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Repellent and Toxicological Impact of Acetone Extracts of *Nicotiana tabacum*, *Pegnum hermala*, *Saussurea costus* and *Salsola baryosma* against Red Flour Beetle, *Tribolium castaneum* (Herbst)

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Abstract.- Acetone extracts of four plants, *Nicotiana tabacum* (Tobacco), *Pegnum hermala* (Hermal), *Saussurea costus* (Kust-e-shireen) and *Salsola baryosma* (Khar booti) at different concentrations *viz*., 2.5, 5, 7.5 and 10% were evaluated for their repellent and toxicological effects against *Tribolium castaneum* (Herbst). The results showed that the highest percent repellency (93.33%) was achieved in *N. tabacum* extract followed by *S. baryosma, P. hermala* and *S. costus*. The highest mortality (6.69%) was recorded in *N. tabacum* extract at 10% concentration after an interval of 72 hours. In conclusion, *N. tabacum* and *S. baryosma* extracts are more effective in terms of their repellent and toxicant action against *T. castaneum* than those of *P. hermala* and *S. costus* and could be recommended for the management of red flour beetle in integration with other control measures.

Keywords: *Nicotiana tabacum, Pegnum hermala, Saussurea costus, Salsola baryosma,* repellency, toxicity, *Tribolium castaneum.*

INTRODUCTION

Almost one third of the total world's food production is destroyed annually by more than 2000 species of field and storage pests, and 43% losses of potential production has occurred in developing countries of Asia (Ahmed and Grainge, 1986). *Tribolium castaneum* (Herbst) is very common and most destructive pest of stored products throughout the world and is generally found in granaries, mills, warehouses and stored grains. Both the larvae and adults not only feed on fresh grains but they can also feed on grains already damaged by other insect pests (Sharaby, 1988). The presence of this pest in stored products results in contamination and economic damage and also decreases their nutritive value as well (Barkholder and Faustini, 1991). Mondal (1994) reported that red flour beetle not only affects the quality and quantity of grains but it also attacks the germ part or embryo portion of grains. Freeman (1973) recorded that 5-10% of the total agriculture production of world is damaged by insects whereas 10-25% post harvest losses have been reported due to the damage caused by the insects, microbes and other factors (Avesi, 1983; Matthews, 1993).

Currently synthetic pesticides are mostly used for the control of stored-grain pests but the frequent use of these pesticides has made the pest strains resistant against these pesticides (Subramanyam and Hagstrum, 1995). Pesticide chemicals which are mostly used for crop protection could be environmentally pollutants and also have adverse effects on animals and human beings (Meena *et al*., 2006, Hashim and Davi, 2003). Phosphine fumigation has become increasingly limited in its use because of resistance developed in stored-grain insects to this fumigant, which is now reported from more than 45 countries (Bell and Wilson, 1995; Chaudhry, 1995).

As a large number of stored-grain pests have been reported to develop resistance against synthetic pesticides now there is need to develop some safe methods for the control of these pests. Insecticidal effect of many plants against pests of stored grain has been demonstrated (Abubakar *et al*., 2000; Boussalis *et al*., 1999; Fields *et al*., 2001). Several products of floral species have been evaluated to act as repellents, toxicants and antifeedants against a

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number of coleopteran species that attack stored grain and their products (Papachristos and Stamopoulos, 2002; Tapondjou *et al*., 2002).

Present study was thus carried out to determine the efficacy of four botanicals, *Nicotiana tabacum*, *Pegnum hermala*, *Saussurea costus* and *Salsola baryosma* against *Tribolium castaneum* (Herbst).

MATERIALS AND METHODS

Collection of insects

The adults and larvae of red flour beetle, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae) were collected from grain market and flour mills located in Faisalabad district and reared in the laboratory to develop a homogenous population.

Rearing of Tribolium castaneum *(Herbst)*

The insect culture was maintained in sterilized glass jars (1.0 kg capacity). The rearing jars were covered with muslin cloth, tightened with rubber bands to avoid the escape of beetles. Wheat flour was used as culture medium. Fifty beetles of *T. castaneum* were released in each jar. The jars were kept in cooled incubators at 30±2ºC and 65±5% R.H with slight modifications given by Mujeeb and Shakoori (2012). After 5 days, the adult beetles were sieved out from the culture medium. The sieved flour along with eggs was again put into the jars and kept again in cooled incubators at optimum rearing conditions to develop a homogenous F_1 population.

Collection of plant materials

The leaves of *P. hermala*, *N. tabacum* and *S. baryosma* were collected from district Layyah and plant parts of *S. costus* were collected from market.

Preparation of extracts

The leaves of test plants were cleaned by washing in water. After shade drying the leaves were grounded in electrical grinder to bring these in the form of powder (Sagheer *et al*., 2013). Extracts were obtained using Rotary Shaker by dipping 50 g powder in 100 ml acetone for 24 hours at 220 revolutions per minute. Chemical extracts obtained were put in clean and air tight lid bottles. The samples were stored in refrigerator before use. Different concentrations *viz*. 2.5, 5.0, 7.5 and 10.0% were prepared from the stock solution of each plant extract using acetone as solvent.

Bioassay studies

Applications of different concentrations were made on the wheat flour (Sagheer *et al*., 2012) Acetone was allowed to evaporate and the air dried treated flour was put in treatment vials. Twenty 3rd instar larvae of *T. castaneum* were released on the treated flour in each vial. These vials were placed in incubator at 30±2°C and 65±5% R.H. Data regarding % age mortality was observed 24, 48 and 72 hours after treatment (Awan *et al*., 2012).

In another bioassay, the repellent effects of these plant extracts against adults of *T. castaneum* were evaluated by using the area preference method. Plant extracts in different doses were applied on filter paper (9 cm diameter) cut in half. The concentrations *viz*., 2.5%, 5.0%, 7.5% and 10.0% of acetone extracts were applied to one half of the filter paper, the other half filter paper was treated with acetone alone. Both the treated and untreated halves of filter paper were joined with staple pins and then were placed in Petri dishes. Twenty adults were put in the centre of each Petri dish. Petri dishes were subsequently covered. Each treatment was replicated thrice and the numbers of insects on the two half paper disks were recorded after 24 hours from the beginning of the test.

Analyses of variance of the collected data were carried out using statistica-6 software. Means of treatments were separated by using Tuckey-HSD test at 5% significant level.

RESULTS AND DISCUSSION

Significant insecticidal activity against larvae and adults of *T. castaneum* was recorded with acetone extract of *N. tabacum, P. hermala, S. costus* and *S. baryosma* in experiments of adult repellency and larval mortality. Higher concentration (10.0%) of all plants in the form of acetone extracts showed higher repellency of adult beetles and mortality of larvae as compared to lower concentration (2.5%). *Nicotiana tabacum* and *S. baryosma* proved more

effective plant extracts as they showed maximum repellency and larval mortality at high concentration (10.0%).

The result reveals that the effect of plant extracts and concentrations is highly significant. Table I shows that maximum mean repellency (93.33%) of the test insect was recorded in those treatments where 10% dose of acetone extract of *N. tabacum* was applied and it differed significantly from all other treatments, while minimum repellency (35.0%) was observed in plant extract of *S. costus* at a concentration of 2.5%. Among all plant extracts *N. tabacum* extract showed the highest average mean repellency (75.67%) while the plant extract of *S. costus* resulted in the lowest average mean repellency (52.50%). The plant extracts of *P. hermala* and *S. baryosma* showed same (62.08%) average mean repellency. Whereas, among doses of plant extract 10% dose of plant extracts resulted in the highest average mean repellency (78.85%) and it decreased with decrease in concentration such as 7.5% (68.34%) > 5% (57.92%) > 2.5% (45.84%).

In all plant extracts the effect of time interval is highly significant (Fig. 1). Among all the plant extract *S. costus* extract showed the lowest mean mortality (0.33%) after a time period of 24 h, while *N. tabacum* extract resulted in the highest mean mortality (3.59%) after an interval of 72 h. *S. baryosma* and *P. hermala* extracts also proved significantly different at various time intervals. There is also significant difference in concentrations and higher concentrations of all plant extracts showed the highest mortality (Fig. 2). The highest mean mortality (4.23%) was achieved by *N. tabacum* extract at the concentration of 10% and it differed significantly from all the concentrations of other tested plants., The lowest mean mortality was recorded at 2.5% dose of *P. hermala* extract as compared to other plant extracts at lower concentrations (Fig. 2). However at a dose of 5% and 7.5% of *P. hermala* extract showed higher mean mortality as compared to *S. costus* extract and lower mortality than *N. tabacum* and *S. baryosma.*

The results also revealed that interaction of time interval and concentrations had no significant effect on percent mean mortality (Table III). In interaction percent mean mortality ranged from 0.0% to 6.69%. The plant extract of *N. tabacum* showed the highest percent mean mortality (6.69%) at a concentration of 10% after an interval of 72 hours followed by *S. baryosma* and *P. hermala* extracts which had 4.35% mean mortality at maximum concentration after an interval of 72 hours. After a time interval of 24 hours at a concentration of 2.5% no mortality was observed in plant extracts of *N. tabacum*, *P. hermala* and *S. coctus*. Similarly, *S. coctus* and *P. hermala* extracts did not result in any mortality at 5% concentration after a time interval of 24 hours and at 2.5% concentration after a time interval of 48 hours, respectively.

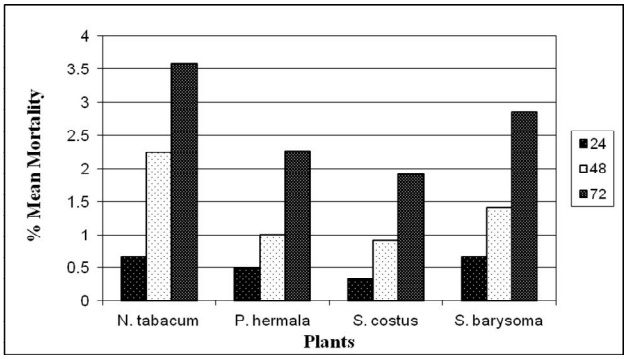

Fig. 1. Mean percent mortality of *Tribolium castaneum* against different plant extract after different exposure intervals.

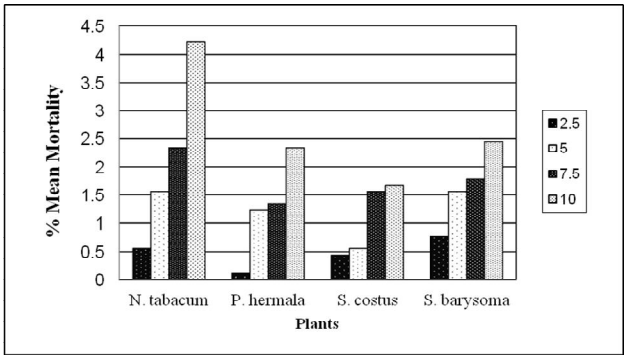

Fig. 2. Mean percent Mortality of *Tribolium castaneum* against different concentrations of four plant extracts.

The present study shows the effect of acetone extracts of four indigenous medicinal plants against *T. castaneum.* Significant insecticidal activity was achieved by *N. tabacum* extract followed by

Table I.- Repellency percentage of *Tribolium castaneum* (Herbst) at different concentrations of tested plant extracts.

Plants	Concentrations of tested plant extracts			
	2.5%	5%	7.5%	10%
N. tabacum	56.67 ± 4.41 c	66.67 ± 3.33 bc	80.00 ± 2.89 ab	93.33 ± 1.67 a
P. hermala	46.67 ± 7.26 bc	58.33 ± 6.67 ab	66.67 ± 4.41 ab	76.67 ± 4.41 a
S. costus	35.00 ± 5.00 cd	46.67 ± 4.41 bc	61.67 ± 4.41 ab	66.67 ± 1.67 a
S. baryosma	45.00 ± 2.89 b	60.00 ± 5.77 ab	65.00 ± 2.89 ab	78.33 ± 4.41 a

Table II.- Average mean comparisons of the data regarding the repellency percentage of *Tribolium castaneum* (Herbst) by plant extracts and their concentrations.

Plants	*N. tabacum*	*P. hermala*	*S. costus*	*S. baryosma*
Avg. Repellency	75.67 ± 3.07 a	62.08 ± 5.69 ab	52.50 ± 3.87 bc	62.08 ± 3.99 ab
Conc. %	**2.5**	**5**	**7.5**	**10**
Avg. Repellency	45.83 ± 4.89 c	57.92 ± 5.04 bc	67.08 ± 3.65 ab	78.75 ± 3.04 a

Table III.- Mean comparison of the data regarding percent mortality of larvae of *T. castaneum* by the interaction of time interval and concentration of plant extracts.

Time (h)	Conc. (%)	*N. tabacum* (% Mortality)	*P. hermala* (% Mortality)	*S. costus* (% Mortality)	*S. baryosma* (% Mortality)
24	2.5	0.00 e	0.00 d	0.00 c	0.33 ± 0.33 b
24	5	0.33 ± 0.33 de	0.67 ± 0.33 bcd	0.00 c	0.67 ± 0.33 b
24	7.5	0.67 ± 0.33 de	0.67 ± 0.33 bcd	0.67 ± 0.33 bc	0.67 ± 0.33 b
24	10	1.67 ± 0.33 cde	0.67 ± 0.33 bcd	0.67 ± 0.33 bc	1.0 ± 0.58 b
48	2.5	0.67 ± 0.33 de	0.00 d	0.33 ± 0.33 bc	0.67 ± 0.58 b
48	5	1.67 ± 0.33 cde	1.01 ± 0.33 bcd	0.33 ± 0.33 bc	1.34 ± 0.67 b
48	7.5	2.33 ± 0.67 bcde	1.01 ± 0.33 bcd	1.67 ± 0.33 abc	1.67 ± 0.58 ab
48	10	4.33 ± 0.33 ab	2.34 ± 0.88 ab	1.33 ± 0.33 abc	2.01 ± 0.33 ab
72	2.5	1.01 ± 0.33 de	0.68 ± 0.33 bcd	1.0 ± 0.58 abc	1.34 ± 0.88 b
72	5	2.68 ± 0.58 bcd	2.02 ± 0.33 abc	1.33 ± 0.33 abc	2.68 ± 0.58 ab
72	7.5	4.02 ± 0.88 bc	2.35 ± 0.58 ab	2.33 ± 0.67 ab	3.01 ± 0.33 ab
72	10	6.69 ± 0.58 a	4.35 ± 0.33 a	3.0 ± 0.58 a	4.35 ± 0.67 a

S. baryosma, *P. hermala* and *S. costus* extract. Similar observations of other plant extracts against other storage insect pests have been reported. For example, Negahban *et al*. (2006) studied the effect of *Artemisia scoparia* against three stored-product insects. The results reveal that the essential oil of *A. scoparia* had significantly high repellent activity against *T. castaneum*. Kumar *et al*. (2004) reported that *T. castaneum* repelled most quickly followed by *Sitophilus oryzae* L. and *Rhyzopertha dominica* (F.) when exposed to pea (*Pisum sativum* L.). The results of the present study are also in contrast with Upadhyay and Jaiswal (2007), they studied the effect of *Piper nigrum* oil and observed that larval and adult mortality increased while the larval survival and adult emergence decreased with increase in the concentration of essential oil.

In the present study it was concluded that the plant extracts of *N. tabacum* and *S. baryosma* are more effective against *T. castaneum*. These plant extracts has shown repellent and toxic effect. Therefore these plant extracts can be used potentially for controlling the population of *T. castaneum*.

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