ORIGINAL ARTICLE

Toxic effects of palpoluck *Polygonum hydropepper* L. and Bhang *Cannabis sativa* L. plants extracts against termites *Heterotermes indicola* (Wasmann) and *Coptotermes heimi* (Wasmann) (Isoptera: Rhinotermitidae)

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Abstract

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Toxic effects of palpoluck *Polygonum hydropepper* L. and Bhang *Cannabis sativa* L. plants extracts against termites *Heterotermes indicola* (Wasmann) and *Coptotermes heimi* (Wasmann) (Isoptera: Rhinotermitidae)

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A research project was carried out aimed at to study the toxic effects of Palpoluck *Polygonum hydropipper* L. and Bhang *Cannabis sativa* L. crude extracts against two species of termites i.e. *Heterotermes indicola* (Wasmann) and *Coptotermes heimi* (Wasmann) at Nuclear institute for Food and Agriculture (NIFA) Peshawar, Pakistan in April 2002. Results revealed that after ten days of feeding maximum percent mortality in case of *Polygonum hydropipper* L. leaf and flower extracts was 28.0, 52.0, 28 and 74.7 for *H. indicola* and

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Coptotermes heimi **respectively, while in control only 10.7 and 12.0% mortality were recorded. Similarly, for the same species of termites the percent mortality in *Cannabis sativa* L. extracts was 54.7, 64.0, 58.7 and 70.7 for leaf and seed extracts respectively, while in control only 12.0 and 10.7% mortality were observed. In each extract mortality was significantly different from that of control. Toxic effects of both extracts (leaf and flower) were more profound against *Coptotermes heimi* than *Heterotermes indicola* during these ten days of feeding. Also the seed extracts caused more mortality than the leaves for both species, suggesting the availability of high contents of toxic materials in seed.**

Key words : toxic effects, Bhang and Polygonum, extracts, *Heterotermes indicola* and *Coptotermes heimi*

The injudicious use of pesticides for the control of termites has generated a number of biological and environmental hazards in air, water, soil and food. These man-created problems have further resulted in phytotoxicity, mammalian toxicity, pesticides residues, insect resistance, insect outbreaks and increased cost of production. More than 1000 species of plants have been reported to have chemicals in leaves, stems, flowers, seeds and roots which have insecticidal property, only a few of them have been used for practical insect control on a commercial scale in the past. The chemical poisons of plants are mostly alkaloids. Alkaloids are plant products, which are nitrogenous in nature. They are heterocyclic compounds having strong effect on the nervous system of animals. The alkaloidal extracts when applied to the insects bring about disturbance in the nervous system and cause death. They are, therefore, basically nerve poisons (Shahid, 2003).

Various scientists (termatologists) divert their attention to discover such type of plants toxicants, which are echo-friendly and also play a better role in the control of these pests. (Zaheer *et al*., 1987; Lin and Wang, 1988; Haung *et al*., 1990; Parihar, 1994; Hutchins, 1997; Parihar and Singh, 1992 and Jalees *et al*., 1993) have studied different plant extracts for their toxicity, attractancy and repellency in various natural products against different termite and insect species.

Keeping in view the economic importance of the two termite genera *Heterotermes indicola* and *Coptotermes heimi* as common problematic pests of agricultural crops, and the importance of some weed plants as a cheap source of insecticides, laboratory trials were conducted to find out the toxic effects of Bhang and Palpoluck plant extracts, which may serve as a toxicant in termite control.

Materials and Methods

1. Collection of Experimental Termites

The experimental termites were collected from a termite-infested orchards/ buildings using a trapping technique used by (Salihah *et al*., 1993). [1]NIFA- TERMAPS* were installed in an infested 6 kanal bungalow at the university town Peshawar. After fifteen days the infested bundles of NIFA-TERMAPS were brought to the entomology laboratory where termites along with the soil and other debris were passed through different sieves and were separated from soil. After separating from soil and debris they were identified with the help of the taxonomic keys (Chaudhry *et al*., 1972) as well as by Dr.Zahoor Salihah and Dr.Abdus Sattar Khan, termite specialists at NIFA. The termites

[1] Please see endnotes.

*NIFA-TERMAP: It is equipment, consisting of five thin slices of 15x8x1.3 cm measurements of poplar wood wrapped in blotting paper tightened with rubber band. Then it is buried in soil encircled by a PVC pipe having a lid to protect it from rain. It is more attractive than the termite's natural environment, and has a great potential for attraction and harbouring thousands of termites inside the crevices of the slices. It captures the maximum numbers of termites, as discovered by NIFA scientists so it is named as NIFA-TERMAP.

were maintained in the laboratory for experimental use. Termite workers along with a few soldiers were used in the experiments.

2. Collection of experimental plants and extracts preparation

2.1 Palpoluck (*Polygonum hydropipper* L.)

The Palpoluck (*P. hydropipper*) plants were collected from Malakandhir Research Farm, NWFP Agricultural University Peshawar-Pakistan. These plants were brought to entomology laboratory of Nuclear Institute for Food and Agriculture (NIFA) Tarnab, Peshawar. The flowers and leaves were removed from the plants were chopped and finely grinded in the pestle and mortar. Then the chopped flowers and leaves were mixed separately in distilled water in the ratio of 1:1. (W/V) and were kept overnight and crude aqueous solutions were prepared from both flowers and leaves by filtering the mixture through a Whitman filter paper No.42. and were stored in refrigerator for further 24 hrs for experimentation. To study the toxic effect of these extracts two termite species, *H. indicola* and *C. heimi*, were used for bioassays.

2.2 Hemp or Bhang (*Cannabis sativa* L.)

These plants were collected from roadside near NIFA and were brought to entomology laboratory. Extracts were prepared from seeds and leaves by the same way as mentioned in 2.1.

3. Bioassays

Force feeding tests were conducted in the Petri dishes (5.5 cm dia.) for both plants. The extracts preparation procedure is discussed earlier in 2.1. Petri dishes were sterilized in the oven at 200ºC for two hours. Circular blotting papers were cut and the bottom of each sterilized glass Petri dish was provided with two of them and the lid of each Petri dish with one. Each filter paper in the bottom was soaked with 0.2 ml of the respective extracts concentrations to the extent that the extracts were fully absorbed. Soaking was carried out with the help of disposable syringe, for each concentration a new syringe was used. Three Petri dishes consisting of untreated filter papers were placed as control in each experiment. Then a population of 22 worker termites and 3 soldiers were added to each Petri dish. The Petri dishes were placed in desiccators having water at the bottom (92% RH) and were kept in the controlled room at temperature of 27±3ºC. Daily observations for 10 days on the mortality of both species i.e. *H. indicola* and *C. heimi* were made and the dead individuals in each Petri dish were sorted out using forceps. Each treatment was replicated thrice for each termite's *sp*. For all the experiments mortality data were analysed using proc. ANOVA (SAS institute, 1985). Duncan Multiple Range Test (DMR) was used for mean separation

Results and Discussion

Results on the effect of leaf and seed extracts of Palpoluck (*P. hydropipper*) and Bhang (*C. sativa*) on percent mortality of *H. indicola* and *C. heimi* are presented in Tables 1-4.

Palpoluck (*Polygonum hydropipper* L.) extracts

Table 1 shows that for *H. indicola* on 1st and 2nd day percent mortality in leaf and seed extract was 13.3,14.7 and 14.7, 20.00 respectively and were similar to each other but were significantly different from the percent mortality in control (2.7 and 5.3). On day 3 percent mortality of 24.00 was recorded in seed extracts followed by 16.00 in leaf extracts and were significantly different from that of control (6.7). On day 4 maximum mortality of 30.7 was recorded for seed extracts followed by 18.7 for leaf and was highly significant from that in control (6.7%). On days 5 and 6 percent mortality recorded in seed and leaf extracts was 32.00, 36.00 and 24.00, 26.7 respectively and was significantly different from control (8%). On days 8, 9 and 10 percent mortality recorded for seed and leaf extracts was significant and higher than percent mortality recorded in control (10.7). For *C. heimi* (Table 2) on day 1, 2 and 3, percent mortality for leaf extract and control was 4.00, 8.00, 12.00 and 1.3, 2.7 and 4.00 respectively which did not differ significantly from each other. However percent mortality recorded for seed extracts of

Table 1. Effect of leaf and seed extracts of *Polygonum hydropipper* L. on the percent mortality of *Heterotermes indicola.*

After days	Leaves	Seeds	Control
1	13.3 a	14.7 a	2.7 b
2	14.7 a	20.0 a	5.3 b
3	16.0 ab	24.0 a	5.3 b
4	18.7 b	30.7 a	6.7 c
5	24.0 a	32.0 a	8.0 b
6	26.7 a	36.0 a	8.0 b
7	28.0 b	49.3 a	8.0 c
8	28.0 b	52.0 a	10.7 c
9	28.0 b	52.0 a	10.7 c
10	28.0 b	52.0 a	10.7 c

Means with in a row followed by the same letter are not significantly different at 5% level of significance
Over all coefficient of variation = 6.52%

Table 2. Effect of leac and seed extracts of *Polygonum hydropipper* L. on the percent mortality of *Coptotermes heimi.*

After days	Leaves	Seeds	Control
1	4.0 b	18.7 a	1.3 b
2	8.0 b	25.3 a	2.7 b
3	12.0 b	30.7 a	4.0 b
4	16.0 b	37.3 a	5.3 c
5	22.7 b	46.7 a	5.3 c
6	28.0 b	53.3 a	8.0 c
7	28.0 b	60.0 a	9.3 c
8	28.0 b	64.0 a	10.7 c
9	28.0 b	69.3 a	10.7 c
10	28.0 b	74.7 a	12.0 c

Means with in a row followed by the same letter are not significantly different at 5% level of significance
Over all coefficient of variation = 5.31%

Table 3. Effect of leaf and seed extracts of Bhang (*Cannabis sativa* L.) on the percent mortality of *Heterotermes indicola.*

After days	Leaves	Seeds	Control
1	24.0 a	28.0 a	5.3 b
2	33.3 a	30.7 a	5.3 b
3	36.0 a	34.7 a	5.3 b
4	38.7 a	38.7 a	8.0 b
5	40.0 a	42.7 a	8.0 b
6	42.7 a	48.0 a	10.7 b
7	44.0 a	52.0 a	10.7 b
8	46.7 a	56.0 a	12.0 b
9	48.0 a	64.0 a	12.0 b
10	54.7 a	64.0 a	12.0 b

Means within a row followed by the same letters are not significantly different at 5 % level of significance
Over all coefficient of variation = 9.05%

Table 4. Effect of leaf and seed extracts of Bhang (*Cannabis sativa* L.) on the percent mortality of *Coptotermes heimi.*

Days	Leaves	Seeds	Control
1	18.0 a	24.0 a	1.3 b
2	22.7 a	30.7 a	2.7 b
3	28.0 a	34.7 a	4.0 b
4	30.7 a	40.0 a	5.3 b
5	42.7 a	49.3 a	5.3 b
6	48.0 a	54.7 a	8.0 b
7	52.0 a	61.3 a	9.3 b
8	54.7 b	66.7 a	10.7 c
9	58.7 b	69.3 a	10.7 c
10	58.7 b	70.7 a	10.7 c

Means within a row followed by the same letters are not significantly different at 5% level of significance.
Over all coefficient of variation = 7.39%

18.7, 25.3 and 30.7 was significantly different from control as well as from leaf extract. On days 4 to10 percent mortalities for leaf, seed extract and control was significantly different from each other. Maximum mortality on each of the 4 to 10 day was caused by seed extract followed by leaf extract and control. Not much literature is available on the test plant against insect pests. Palpuluck (*P. hydropeper*) is a common weed almost everywhere in Peshawar valley - Pakistan. Being a very cheap source, further studies are needed for the isolation of the factor (alkaloids) in the said plant.

Bhang (*Cannabis sativa* L.) extracts

For *H. indicola* as clear from the Table 3 that from 1 day to day 10 percent mortalities for leaf and seed extracts were not significantly different from each other but differed significantly from that of control. Maximum mortality on each day was recorded in seed extract followed by leaf extracts and control. The percent mortalities recorded for *C. heimi* of leaf and seed extracts up to day 7 was not significantly different from each other but were significantly different from the percent mortality recorded in control. However mortality from day 8 to day 10 differed significantly between leaf-seed extracts and between each extract and control (10.7%), as illustrated in Table 4. Jalees *et al.* (1993) also determined the insecticidal properties of *C. sativa* against the larvae of *Anopheles stephensi, Culex quinquefasciatus* and *Aedes aegypti* in the laboratory and found that 4% concentration killed all the larvae within 24 h. We also found the toxicity of this plant against *H. indicola* and *C. heimi*. The results of this study are similar to those of Jalees work because the mortality due to this plant was highly significant than that of the control. The insecticidal effects of *C. sativa* [hemp] were also found most effective by Parihar and Singh (1992) to cause early larval mortality of *Heliothis armigera*. Hiremath and Ahn (1997) concluded that *Cannabis sativa* L. caused 50% adult female mortality in a pest of rice, the paddy brown plant hopper (*Nilaparvata lugens*), so it may be utilized to reduce the pesticide application. McPartland (1997) reported *Cannabis sativa* L. as a pest repellent and pesticide. Dried leaves and flowers of this plant repell or kill insects, mites and nematodes. Plant extracts (either aqueous or polar organic solvent extracts) can also kill or repel insects, mites, and nematodes. Thomas *et al.* (2000) also showed that *C. sativa* L. caused 100% mosquito larvae mortality and also found that the aqueous oil extract was more toxic than the ethanolic extract. We also tested the seed and leaf aqueous extracts of *C. sativa* L. and found that both treatments cause mortality that was significantly different from that of control. Also the lethal time (LT_{50}) indicated that the *Polygonum hydropipper* seed extracts have lower LT_{50} value, meaning that it caused 50 % mortality in less time than the leaf extracts. The seed extract LT_{50} was also lower than those of leaf extracts for both termites as well as of both plants indicating the availability of high

Table 5. LT_{50} (days) for both termite species against plants extracts of *Cannabis sativa* L.

Treatments	LT_{50} (Days) ± SD		Termite Species
	Lower limit	Upper limit	
Seeds extracts	4.7±0.33	8.9±0.94	*Heterotermes indicola* W.
Leaves extracts	7.73±0.89	9.69±1.29	*H. indicola* W.
Seeds extracts	4.44±0.65	8.74±1.03	*Coptotermes heimi* W.
Leaves extracts	5.17±0.713	8.69±0.939	*C. heimi* W.

Table 6. LT_{50} (days) for both termite species against plants extracts of *Polygonum hydropipper* L.

Treatments	LT_{50} (Days) ± SD		Termite Species
	Lower limit	Upper limit	
Seeds extracts	5.7±0.43	7.6±0.94	*Heterotermes indicola* W.
Leaves extracts	6.73±0.89	8.79±1.29	*H. indicola* W.
Seeds extracts	4.17±0.713	6.69±0.875	*Coptotermes heimi* W.
Leaves extracts	6.04±0.65	7.14±1.03	*C. heimi* W.

contents of toxic materials in seed extracts and also showing the higher susceptibility of *Coptotermes haimi* against these extracts than the *Heterotermes indicola* (Tables 5 and 6).

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