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Growth Inhibiting Effects of Plant Extracts Against the Grain Moth, *Sitotroga cerealella* (Oliv.) (Gelechiidae: Lepidoptera)*

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Abstract.- Extracts of *Acorus calamus* (L.) (sweet flag), *Azadirachta indica* (A. Juss) (neem) and *Curcuma longa* (L.) (turmeric) prepared in petroleum ether, acetone and ethanol were evaluated as growth inhibitor against *Sitotroga cerealella* (Oliv.). Among these extracts, petroleum ether extract of sweet flag at application rates of 1000, 500 and 250 µg/g and its acetone extract at 1000 and 500 µg/g completely inhibited emergence of adults. Petroleum ether extract of neem was next to sweet flag. Turmeric was less effective than neem extract especially at 1000 and 500 µg/g application rates.

Key words: *Acorus calamus*, sweet flag, *Azadirachta indica,* neem, *Curcuma longa,* turmeric, *Sitotroga cerealella,* growth inhibition, botanical pesticides

INTRODUCTION

The important insect pests of stored cereals in Pakistan include *Tribolium castaneum* (Hbst.) (Red flour beetle), *Rhizopertha dominica* (F.) (Lesser grain borer), *Sitophilus oryzae* (L.) (Rice weevil), *Trogoderma granarium* Everts (Khapra beetle) and *Sitotroga cerealella* (Oliv.) (Angumois grain moth) (Iqbal *et al*., 1992). Their control is primarily dependant upon continued application of liquid insecticides (pyrethroids) and gaseous insecticides (methyl bromide and phosphine) (White and Leesch, 1995). Although effective, their repeated use for several decades has disrupted natural biological system and led to development of resistance to insecticides, undesirable effect on non target organisms and environment and human health (Champ and Dye, 1977; Subramanyam and Hagstrum, 1995; Irshad and Gillani, 1989, 1991; Irshad *et al*., 1992; Irshad and Iqbal, 1994). The adverse effects of pesticides enforced the scientists and chemists throughout world to search for safer pesticides. Throughout history, plant products have been successfully exploited as insecticides, repellents and antifeedents.

Many plant extracts are known to possess insecticidal activity against various stored product insects (Desmarchelier, 1994; Shaaya *et al*., 1997; Schmutterer, 1990, 1995). Some indigenous plants of Pakistan have been studied for repellent effects on red flour beetle, *T. castaneum* (Jilani *et al*., 1989, 1991, 1993), as grain protectant against insects (Jilani and Haq, 1984). Jilani *et al.* (2006) and Rehman *et al*. (2009) also tested some plant extracts against peach fruit fly (*Bactocera zonata* Saunders).

The present study has been carried with a view to access the growth inhibiting affects of the plant extracts of sweet flag, *Acorus calamus* (L.), turmeric, *Curcuma longa* (L.) and neem, *Azadirachta indica* (A. Juss) using solvents of various polarities *viz*. petroleum ether (low), acetone (medium) and ethanol (high), separately against grain moth. These plants are abundantly available and possess medicinal properties and are effective against storage pests (Iqbal *et al*., 2010).

MATERIALS AND METHODS

Sitotroga cerealella (grain moth) was reared in the laboratory on soft wheat under controlled temperature 27±1°C and 55±5% R.H. Rhizomes of *A. calamus* (L.) (sweet flag) and *C. longa* (L.) (turmeric) and seeds of *A. indica* (A. Juss) (neem) were purchased from the local market and were grinded to fine powder of 60 mesh. They were extracted on Soxhlet's extractor separately with

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petroleum ether (60-80°C), acetone and ethanol to get the compounds of various polarities. The left over solvent, if any, was evaporated on rotary evaporator and extracts were made solvent free in a vacuum desiccator. Four concentrations (1000, 500, 250 and 125 µg/g) were used with acetone as a carrier solvent. Three replicas of each concentration were applied. Wheat grains were used as host material to determine the effect of plant derivatives as growth inhibitors.

Wheat grains in lots of 50 g each were treated with 1000, 500, 250 and 125 µg/g of plant extract in 5 ml of acetone. In control, grains were treated with acetone only. After complete evaporation of acetone, 24 hours old 100 eggs of *S. cerealella* were placed in each treatment and as well as in control. The glass jars were covered with muslin cloth and kept in the controlled environment. After 4 weeks the adults were counted for 15 consecutive days. Data of adult emergence was recorded separately for each treatment and afterwards it was pooled for comparison of plants, solvents, application rates and their interactions. Data was analyzed with Duncan's Multiple Range Test at 5% probability.

RESULTS

Sweet flag extract in petroleum ether at 1000, 500 and 250 µg/g completely inhibited emergence of adults. Only 0.3 adults emerged at 125 µg/g as compared with 75.0 emerged in control (Table I). Its acetone extract also inhibited adult emergence at 1000 and 500 µg/g but not so in 250 and 125 µg/g. In petroleum ether extracts neem was next to sweet flag. Turmeric was less effective than neem extract especially at 1000 and 500 µg/g application rates. However, in case of acetone extract, turmeric was slightly better than neem at 1000 and 500 µg/g. Although neem was better than turmeric extract at 250 and 125 µg/g but was not significantly different. In the case of ethanol extract neem was significantly better than sweet flag at 1000 µg/g. There was significant difference of sweet flag with other plant extracts. It was also observed that all the plant extracts were significantly different from the control.

Our results indicate that sweet flag was the most active in inhibiting *S. cerealella* emergence. In grains treated with sweet flag extract, only 20.4 adults emerged which were significantly lower than 44.9 recorded in grains treated with neem extract. In turmeric extract 51.0 adults emerged which were significantly higher than those in neem and sweet flag extracts (Table II). This may be due to the ovicidal action of the active component "asarone" of sweet flag as shown by Schmidt and Risha (1990) and Zaidi *et al*. (2004).

Table I.- Mean number of *S. cerealella* adults emerged in wheat treated with various application rates of plant extracts in different solvents.

Plant	Application rate (µg/g)	Number emerged in different solvents					
		Pet. ether		Acetone		Ethanol	
Sweet flag	1000	0.0	e	0.0	e	53.7	bc
	500	0.0	e	0.0	e	56.0	abc
	250	0.0	e	0.7	e	61.0	abc
	125	0.3	e	7.7	d	65.7	ab
Neem	1000	25.7	d	53.3	abc	40.3	d
	500	28.0	cd	53.3	abc	49.3	bcd
	250	36.7	bc	56.3	abc	49.3	cd
	125	44.3	b	52.3	abc	49.7	bcd
Turmeric	1000	40.3	bc	39.0	c	47.7	cd
	500	44.3	b	43.7	bc	55.0	bc
	250	49.0	b	59.3	abc	59.0	abc
	125	51.0	b	61.3	ab	62.7	abc
Control		75.0	a	74.7	a	74.3	a
LSD value		11.9		10.9		12.8	

Each value is mean of 3 replications (number of eggs =100).
Values having same letters in a column are non-significant ($P \leq 0.05$).

Table II.- Mean number of *S. cerealella* adult progeny emerged from 100 eggs in wheat grains treated with various plant extracts.

Plant extract	Number emerged
Neem	44.9 ^{b}
Sweet flag	20.4 ^{c}
Turmeric	51.0 ^{a}
LSD value	3.3

Each value is mean of 3 solvents (petroleum ether, acetone, ethanol); 4 concentrations (1000, 500, 250, 125 µg/g) and 3 replications; Values having same letters in a column are non-significant ($P \leq 0.05$).

Extracts prepared in different solvents depicted that plant extracts prepared in petroleum ether were the most effective (26.6) as compared with those in acetone and ethanol (35.6 and 54.1, respectively) (Table III).

Adult emergence of *S. cerealella* from grain treated with various plant extracts was dependent on application rate of plant extracts (Table IV). Comparatively lower adult emergence (33.3) was at application rate of 1000 µg/g as compared with 36.6, 41.3 and 43.9 adults in grain treated at 500, 250 and 125 µg/g, respectively. Emergence at 1000 µg/g was significantly lower than that at 250 and 125 µg/g but not from that at 500 µg/g. At application rate of 500 µg/g or less, 50 % reduction in application rate caused significant reduction in adult emergence.

Table III.- Mean number of *S. cerealella* adult progeny emerged from 100 eggs in wheat grains treated with various solvents.

Solved	Number emerged
Petroleum ether	26.6 ^{c}
Acetone	35.6 ^{b}
Ethanol	54.1 ^{a}
LSD value	3.3

Each value is mean of 3 plants (neem, sweet flag, turmeric); 4 concentrations (1000, 500, 250, 125 µg/g) and 3 replications; Values having same letters in a column are non-significant ($P \leq 0.05$).

Table IV.- Mean number of *S. cerealella* adult progeny emerged from 100 eggs in wheat grains treated at different application rates.

Application rate (µg/g)	Number emerged
1000	33.3 ^{c}
500	36.6 ^{c}
250	41.3 ^{b}
125	43.9 ^{a}
LSD value	3.8

Each value is mean of 3 plants (neem, sweet flag, turmeric); 3 solvents (petroleum ether, acetone, ethanol) and 3 replications; Values having same letters in a column are non-significant ($P \leq 0.05$).

On the basis of adults emerged, all the three plant extracts and solvents were significantly different from each other. Application rate of 1000 and 500 µg/g were also significantly different from others (Tables II, IV).

Interaction of various plant extracts into different application rates showed significantly lower (17.9) *S. cerealella* adults at 1000 µg/g of sweet flag extracts which were significantly lower than those emerged from the grain treated with different application rates of neem or turmeric (Table V). It was also significantly different from its application rate of 125 µg/g. However, adults emerged at 500 and 250 µg/g of sweet flag were not significantly lower than at 1000 µg/g. Neem extracts comparable at all application rates allowed lower number of adults than those in turmeric.

Table V.- Mean number of *S. cerealella* adult progeny emerged from 100 eggs in wheat grains treated with various plant extracts at different application rates (plant extract and application rate interaction).

Plant extract	Mean emergence at different application rates (µg/g)			
	1000	500	250	125
Neem	39.8 ^{d}	43.6 ^{cd}	47.4 ^{bc}	48.8 ^{abc}
Sweet flag	17.9 ^{f}	18.7 ^{f}	20.6 ^{f}	24.6 ^{e}
Turmeric	42.3 ^{cd}	47.7 ^{abc}	55.8 ^{ab}	58.3 ^{a}
LSD value		6.6		

Each value is mean of 3 solvents (petroleum ether, acetone, ethanol) and 3 replications; Values having same letters in columns and rows are non-significant ($P \leq 0.05$).

Table VI.- Mean number of *S. cerealella* adult progeny emerged from 100 eggs in wheat grains treated with various plants extracted in different solvents (plant and solvent interaction).

Plant	Mean emergence at different application rates (µg/g)		
	Petroleum ether	Acetone	Ethanol
Neem	33.7 ^{c}	53.8 ^{ab}	47.2 ^{b}
Sweet flag	0.1 ^{e}	2.1 ^{d}	59.1 ^{a}
Turmeric	46.2 ^{b}	50.8 ^{ab}	56.1 ^{a}
LSD value		5.7	

Each value is mean of 4 concentration (1000, 500, 250, 125 µg/g) and 3 replications; Values having same letters in columns and rows are non-significant ($P \leq 0.05$).

Interaction of plant extract into solvent revealed that petroleum ether extracts of all the plants were most effective growth inhibitors allowing minimum number of *S. cerealella* adults to emerge. It was also found that petroleum ether extract of sweet flag was the most effective growth inhibitor. It allowed only 0.1 adults to emerge which were significantly lower than those recorded in other treatments. It was followed by 2.1 adults in its acetone extract which was also significantly lower than all other treatments (Table VI). Similarly in petroleum ether extract of neem, 33.7 adults emerged which were also significantly lower than rest of the treatments except petroleum ether and acetone extract of sweet flag. However, ethanol extract of sweet flag was the least effective showing 59.1 adults. It was not significantly different from acetone extract of neem and turmeric and ethanol extract of turmeric.

An interaction of solvent and application rate indicated that emergence of adults was minimum at 1000 µg/g of all the solvent extracts and all other comparable concentrations. Petroleum ether extracts showed significantly lower emergence of adults at each application rate followed by acetone and ethanol extracts. Minimum number of 22.0 adults emerged in the grains treated with petroleum ether at 1000 µg/g. It was followed by 24.1, 28.6 and 31.9 at 500, 250 and 125 µg/g respectively. In acetone and ethanol number of adults emerged were higher than petroleum ether at the comparative application rate (Table VII).

DISCUSSION

Petroleum ether extracts are the most promising growth inhibitor because of being highly volatile due to low polarity of compounds. It revealed that the vapors were ovicidal, rather than having lethal effect on other developmental stages. Such action of sweet flag oil had also been recorded by Yadava (1971) and Rahman and Schmidt (1999). Insects exposed to sweet flag oil vapors can lay immature eggs which could not hatch. The effect of petroleum ether extract of sweet flag was comparatively more visible on earlier stages (larvae) of metamorphosis. Conversely, the effect of ethanol extract of neem was slightly better shown up on later stages (pupation and adult emergence). This phenomenon may be due to the presence of asarone in petroleum ether extract of sweet flag and azadirachtin in ethanol extract of neem which are the active components. Direct effects of azadirachtin are on cells and tissues and indirect effects are exerted via the endocrine system. Neem seeds contain many related triterpenoids in addition to azadirachtin including 3-Tigloyl-azadirachtol (azadirachtin B), nimbin and salanin. Studies with several insects indicated that IGR effects are induced by azadirachtin applications. It is known that neem interferes with many life processes. In the adult (Schmidt and Pesel, 1987; Saxena, 1989) during larval development (Sieber and Rembold, 1983) and during metamorphosis of insects (Schluter *et al.*, 1985; Koul *et al.*, 1987).

Table VII.- Mean number of *S. cerealella* adult progeny emerged from 100 eggs in wheat grains treated with various solvents at different application rates (solvent and application rate interaction).

Solvent	Mean emergence at different application rates (µg/g)			
	1000	**500**	**250**	**125**
Petroleum ether	22.0 ^{g}	24.1 ^{fg}	28.6 ^{ef}	31.9 ^{de}
Acetone	30.8 ^{ef}	32.3 ^{e}	38.8 ^{d}	40.4 ^{c}
Ethanol	47.2 ^{b}	53.4 ^{ab}	56.4 ^{ab}	59.3 ^{a}
LSD value	6.6			

Each value is mean of 3 plants (neem, sweet flag, turmeric) and 3 replications; Values having same letters in columns and rows are non-significant (P ≤ 0.05)

With the current thrust on sustainable agriculture and organic farming, the use of natural products has assumed greater practical significance at global level. All these plants are used for medicinal purposes and as spices in daily food in Pakistan and some other countries. They are not likely to leave harmful effect on the commodity. In the present global situation, there is advocacy of lesser use of pesticides, depending upon the circumstances, such as the extent and severity of the pest, stage of host, availability and effectiveness of pesticide, application methods and use of either botanical pesticide or traditional pesticide in an integrated manner. Extracts of plants effects the growth of the pest species and this has been well documented. Their development to be used at mass scale is still limited. More studies are required for their efficient and economical use in the country.

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