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Insecticidal and antifeedant effect of *Pedalium murex* Linn. root and on *Spodoptera litura* (Fab.) (Lepidoptera: Noctuidae)

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Impact of ethanol extract of *Pedalium murex* (Linn) (Family: Pedaliaceae) root (0.1,0.2, 0.4, and 0.8%) were screened for its antifeedant and insecticidal activities against third, fourth and fifth instar larvae of *Spodoptera litura* (Fab.) by leaf-dip method. The larval mortality more than 50 percent at higher concentration (0.8%) was observed in the ethanol root extract. Stage dependant LC_{50} value was observed for *S. litura* (0.100, 0.118 and 0.258% for third, fourth and fifth nymphal instars). *P. murex* reduced the food consumption index, growth rate, approximate digestability, eficiency of conversion of ingested food, efficiency of conversion of digested food of *S. litura* indicating the antifeedant activity of this plant. Qualitative analysis of *P. murex* root extract revealed that it contains phytochemical such as, steroids, terpenoids, phenolics, saponines, tannins and flavanoids. Phenol, 2-(5,6-dimethyl pyrazinyl) methyl (molecular weight 214); O-Terphenyl-13C (molecular weight 230) and 3,3A, 4,9B-Tetrahydro-2H-Furo(3,2-C)(1) Benzopyran (molecular weight 206) were identified from the ethanol root extract of *P. murex* by using GC-MS. *P. murex* impact was more than the neembased biopesticide neemgold. Hence this plant can be explored as biopesticidal plant in the near future.

Key words: *Pedalium murex, Spodoptera litura,* insecticidal, antifeedant, phytochemical analysis

Introduction

Spodoptera litura (Fab.) (Lepidoptera: Noctuidae) is a polyphagous insect pest widely distributed throughout Asia (Hadapad *et al*., 2001). It has a wide range of host, feeding on 112 species world wide, of which 40 species are known from India (Singh *et al*., 1998 and Paulraj, 2001). Traditional farmers have been practicing synthetic pesticides to eliminate *S. litura* and hence it has developed resistance against almost all the commonly using pesticides in this aerea. Human health problem and environmental hazards caused by the

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indiscriminate use of chemical pesticides during past three decades have leads to the scientists to look for less persistent and biodegradable alternatives (Muraleedharan and sheeladevi, 1992; Mehrotra, 1992 and Sahayaraj *et al*., 2003). For this purpose, medicinal as well as weed plants that have been occasionally attacked by the pests were screened and are being reported to contain bio-pesticidal property (Selvaraj and Sahayaraj, 2005). These novel bioactive compounds isolated from the insectidial plants have been integrated in the Biointensive Integrated Pest Management (BIPM) programme for many crops. Biological, physiological and biochemical impact of many insecticidal plants on different insect pests has been reported by many authors. Pedaliaceae such as *Seasamum orientale* Linn. and *S. indicum* Linn. have been used as insecticidal plant against green gram pulse beetle *Callosobruchus chinensis* Linn. and *Sitophilus oryzae* Linn. (Sujatha and Punniaiah, 1985; Choudhary, 1990). This study was aimed to found out the insecticidal activity of *P. murex* root ethanol exract on *S. litura* third, fourth and fifth instar larvae.

Materials and methods

Collection and rearing of pest

Egg masses and larva of *S. litura* were collected from groundnut and castor fields in and around Tirunelveli district, Tamil Nadu, India. Collected leaves with egg masses were transfered on to the filter paper and kept in petridishes under laboratory conditions (27^0 C $\pm$ 1^0 C temperature; 65 - 70% RH, 11 L : 13 D). Newly hatched first instar larva were reared in plastic trough (28 x 21 x 9 cm) on castor leaves. Laboratory emerged third, fourth, and fifth instar larvae (<3 hours old) of *S. litura* were used for this experiment.

Collection and extraction of plants

Pedalium murex (L.) (Family: Pedaliaceae) was chosen for the present study. It was collected from St. Xavier's College campus, Palayamkottai, Tamil Nadu, India. Collected plants were washed thrice in tap water and once in 0.5% of sodium hypochloride and distilled water subsequently. Roots were removed from the plant and were shade dried for two weeks. The plants were powdered in a domestic grinder and stored in refrigerator for further use. From the stock, 250 grams of powder was extracted with 500 ml of ethanol using soxhelet apparatus for about 24 hrs. Ethanol extracts were concentrated using a distillation unit, air dried and stored at -4^0C for further experimental purpose.

Preparation and treatment

One gram of the crude ethanol extract of *P. murex* was dissolved in 5 ml of water. After thorough mixing the extract was again mixed with 95 ml of water. This extract was treated as 5% plant extract and used for the preliminary range finding tests and also to detect the concentrations of extracts which causing 100% larval mortality. Based on this preliminary concentration, we have prepared different concentrations such as 0.1, 0.2, 0.4 and 0.8% and used for this studies.10g castor leaves were soaked in 0.1% of *P. murex* extract for five minutes. Control leaves were soaked in water. After five minutes, leaves were air dried for another 5 minutes and were supplied to the pests. Five weighed third, (less than 3 hours old) instar larvae of *S. litura* were released into the plastic vials (500 ml capacity). *P. murex* ethanol extract and water treated castor leaves were provided to experimental and control category of *S. litura* respectively. Then the vials were covered with muslin cloth for aeration. Similar procedure was also followed for other concentrations such as 0.2, 0.4 and 0.8% and also for fourth and fifth instars. Six replications were made for each concentration, standard and control. For standard a neembased hebal pest repellent/antifeedant insecticide Neemgolad (SPIC, Thoothukudi, Tamil Nadu, India) was used at field concentration (3%). The larvae were allowed to feed the treated leaves for a period of 96 hours continuously. After 72 and 96 hours of treatment, number of larvae died was recorded. By using this data, LC_{50}, chi-square value, regression equation, lower and upper fiducial limits were calculated using (Finney, 1971) formula. Furthermore, any change in the morphology of the larvae was also noticed.

Antifeedant study

Energetics was considering as- tool for antifeedant activity 96 hours LC_{50} concentration was diluted 10 times with water. 10 g castor leaves were treated with the same; shade dried and provided to the 3 hours old third instar leaves for four days continiously. After every 24 hrs of treatment, unconsumed leaves, faecal pellets and larval weight were taken with the help of monopan balance (Dhona). Similar parameters were recorded after 48, 72 and 96 hrs of the experiment. Similar procedure was followed for fourth and fifth instars of *S. litura* with control (water), standard (neemgold) and experimental plant. In order to findout the dry weight of the castor leaves, fresh leaves were weighed and they were placed in an oven at 50^0 C. After 12 hrs, the dry weight of the leave was noted. Similar procedure was also followed for the faecal pellets and

S. litura larva. Procetures and formula were used to study energetics (WALDBAURI, 1968).

Phytochemical analyses of Petalium murex root extract

Steroids, alkaloids, reducing sugar, phenolic compounds, saponins, zanthoproteins, tannins and flavonoids were tested using Brindha *et. al*., (1981). The ethanol extracts of *P. murex* was subjected to the compound identification using HPLC, one mg of ethanol extract was dissolved in 1 ml HPLC grade imported methanol and the required quantity of sample was injected into the HPLC unit. The spectrum and their mass peak were compared and identified with the Tutor and Wiley library records. Student 't' test was performed to found out the significance between control to standard and *P. murex* treatments. The significance was expressed at 5% level.

Result and discussion

Phytochemistry

Preliminary phytochemical analysis of *P. murex* ethanol extract showed the presence of reducing sugars, phenolic compounds, saponins, xanthoprotein, alkaloids, triterpenoids, tannins and flavanoids. (Srinivasa *et al*., 1999, Suganthy, 2000, Sundararajan and Ananthakrishnan, 2002) were used ethanol as a solvent for the extraction of different secondary metabolites of plants. Since the polarity of ethanol is higher, most of the secondary metabolits of *P. murex* dissolved in ethanol. Saponins and their derivatives inhibit the larval growth and development (Suresh *et al.,* 2002). Furthermore tannine combine with protein inhibite the enzyme activity and reduce the availability of protein in haemolymph insect (Chan *et al.,* 1982). Insecticidal activity of *P. murex* might be due to the presence of saponins and tannins present in this extract. HPLC analyses revealed that *P. murex* ethanol extract consists of theree major compounds such as phenol, 2-(5,6-dimethyl pyrazinyl) methyl (Molecular weight 214); O-Terphenyl-13C (Molecular weight 230) and 3,3A,4,9B-Tetrahydro-2H-Furo(3,2-C)(1)Benzopyran (Molecular weight 206). More studies are essential to test these compounds either individually or in combination and recommend them for industrial usage.

Median lethal concentration

LC_{50} values of *S. litura* with *P. murex* treated castor leaves are presented in Table 1. It showed that *S. litura* with higher concentration of *P. murex* (above 0.4%) died at the early period of the treatment. But those animals,

Table 1. Impact of *Pedalium murex* root ethanol extract on the LC_{50} regression equation, variance, chi-square, lower (LFL) and upper fiducial limit (UFL) on *S. litura* third, fourth and fifth instar larvae of *Spodoptera litura.*

Duration (in Hrs.)	Life stages	Regression equation	LC_{50}	Vari-ance	Chi-square	LFL	UFL
72	Third instar	Y = 1.339 x + 5.51	0.042	0.0975	1.14	0.032	0.238
	Fourth instar	Y=1.216 x + 4.13	0.052	0.1145	0.62	0.041	0.170
	Fifth instar	Y = 0.877 x + 4.69	0.244	0.0336	1.32	0.103	0.372
96	Third instar	Y=1.578 x + 5.0	0.100	0.031	0.61	0.021	0.219
	Fourth instar	Y=2.336 x + 4.83	0.118	0.019	2.08	0.003	0.378
	Fifth instar	Y=0.937 x + 4.61	0.258	0.027	0.75	0.011	0.218

which fed with lesser concentration (below 0.2%), failed to complete the moulting and died either in the larvae or in pupae. Those fed with least concentration (0.1%) were transformed into normal adults. But some of the larvae were failed to normal growth and development. Because, haemolymph was expelled out from the *S. litura* larvae and they died after two to four hours of haemolymph expelsion. From the result it was very clear that LC_{50}value was life stage dependent factor. For instance LC_{50} value for third instar *S. litura* was 0.100% and it gradually increased when the pest grew older (0.118 and 0.258% for fourth and fifth instars respectively).

Antifeedant activity by energetics

Food consumption was higher in control and lower in *P. murex* treated castor leaves fed *S. litura* larvae. As observed for the food consumption, the growth rate was also higher in control as well as in third instars. It was gradually decrease when *S. litura* grew older. Among all the categories the growth rate was very minimum at 0.8% *P. murex* root ethanol extract. From the result it is very clear that the nutritional requirements of *S. litura* differs when it was provided with either neemgold treated castor leaves or *P. murex* treated leaves. Irrespective of the life stages, food consumption index was higher in control followed by neemgold and *P. murex* (Table 2). It was higher in third instar and gradually decreased when the pest grew older. As observed for third instar the approximate digestability was also higher in third instar

followed by fourth and fifth instar. This parameter is an indirect indication of the amount of food converted into body biomass. Food on which insect's greater body weight is always classified as better source of energy (Ananthakrishnan, 1992). According to this hypothesis our study also revealed that *S. litura* fed *P. murex* was highly reduce the body weight. The comparison between ECI of control and *P. murex* were statistically significant at 5% level in all the life stages of *S. litura*. Earlier (Waldbaur, 1968) reported ECI values rise and fall with AD. It could be conducted that *P. murex* treated food was quickly converted to body substance in response to the various bioactive components of *P. murex* to *S. litura*. In insects that develop eggs following adult ecdysone, vitellogenesis is often triggered in response to food intake. In response dietary cues, vitellogenesis is synthesized by fat body, transported in haemolymph, endocytosed by the ovarian follicle and incorporated as yolk protein (vitellin) in to developing oocytes. We have hypothesized not initial magnitute of synthetic response to diet is determined by dietary quality. Control treatment reaches higher ECD values as comparatively to neem biopesticide and *P. murex* ethanol extract. This study clearly revealed that *P. murex* highly reduces the food consumption index, approximate digestability, growth rate, efficiency of conversion of ingested food and efficiency of conversion of digested food. Hence *P. murex* root extract can be explored in *S. litua* management.

Table 2. Impact of *Pedalium murex* root ethanol extract on food consumption index (FCI), growth rate (GR) and aproximate digestability (AD) (mg dry weight/ day), efficiency of conversion of ingested food (ECI) and efficiency of conversion of digested food (ECD) (%) on third, fourth and fourth instar larvae of *Spodoptera litura*.

Treatments	FCI	GR	AD	EC	ECD
			Third Instar		
P. murex	23.18 ± 6.85	19.36 ± 2.85	1.05 ± 0.02	3.56 ± 0.62	3.35 ± 0.94
Neemgolad	31.10 ± 10.85	25.12 ± 3.83	1.2 ± 0.26	4.49 ± 0.34	6.46 ± 1.19
Control	47.08 ± 12.86	31.2 ± 6.22	15.5 ± 0.60	6.41 ± 0.39	7.33 ± 0.94
			Fourth Instar		
P. murex	19.68 ± 7.58	18.20 ± 1.90	29.95 ± 2.09	11.30 ± 4.01	10.55 ± 4.04
Neemgolad	31.82 ± 8.15	14.3 ± 0.76	38.2 ± 4.23	10.86 ± 4.58	15.68 ± 5.48
Control	37.50 ± 10.82	17.05 ± 1.91	41.0 ± 3.74	19.43 ± 6.05	22.77 ± 6.32
			Fifth Instar		
P. murex	8.69 ± 0.76	9.79 ± 1.21	6.02 ± 10.34	9.17 ± 2.97	8.74 ± 4.89
Neemgolad	10.39 ± 0.37	11.16 ± 27.27	10.5 ± 7.82	7.72 ± 1.93	15.68 ± 4.69
Control	11.21 ± 0.67	12.46 ± 16.45	11.82 ± 8.42	19.28 ± 3.75	22.79 ± 6.82

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