Pak J Sci Ind Res 2002 45 (1) 46-49

Laboratory studies on Neem Kernels and Leaves as Wheat Protectants

Shams Zahoor, Roshan A Qureshi*, Shakira Quadri, Zamir Ahmad and Yazdana M Rizki

Applied Biology and Marine Resources Research Centre, PCSIR Laboratories Complex, Karachi-75280, Pakistan

(Received 5 January 2000; accepted 22 February 2001)

The extracts of neem seed kernels and leaves and their powders were tested for the repellent and antifeedant activities against *Tribolium castaneum* (Herbst) and *Rhizopertha dominica* (Fabricius) respectively. The best repellent activity class (iv) was observed in fresh seed kernels extracted in ethyl alcohol at 150 mg cm^2. Whereas the benzene, hexane and acetone extracts of the seed kernels were not effective at the same dose. Dry powder of fresh seed kernels protected wheat from infestation upto one year at higher dose.

Key words: Neem, Stored grain, Insect repellency, *Azadirachta indica*, wheat, *T. castaneum, R. dominica.*

Introduction

The protection of stored products by the use of plant materials is a common practice among farmers, particularly in tropical and subtropical regions. Over the last 40 years many synthetic insecticides have been used in agriculture and household which do not fulfil the requirements of integrated pest management (IPM). The negative impact on the environment and pest resistance led to the search for new compounds of natural origion. A number of investigators (Jacobson 1975; Grainge *et al* 1985: Sighamony *et al* 1986; Delobel and Malonga 1987; Talukder *et al* 1998) screened numerous plant extracts for their anti-insect activities. More emphasis was given to the neem tree *Azadirachta indica* (A. Juss), family Meliaceae which has long been used for medicinal and insecticidal properties in India and Africa.

Today it is well known that the leaves and seeds of *A. indica* are composed of 3 or 4 related compounds which belong to the general class of natural products called "triterpenes", more specially "limonoids" such as azadirachtin, meliantriol, salamin, nimbin, nimbidin with strong antifeedant growth regulatory and repellent properties for a number of insects, (Jilani *et al* 1988; Mohiuddin *et al* 1993; Schmutterer 1990; Akbar *et al* 1996).

The present study was undertaken to observe the repellent and antifeedant activities of the seed kernels and leaves of *Azadirachta indica* and their powders for protecting wheat against the attacks of *Tribolium castaneum* (Herbst) *Rhizopertha dominica* (Fabricius).

Materials and Methods

Rearing of test insects. The test insect *Tribolium castaneum* (Herbst) and *Rhizopertha dominica* (Fabricius) were maintained on wheat flour with 5% yeast and whole wheat grain respectively at 29±1°C and 60±5% R.H.

Preparation of plant material. The green seeds and yellow leaves were collected and dried at room temperature. The seed kernels and leaves were ground into fine powders and extracted with ethyl alcohol, benzene, hexane and acetone in soxhlet for 24 h. The solvent was evaporated under reduced pressure using rotary evaporator.

Bioassay. The repellency method followed is that of Malik and Naqvi (1984), with minor modification. The filter paper strips (Whatman No.1) 9 x 9 cm^2 dimensions were treated with the appropriate dilutions of extracts in acetone so as to deposit 150, 300 and 600 μg cm^2. The untreated strips were attached side to side with treated strips. After complete evaporation of acetone two glass rings (4.5 cm high, 8 cm diameter) were placed over these matched papers. Ten 2-3 week old *T. castaneum* adults were released into each ring and number of insects on treated and untreated halves were observed twice daily for five days. The overall average percentage of repellencey value for eight weeks were assigned as repellencey class by using the following scale.

Class I: 0.1-20%; Class II: 20.1-40%; Class III: 40.1-60%; Class IV: 60.1-80%; Class V: 80.1-100% (McGovern *et al* 1977).

Protection of wheat grain by surface treatment. The extracts of seed kernels and leaves were dissolved in acetone and the appropriate dilutions were added to 10 g of whole wheat grain so as to obtain 0.25, 0.5 and 1% concentration (w/w). Dry powder of kernels was thoroughly mixed with wheat at 1.25-10% and leaf powder at 30% concentration (w/w). Each treatment was replicated three times. Ten 1-2 weeks old adults were observed weekly for one month and total surviving adults number and weights of damaged and undamaged seeds were noted after two months. The

*Author for correspondence

percentage weight loss of grain was calculated as follows according to the formula reported by Golob *et al* 1982:

$$\% \text{ Wt.loss} = \frac{(UNd - DNu) \times 100}{UX (Nd + Nu)}$$

Where U = Weight of undamaged grain
D = Weight of damage grain
Nd = Number of damage grain
Nu = Number of undamaged grain

Results and Discussion

It was observed that the repellent activity of fresh kernel extracts against *T. castaneum* was maximum (Class IV) at 150 μg cm^{-2} dose and persisted upto eight weeks whereas the extracts of one year old seed kernels showed repellent activity of class III at 600 μg cm^{-2} dose (Table 1).

The kernel extract was more effective than leaf extract at 0.25% conc. (Table 2) and kernel powder was more effective than leaf powder. The effectiveness of kernel powder persisted upto six months at 5% conc. and one year at 10% conc. (Table 3). The laboratory studies have shown that the alcoholic extracts of fresh seed kernel can provide protection to stored wheat from *T. castaneum* and *R. dominica* infestation (Table 1 and 2). According to Feuerhake (1984) *A. indica* limonoids are highly soluble in alcoholic solvents and the yields of active ingredients vary from 0.2 to 6.2% upon extraction. The fresh kernels contain 2-4 mg of azadirachtin per gram of kernels. It is also observed that extracts of one year old kernels lose their repellent activity and require 600 μg cm^{-2} dose to achieve class III repellency (Table 1). When exposed to sun light or high temperatures, the neem compounds lose their pest control potential.

The fresh kernel powder can protect the wheat from *R. dominica* at higher concentrations (Table 3). At 10% conc. no loss of grain was observed upto one year. These findings support the observation of Siddiqui, (1980) who revealed that powder of neem seeds protect wheat against *Trogoderma granarium* over periods extending from 207 to 490 days. Use of plants for pest infestation control in stored grains, therefore, seems to offer a practical solutions, especially in developing countries, where plants are found in abundance throughout the year.

Table 1
Average repellency of neem seed kernel extracts against *Tribolium castaneum* adults

Solvent used	Conc. μg cm^{-2}	% Repellency after weeks 1st	2nd	4th	8th	Mean Repellencey %	Repellency class
*Ethyl alcohol	150	95.4±2.1	80.0±1.4	62.2±6.5	52.5±3.1	72.5	IV
•Benzene	150	63.6±5.6	42.5±7.9	21.3±6.0	20.7±9.3	37.0	II
	300	63.9±3.8	49.8±3.7	42.1±5.0	26.4±4.7	45.6	III
	600	70.2±4.7	59.4±6.0	45.6±3.6	30.0±3.7	51.3	III
•Acetone	150	27.3±4.1	20.6±7.0	17.8±6.4	16.6±4.4	20.6	II
	300	47.8±7.0	40.5±6.6	32.5±1.6	12.9±2.9	33.4	II
	600	58.9±4.4	47.5±2.1	38.6±3.8	24.4±2.5	42.4	III
•Hexane	150	36.3±10.2	25.0±7.5	14.2±9.6	13.3±4.8	22.2	II
	300	47.1±5.7	35.6±2.9	24.0±3.3	17.1±2.7	30.9	II
	600	53.9±4.4	49.5±2.1	36.7±3.8	23.9±2.9	41.0	III
•Hexane + Acetone (1:1)	150	63.1±10.1	41.6±7.3	21.9±7.7	17.8±7.5	36.1	II
	300	65.0±5.7	44.5±5.1	33.1±4.7	25.3±4.3	41.9	III
	600	69.6±3.1	51.8±2.9	42.1±5.4	28.7±1.7	48.1	III
Neem Seed Covering	150	20.6±4.4	18.1±9.9	8.0±4.3	6.1±5.8	13.2	I
	300	30.6±43.0	26.5±4.3	14.2±5.8	7.9±5.0	19.8	I
	600	41.5±3.0	38.1±5.1	28.2±2.9	12.5±5.3	30.1	II
Control	-	11.7±6.2	7.2±5.6	15.7±3.9	7.4±6.0	10.5	I

* Extract of fresh seed kernel; • Extraction of seed kernel after one year ; ± S.D. or S.E.

Table 2
Protection of wheat grain by surface treatment with neem seed kernel/extract against *Rhizopertha dominica* (F.)

Extracts	% Conc. w/w	*Mean %mortality after one month	Mean No. of F_1 generation after 2 months	% Reduction of F1 generation	Mean % wt. loss of grains
Kernel	1.0	100	Nil	100	No loss
	0.5	100	Nil	100	No loss
	0.25	80.0±5.8	Nil	100	0.5±0.1
Yellow	1.0	56.7±3.3	20.0±9.9	88.8	1.2±0.2
leaves	0.5	46.7±3.3	40.3±6.0	77.5	3.9±0.4
	0.25	40.0±11.6	41.7±1.7	76.7	8.7±1.6
Green	1.0	56.7±12.0	29.3±10.5	83.6	6.2±0.5
leaves	0.5	33.3±6.6	48.3±2.6	73.0	12.9±4.5
	0.25	33.3±3.3	56.0±5.0	68.7	18.5±3.6
Control	-	10.0±5.8	179.0±6.6	-	41.3±5.8

-Average of three replicates; ± Standard error of the mean.

Table 3
Effect of dry powder of neem seed kernels and leaves against *Rhizopertha dominica* (F.)

Neem parts used and formulation	% concentration (w/w)	Mean % mortality in one month	Mean No. of F_1 generation after 2 months	Mean % weight loss of grains
Dry kernel powder	10*.0	100.0	Nil	No loss
	5.0	48.4±6.0	14.0±6.3	2.8**±1.3
	2.5	36.7±6.7	84.0±14.0	11.5±1.7
	1.25	33.3±3.3	144.0±10.1	15.0±3.5
Pieces of kernel	30.0	70.0±5.8	15.0±3.5	2.2±0.5
Whole kernel	30.0	30.0±1.0	54.0±3.5	13.1±8.2
Yellow leaves powder	30.0	60.0±5.8	55.0±9.5	11.1±2.9
Green leaves powder	30.0	56.6±6.6	73.0±5.6	21.8±3.3
Control	-	10.0±5.8	178.0±6.4	44.5±3.4

* Observed upto one year ; ± Standard error of the mean; **Weight loss of grain after six months

References

Akbar S, Ahmad S, Hassan M U 1996 Efficacy of neem products and insecticides against white backed plant hopper, *Sogatella furcifera* (Horv). *Pak J Zol* **28** 5-7.

Delobel A, Malonga P 1987 Insecticidal properties of six plant materials against *Crayedon serratus* (Ol.), Coleoptera, Bruchidae. *J Stored Product Res* **23** 173-176.

Feuerhake K J 1984 Effectiveness and sensitivity of technical solvents for the extraction of neem seed components with insecticidal activity. *Proc 2nd Int Neem Conf* 1983, pp 103-113.

Golob P, Mwambula J, Mhvango V, Ngulube F 1982 The use of locally available materials as protectants of maize grain against insect infestation during storage in Malawi. *J Stored Product Res* **18** 67-74.

Grainge M, Ahmad S, Mitchell W C, Hylin J V 1985 Plant species reportedly possessing pest control properties. *EWC/UH database East/West Resource system Ist*, Honolulu, Hawaii, USA.

Jacobson M 1975 *Insecticides from plants. A review of the literature (1954-1971). Agric Hand Book No.461*, USDA.

Jilani G, Saxena R C, Rueda 1988 Repellent and growth-inhibiting effects of turmeric oil, sweet flag oil, neem oil and Margosan-0 on red flour beetle (Coleoptera:

Tenebrionidae). *J Econ Entomal* **81** 1226-1230.

McGovern T P, Gillenwater H B, McDonald 1977 Repellents for adult *Tribolium confusum*. *J Ga Ent Soc* **12** 79-84.

Malik M M, Naqvi S H M 1984 Screening of some indigenous plants as repellents or antfeedants for stored grain insects. *J Stored Product Res* **20** 41-44.

Mohiuddin S, Qureshi R A, Ahmad Z, Qureshi S A 1993 Laboratory evaluation of some vegetable oils as protectants of stored products. *Pak J Sci Ind Res* **36** 377-379.

Siddiq S A 1980 Efficacy and persistence of powdered neem seeds for treatment of stored wheat against *Trogoderma granarium*. *Proc Int Neem Conf,* Rottech-Egern, p 251-258.

Sighamony S, Anees I, Chandrakala T, Osmani Z 1986 Efficacy of certain indigenous plant products as grain protectants against *Sitophilus oryzae* and *Rhyzopertha dominica*. *J Stored Product Res* **22** 21-23.

Schmutterer H 1990 Properties and potential of natural pesticides from the neem tree *Azadirachta indica*. *Ann Rev Entomol* **35** 271-297.

Talukder D, Malek M A, Khanum L A M, Dey K C 1998 Toxicity of some indigenous plant seed oil against *Tribolium confusum* Dural (Coleoptera: Tenebrionidae) adults. *Pak J Zool* **30**(4) 331-334.