6.25 ppm, respectively. In control no mortality were observed after 48 h. Generally, *H. indicola* mortality increased with increase in concentrations of all the insecticides.

The present results revealed that Chlorpyrifos and Cadusafos caused more than 80% mortality of *H. indicola* at the higher

concentrations and more than 60% mortality at the lower concentrations after 8, 24 and 48 h, which showed its fast acting lethal effect against the test insect. During this time, however, mortality with agenda were nil after 8h and below 35% after 24 and 48 h. After 15 days of treatment, however, the results of toxicity experiments were totally different than achieved earlier. It was found that agenda, except chlorpyrifos at 100 ppm after 8 and 24 h, gave significantly higher mortality of *H. indicola* at all concentrations after 8, 24 and 48h. These present results comparable to those reported by Sheikh *et al*., [2], where they recorded highest mortalities of *H. indicola* at higher concentrations of chlorpyrifos (480 ppm) and cadusafos (100 ppm). The findings of Manzoor *et al*., [13] also supported our results and recorded highest mortality of 97% when *H. indicola* was treated with Bifenthrin, imidacloprid, chlorfenapyr and cadusafos. Agenda (Fipronil) was found to have slow acting effect as it required longer time to show its lethal effect also supported by Manzoor *et al*., [13] and reported fipronil as slow acting toxicant against *H. indicola*. After 15 days, more than 80% mortalities were achieved with all concentrations of agenda after 48 h of treatment. Our findings are in accordance with those of Saljoqi *et al*., [14] wherein they recorded mortality of 90-60% with fipronil after ten days. The work of Iqbal and Saeed [15] also corresponds to our outcomes where slow lethal effect of fipronil was noted. The present results are also at par with those reported by Sattar *et al*., [16]. According to them the slow acting toxicity in *H. indicola* was inversely proportional to insecticide concentrations. As insecticides concentrations decreased in distilled water solution the slow-acting toxicity increased. The most desirable results of 87.30% and 100% mortality were observed after 12 and 15 days, respectively, with 0.000625% concentration of Agenda.

4. Conclusion and Recommendations

Toxicity of Chlorpyrifos and Cadusafos against *H. indicola* was highest at the higher concentrations after 8, 24 and 48 h but toxicity decreased with time. Agenda, on the other hand, showed slow acting lethal effect after 8, 24 and 48 h of treatment, but its toxicity increased with time. Chlorpyrifos and Cadusafos are recommended for quick knockdown of *H. indicola*, while agenda as slow acting insecticide for long time control of the termite species.

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