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Evaluation of some indigenous plant extracts as toxicants against lesser mealworm, *Alphitobius diaperinus* (Panz.) and rice weevil, *Sitophilus oryzae* (L.)

M. Shahjahan[1], M. Kamruzzaman[2] and F. Diba[3]

[1]Department of Entomology, Bangladesh Agricultural University, Mymensingh, [2]Bangladesh Institute of Nuclear Agriculture (BINA), P.O. Box 4, Mymensingh and [3]Department of Agronomy, Bangladesh Agricultural University, Mymensingh

Abstract

Experiments were conducted to evaluate the toxicity of six botanicals, dholkalmi, *Ipomoea fistulosa*; dutura, *Datura fastuosa*; eucalyptus, *Eucalyptus citriodora*; hatisur, *Helitropium indicum*; khetpapri, *Hedyotis corymbosa* and urmoi, *Sapium indicum* against lesser mealworm, *A.* (Panz) *diaperinus* and rice weevil, *S.* (L.) *oryzae*. Leaf and seed extracts of botanicals were prepared by using acetone, ethanol and water as solvents. The Results showed that all the extracts of test plants had toxic effect on both the insects. The dholkalmi bark extract was toxic for lesser mealworm (mortality, 47.92%) and dholkalmi leaf extract was found to be the most toxic for rice weevil (mortality, 44.89%). Ethanol extract showed highest toxic effect followed by acetone and water extracts. Effectiveness of all the plant extracts increased proportionally with the increase of doses and decreased proportionally with the increase of time. The seed/bark extract was more effective than leaf extract.

Keywords : Plant extracts, Lesser mealworm, Rice weevil, Toxicity

Introduction

The lesser mealworm is a cosmopolitan pest of stored products. In Bangladesh, this pest generally attacks husked rice and wheat. The rice weevil is a serious pest of rice, wheat and other grains in storage. It damages stored grains in Bangladesh causing serious loss to national economy. They attack moist and badly preserved grains and reduce wheat seed viability by feeding on the embryo (Gautam, 1989). Synthetic pesticides have been used for many years to control agricultural insect pests including those damaging durable food crops in storage. However, considerable problems may arise from the continued application of these insecticides including genetic resistance of pest species, toxic residues, increasing cost of application, environmental pollution and hazards from handling etc. Thus, there is an urgent need for safe but effective and biodegradable pesticides with no toxic effects on non-target organisms. This has created a world wide interest in the development of alternative strategies, including the search for new types of insecticides, re-evaluation and use of age-old traditional botanical pest control agents (Heyde *et al.*, 1984). Botanical insecticides are broad-spectrum, safe to apply, unique in action and can be easily processed and are less expensive. The local botanicals like bonkalmi (*I. sepiara*) and dutura (*D. fastuosa*) were found very effective in killing on an average about 88% of hispa adult at 48 hours after application (Karim, 1986). The present experiment was, therefore, undertaken to study the toxic effect of dholkalmi, dutura, eucalyptus, hatisur, khetpapri, urmoi leaf and seed/bark extracts in controlling lesser mealworm and rice weevil.

Materials and Methods

Experiments on the insecticidal effects of some plant leaf, seed and bark extracts against lesser mealworm, *Alphitobius diaperinus* and rice weevil, *Sitophilus oryzae* were conducted in the laboratory of the Department of Entomology, Bangladesh Agricultural University, Mymensingh during the period from April 1999 to April 2000. The lesser mealworm and rice weevil were reared separately in round plastic jars (12 x 23 x 6.5 cm in size). Lesser mealworms were given wheat grains and rice weevils were given rice grains. Experiments were conducted in growth chamber at 18.7–28.98^{0}C temperature and 73.34–87.90% relative humidity.

Preparation of plant extracts

The test plant materials (leaves, seeds, barks) of dholkalmi, dutura, eucalyptus, hatisur, khetpapri and urmoi were collected from different areas of Bangladesh for this experiment. Fresh leaves, seeds and barks were washed in water and then air-dried in shade. The air-dried plant materials were then oven-dried at 60^{0}C. Oven-dried plant materials were ground manually and passed through a 25-mesh sieve to obtain fine dust and to preserve them into airtight plastic container, till their use in extract preparation. Thirty grams of each category of dust were taken in a 600 ml beaker and separately mixed with 300 ml of different solvents (acetone, ethanol and distilled water). Then the mixture was stirred for 30 minutes in a magnetic stirrer (at 6000 rpm) and left to stand for next 24 hours. The mixture was then filtered through a fine cloth and again through filter paper (Whatman No. 1). The filtered materials were taken into a round bottom flask and then condensed by evaporation of solvent in a water bath at 45^{0}C, 55^{0}C and 80^{0}C temperature for acetone, ethanol and water extracts, respectively. After the evaporation of solvent from filtrate, the condensed extracts were preserved in tightly corked-labelled bottles and stored in a refrigerator until their use for insect bioassays. Different concentrations of each category of plant extracts were prepared by dissolving them in the water prior to insect bioassay.

Insect bioassay

The toxicity test was conducted according to the method described by Talukder and Howse (1993) with slight modifications. The adult insects were chilled for a period of 10 minutes. Then the immobilized insects were picked up individually by using a small suction tube. One μl solution each of different concentrations was applied to the dorsal surface of the thorax of each insect with a micropipette. The doses were 7.5, 10.0, 12.5 and 15.0% for lesser mealworm and 2.5, 5.0, 7.5 and 10.0% for rice weevil. Thirty insects, in 3 replicates of 10 insects each, were treated at each dose. In the control, same number of insects was treated with solvent only. After treatment, the insects were transferred into 9 cm diameter petri dish (10 insects/petri dish) containing food. Insects were examined daily and those that did not move or respond to gentle touch were considered as dead. Insect mortalities were recorded at 24, 48 and 72 hours after treatment (HAT). Observed mortalities of the ~~insects were~~ corrected by Abbott's formula (1987) : corrected mortality = (observed mortality - control mortality/100-control mortality) x 100. The corrected mortality data were statistically analysed by completely randomized design (factorial CRD) after arcsine √percentage transformation. Mean values were compared by DMRT (Duncan, 1951). The corrected mortality data were analysed by probit analysis designed by Finney (1971) using Mstat Statistical Software in a micro computer.

Results and Discussion

The effects of dholkalmi, dutura, eucalyptus, hatisur, khetpapri and urmoi were investigated on lesser mealworm, *Alphitobius diaperinus* and rice weevil, *Sitophilus oryzae*. The results are presented in the Tables 1-8.

Plant leaf extracts

Average mortality percentage of lesser mealworm at 24, 48 and 72 HAT revealed that dholkalmi leaf extract possessed the highest toxic effect (42.07%) followed by dutura (36.15%) (Table 1). The order of toxicity was : dholkalmi > dutura > urmoi > eucalyptus > hatisur > khetpapri. Among the solvents, the highest toxicity effect was observed in ethanol extract (42.29%) and it was significantly different from acetone (35.56% and water (26.85%) extracts (Table 2). Average mortality percentages were directly proportional to the level of dose and time after treatment.

Observations on average mortality percentage of rice weevil at 24, 48 and 72 hour after treatment (HAT) indicated that dholkalmi leaf extract (36.67%) possessed the highest toxic effect, whereas hatisur leaf extract (21.04%) possessed the lowest toxic effect (Table 1). The order of toxicity was : dholkalmi > khetpapri > urmoi > eucalyptus > dutura > hatisur. Mortality percentage was directly proportional to time. Among the solvents, ethanol extract was found more toxic (37.89%) and significantly different from acetone (29.74%) and water (18.00%) extracts (Table 2). Mortality percentage was also found directly proportional to the level of doses of plant extracts (Table 3).

Table 1. Mean mortality percentages of lesser mealworm and rice weevil treated with leaf extracts of different plants by topical application method at different HAT

Name of the plants	Mortality percentage							
	Alphitobius diaperinus				*Sitophilus oryzae*			
	24 HAT	48 HAT	72 HAT	Average value	24 HAT	48 HAT	72 HAT	Average value
Dholkalmi	36.67	43.33	46.22	42.07a	27.78	37.33	44.89	36.67a
Dutura	29.11	37.56	41.78	36.15b	17.11	22.89	25.78	21.92d
Eucalyptus	28.89	36.44	39.78	35.03bc	21.78	29.11	32.44	27.27c
Hatisur	27.56	33.11	36.44	32.37cd	15.56	20.67	26.89	21.04d
Khetpapri	24.67	31.78	35.33	30.59d	25.33	31.56	40.22	32.37b
Urmoi	29.33	36.22	40.00	35.18bc	24.22	32.44	37.78	31.48b
Sx		0.842		0.486		1.073		0.618
Probability level		NS		0.01		NS		0.01

Table 2. Effect of different leaf extracts of different solvents on the mortality of lesser mealworm and rice weevil by topical application method at different HAT

Name of the solvents	Mortality percentage							
	Alphitobius diaperinus				*Sitophilus oryzae*			
	24 HAT	48 HAT	72 HAT	Average value	24 HAT	48 HAT	72 HAT	Average value
Acetone	30.78 d	37.56 c	41.33 b	35.56 b	21.22 d	30.00 c	38.00 b	29.74 b
Ethanol	35.67 c	43.89 b	47.33 a	42.29 a	30.56 c	38.89 b	44.22 a	37.89 a
Water	21.67 f	27.78 e	31.11 d	26.85 c	14.11 f	18.11 e	21.78 d	18.00 c
Sx	0.595			0.343	0.759			0.438
Probability level	0.05			0.01	0.05			0.01

HAT = Hour after treatment
NS = Not significant
Within column values followed by different letter(s) are significantly different by DMRT

Table 3. Mean effect of plant leaf extract in different dose level on the mortality of lesser mealworm and rice weevil by topical application method at different HAT

Mortality percentage									
Alphitobius diaperinus					*Sitophilus oryzae*				
Doses (%)	24 HAT	48 HAT	72 HAT	Average value	Doses (%)	24 HAT	48 HAT	72 HAT	Average value
7.5	25.18 i	32.03 h	35.56 gh	30.92 d	2.5	16.67 h	23.56 fg	28.15 ef	22.79 d
10.0	33.70 h	43.14 f	47.22 de	41.36 c	5.0	23.15 g	31.30 e	37.04 bd	30.49 c
12.5	39.44 fg	49.81 cd	53.70 bc	47.65 b	7.5	29.63 ef	39.26 d	47.41 bc	38.76 b
15.0	48.15 d	57.03 b	63.15 a	56.23 a	10.0	40.37 cd	50.20 b	58.70 a	49.75 a
Sx	0.768			0.443		0.980			0.565
Probability level	0.01			0.01		0.01			0.01

HAT = Hour after treatment
Within column values followed by different letter(s) are significantly different by DMRT

Plant seed/bark extracts

Observations on average mortality percentage of lesser mealworm at 24, 48 and 72 HAT indicated that dholkalmi seed extract had the most toxic effect (47.92%) and the lowest in khetpapri seed extract (40.22%) (Table 4). The order of toxicity of six plant seed extracts was : dholkalmi > hatisur > dutura > urmoi > eucalyptus > khetpapri. Considering the solvents, ethanol extract showed the highest toxic effect (52.30%) followed by acetone (51.11%) and water (28.44%) extracts (Table 5). The mortality of lesser mealworm gradually increased with the increase of doses (Table 6). This results suggest that dholkalmi and dutura extracts can be used as toxicant agents in storage management systems.

Mortality percentage of rice weevil at 24, 48 and 72 HAT indicated that the dutura seed extract possessed the highest toxic effect (38.14%) followed by khetpapri (37.40%) (Table 4). The order of toxicity of six plant seed extracts was : dutura> khetpapri> dholkalmi > urmoi > eucalyptus> hatisur. Among three solvents, ethanol extract showed the highest toxic effect (43.70%) than that of acetone (31.93%) and water (29.00%) extracts (Table 5). The doses of plant extracts were found to have a profound influence on the mortality of rice weevil at different hours after treatment. Irrespective of plant, the lowest mortality was caused by the dose of 2.5%. The effectiveness of the extract increased gradually with increase of doses and the highest mortality was caused by the highest dose (Table 6).

Table 4. Mean mortality percentages of lesser mealworm and rice weevil treated with seed/bark extracts of different plants by topical application method at different HAT

Name of the plants	Mortality percentage							
	Alphitobius diaperinus				*Sitophilus oryzae*			
	24 HAT	48 HAT	72 HAT	Average value	24 HAT	48 HAT	72 HAT	Average value
Dholkalmi	41.78	49.56	52.44	47.92 a	27.56	36.44	40.22	34.74 b
Dutura	38.44	44.89	47.33	43.55 c	32.22	40.00	42.22	38.14 a
Eucalyptus	34.89	42.44	46.22	41.18 cd	28.44	34.00	40.44	34.29 b
Hatisur	41.33	49.11	51.78	47.40 b	24.00	31.56	34.67	30.07 c
Khetpapri	34.00	41.11	45.56	40.22 d	32.22	38.89	41.11	37.40 ab
Urmoi	38.00	44.67	47.56	43.41 c	28.67	36.00	38.67	34.44 b
Sx	0.881			0.508	1.077			0.622
Probability level	NS			0.01	NS			0.01

HAT = Hour after treatment

NS = Not significant

Within column values followed by different letter(s) are significantly different by DMRT

Table 5. Effect of different seed/bark extracts of different solvents on the mortality of lesser mealworm and rice weevil by topical application method at different HAT

Name of the solvents	Mortality percentage							
	Alphitobius diaperinus				*Sitophilus oryzae*			
	24 HAT	48 HAT	72 HAT	Average value	24 HAT	48 HAT	72 HAT	Average value
Acetone	45.22 d	52.56 c	55.56 ab	51.11 a	26.89 f	33.00 d	35.89 c	31.93 b
Ethanol	46.67 d	53.56 bc	56.67 a	52.30 a	36.78 c	45.44 b	48.89 a	43.70 a
Water	22.33 g	29.78 f	33.22 e	28.44 b	22.89 f	30.22 e	33.89 cd	29.00 b
Sx	0.623			0.359	0.761			0.439
Probability level	0.05			0.01	0.05			0.01

Table 6. Mean effect of plant seed extracts in different dose level on the mortality of rice weevil and lesser mealworm by topical application method at different HAT

Mortality percentage									
Alphitobius diaperinus					*Sitophilus oryzae*				
Doses (%)	24 HAT	48 HAT	72 HAT	Average value	Doses (%)	24 HAT	48 HAT	72 HAT	Average value
7.5	30.56 h	38.52 g	40.74 g	36.60 d	2.5	19.63 h	25.74 g	27.78 fg	24.38 d
10.0	41.85 g	50.19 f	55.74 e	49.25 c	5.0	31.67 f	41.85 de	45.74 d	39.75 c
12.5	51.48 ef	62.78 d	67.30 cd	60.43 b	7.5	39.63 e	49.30 c	52.78 c	47.22 b
15.0	66.29 c	74.63 b	78.52 a	73.14 a	10.0	52.22 c	62.22 b	67.03 a	60.49 a
Sx	0.804			0.464		0.983			0.567
Probability level	0.01			0.01		0.05			0.01

HAT = Hour after treatment
Within column values followed by different letter(s) are significantly different by DMRT

Probit statistics

The results of the probit analysis for the estimation of LC_{50} values and their 95% fiducial limits and the slope of regression lines at 24, 48 and 72 HAT for the mortality of lesser mealworm are presented in Table 7. The LC_{50} values of dholkalmi (11.21%), dutura (13.44%), eucalyptus (14.02%) hatisur (13.47%), khetpapri (14.80%) and urmoi (12.57%) at 24 HAT indicated that dholkalmi was the most toxic (Table 7) followed by urmoi. At 48 HAT, it was observed among six plants that dholkalmi possessed the highest toxic effects (9.00%), whereas khetpapri possessed the least (12.04%) toxic effect. At 72 HAT, similar trend of results was also observed. The chi-square values of different plant extracts at different HAT were insignificant at 5% level of probability and did not show any heterogeneity of the mortality data.

The results of the probit analysis for the estimation of LC_{50} values and their 95% fiducial limits and the slope of regression lines at 24, 48 and 72 HAT for the mortality of rice weevil are presented in Table 8. The LC_{50} values of dholkalmi (12.71%), dutural (11.08%), eucalyptus (17.73%), hatisur (19.47%), khetpapri (10.89%) and urmoi (11.50%) at 24 HAT (Table 8) indicated that khetpapri was the most toxic followed by dutura. At 48 HAT, comparison of LC_{50} values showed that dholkalmi extract (7.34%) was highly toxic and it was followed by khetpapri extract (7.50%). When the LC_{50} values were compared at 72 HAT, it was found that dholkalmi extract (5.29%) maintained its highest toxicity followed by khetpapri extract (6.30%). From the above probit results it is clear that dholkalmi will be more effective for controlling the rice weevil. The chi-square (χ^2) values of different plant extracts at different hours after treatment were insignificant at 5% level of probability. Insignificant χ^2 value did not show any heterogeneity of the mortality data.

Table 7. Relative toxicity of different plant extracts treated against lesser mealworm at 24, 48 and 72 HAT

Name of extracts	No. of insect used	LC_{50} value (%)	95% fiducial limit	χ^2 value	Slope±SE
24 HAT					
Dholkalmi	720	11.21	10.20–12.27	1.51	2.32±0.01
Dutura	720	13.44	11.87–15.20	1.17	2.19±0.01
Eucalyptus	720	14.02	12.16–16.18	0.88	2.06±0.01
Hatisur	720	13.47	11.90–15.20	0.14	1.93±0.01
Khetpapri	720	14.80	13.07–16.76	1.71	2.66±0.01
Urmoi	720	12.57	11.69–13.48	2.66	3.43±0.01
48 HAT					
Dholkalmi	720	9.00	8.03–10.06	0.13	2.32±0.01
Dutura	720	10.48	9.57–11.45	0.07	2.33±0.01
Eucalyptus	720	11.14	10.28–12.07	0.02	2.62±0.01
Hatisur	720	10.47	9.47–11.58	0.76	2.09±0.01
Khetpapri	720	12.04	11.04–13.09	1.00	2.66±0.01
Urmoi	720	10.79	10.13–11.48	0.71	3.42±0.01
72 HAT					
Dholkalmi	720	8.31	7.32–9.39	0.50	2.46±0.01
Dutura	720	9.53	8.68–10.44	0.004	2.56±0.01
Eucalyptus	720	10.09	9.28–10.76	0.46	2.96±0.01
Hatisur	720	9.51	8.55–10.56	0.74	2.26±0.01
Khetpapri	720	10.77	10.02–11.56	0.22	2.93±0.01
Urmoi	720	10.00	9.37–10.66	0.84	3.39±0.01

HAT = Hour after treatment

Values were based on two plant parts, three solvents, four concentration, three replications of 10 insects each

χ^2 = Goodness of fit

The tabulated value of χ^2 is 5.99 (d.f = 2 at 5% level)

Table 8. Relative toxicity of different plant extracts treated against rice weevil at 24, 48 and 72 HAT

Name of extracts	No. of insect used	LC_{50} value (%)	95% fiducial limit	χ^2 value	Slope±SE
24 HAT					
Dholkalmi	720	12.71	9.05–17.82	1.15	1.22±0.008
Dutura	720	11.08	9.01–13.58	5.25	1.76±0.008
Eucalyptus	720	17.73	10.45–29.99	1.29	1.04±0.008
Hatisur	720	19.47	12.11–31.26	0.98	1.26±0.008
Khetpapri	720	10.89	8.34–14.22	2.20	1.32±0.008
Urmoi	720	11.50	8.95–14.72	2.99	1.50±0.008
48 HAT					
Dholkalmi	720	7.34	6.44–8.37	0.60	1.90±0.008
Dutura	720	8.46	7.08–10.06	2.72	1.55±0.008
Eucalyptus	720	9.21	7.36–11.51	0.35	1.34±0.008
Hatisur	720	17.36	10.05–29.92	3.58	0.98±0.008
Khetpapri	720	7.50	6.42–8.74	1.60	1.62±0.008
Urmoi	720	8.05	6.67–9.68	1.50	1.42±0.008
72 HAT					
Dholkalmi	720	5.29	4.53–6.18	1.09	1.44±0.008
Dutura	720	7.20	6.26–8.26	5.51	1.71±0.008
Eucalyptus	720	8.03	6.96–9.26	1.21	1.54±0.008
Hatisur	720	11.18	8.06–15.48	5.77	1.12±0.008
Khetpapri	720	6.30	5.59–7.11	3.62	1.94±0.008
Urmoi	720	6.79	5.88–7.83	2.02	1.67±0.008

HAT = Hour after treatment
Values were based on two plant parts, three solvents, four concentration, three replications of 10 insects each
χ^2 = Goodness of fit
The tabulated value of χ^2 is 5.99 (d.f = 2 at 5% level)

From the above probit results it is clear that all the six plants would be effective for controlling rice weevil and lesser mealworm. The present study revealed the reduction of pest population due to use of leaf and bark extract of dholkalmi and seed extract of dutura and agreed with the previous findings of Malik and Naqvi (1984), Karim (1986), Saxena and Sumithra (1988), Khalequazzaman and Islam (1992) and Mohamed (1997).

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