International Journal of Entomology Research
ISSN: 2455-4758
Impact Factor: RJIF 5.24
www.entomologyjournals.com
Volume 3; Issue 2; March 2018; Page No. 130-138

Oviposition deterrent, repellent and ovicidal activity of *Pterolobium hexapetalum* (Fab.) against the stored grain pest, *Callosobruchus maculatus* (Coleoptera: Chrysomelidae)

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Abstract
The oviposition deterrent, repellent and ovicidal activities of *P. hexapetallum* against the stored pest - *Callosobruchus maculatus. P. hexapetalum* leaves was examined with increasing polarity of different organic solvents like hexane, dichloromethane and ethanol at various concentrations of 200-600 ppm against *C. maculatus* under the laboratory conditions. After 24 hours there was a significant activity noted against *c. maculatus* but the highest activity was detected after 72 hours in the ethanol extract for the concentration of 600ppm. The remaining two organic solvents also exhibited oviposition deterrent, repellent and ovicidal activities but ethanol portrayed the peak activity. Therefore, *P. hexapetalum* has the best phytopesticidal constituents to treat against the stored pest and it also creates no harm to the environment because of its easy biodegradable and non-toxic nature.

Keywords: *Callosobruchus maculatus, Pterolobium* hexapetalum, oviposition deterrent activity, repellent activity, ovicidal activity, phytopesticidal

1. Introduction
Insects often cause serious menace to stored food grains throughout the world rather than any other agents like fungi, moulds, bacteria etc. It has been estimated that, insects cause heavy loss in weight and results in very bad quality and also reduce the quantity of products [1]. Cowpea – *Vigna unguiculata* is one of the most important crops which are being used daily in our day to day life.
This cowpea can be easily damaged continuously by the pulse beetle, *Callosobruchus maculatus* (*C. maculatus*). *C. maculatus* is also termed as pulse beetle because it is the host for the variety for beans such as cowpea, mungbean, Bambara, ground nuts; leguminous seeds like chick pea, green gram, red gram, soya bean, lentil, pea, peanut and haricot beans. *C. maculatus* may be observed abundantly in the cowpea seeds at the time of storage and remaining seeds are being infested throughout the tropics [2].The crop is susceptible to variety of pests in the field and this *Callosobruchus maculatus* infestation starts in the field itself but its activity will be more at the time of storage. 90% of the cowpea seeds were damaged by *C. maculatus* and within 3-5 months of storage there will be 100% infestation [3].
These cowpea weevil's infestation on stored grains prevent germination of seeds because initially in the field itself eggs were laid and was firmly deposited on the pod and at the time of storage the eggs deposition take place directly on the seed and will create a small hole on the seed to pierce inside and complete its remaining life stages. Finally, emerging out as a complete matured adult insect. Seed coat or testa is one of the most important factors for oviposition stimulation. Adults do not feed on seeds, but gravid female starts laying eggs on the seed (4). Within two days *C. maculatus* reaches maximum eclosion. Female *C. maculatus* has the potency to lay about 100 eggs in its life time [5]. Also, the egg hatches into the seed only after the oviposition of 6 days [6]. The average life span of adult male and female *C. maculatus* is 7 days and under rare condition it will be two weeks [7]. Since, this bruchid activity is vital and consistently destroying the agriculturally stored products which results in the great loss in the commercial value of seeds. Therefore, in the present context an immediate and most stimulated action has to be achieved to prevent a enormous loss to our farmers. Under this situation, the activity of *C. maculatus* was controlled by synthetic insecticides and fumigants. But these, synthetic insecticides resulted in serious threat to environment, cost of application is high, non biodegradable quality, moreover, the pest started developing resistance and also application of synthetic pesticides results in lethal effects to non-target organisms [8]. Therefore, the present study hampers the tempo of exploration of green pesticide as an emergency need to develop safer and eco-friendly environment.

2. Materials and Methods
2.1. The preliminary qualitative phytochemical screening was followed by the methodology of Harborne and Kokate [9-10].

2.2. Collection of plant material: Fresh leaves of the plant *Pterolobium hexapetallum* was collected from Javadhi hills, Vellore district, Tamil Nadu. The collected plant was stored in a zip lock cover to prevent the effect of humidity and evaporation. The collected leaves were washed thoroughly with tap water, shade dried and ground to a fine powder with an electric blender.

2.3. Preparation of plant extracts: The extraction of plant sample was done with three different solvents: hexane,

dichloromethane and ethanol. About 100g of sample was taken and soaked with 500 ml of hexane solvent for 24 hours in a brown glass bottle and kept in a rotator shaker for continuous shaking. After the completion of 24hours, the filtration was carried out in a Whatman No.1 filter paper. The filtrates were then placed in a rotary evaporator and the boiling points were maintained separately for each solvent with a rotary speed of 3-6 rpm for 2 hours. The crude obtained from the evaporator is again allowed to air dried to remove traces of hexane solvent. Then, the crude is stored in a brown vial for further study. Now, the residue left in the filter paper was shade dried for complete evaporation of the solvent and then it is allowed to extract with Dichloromethane and followed by Ethanol. The crude of all the extracts were stored in brown vials and kept in a refrigerator.

2.4 Culture of *Callosobruchus maculatus*: To rear *C. maculatus* in the laboratory, infested chickpea grains (host) were collected from the provision stores and it is introduced into the uninfected grains which were stored in a transparent airtight plastic container. A part of the cap of the container is covered with muslin to provide proper ventilation and the beetles were reared on grains for about a year (approximately 10 generations) prior to the experiments. One-two day's old F10 generation insects were used for the study.

2.5. Bioassay

2.5.1. Repellent activity of different solvent extracts of *Pterolobium hexapetallum* against *Callosobruchus maculatus*: To study the repellent activity of hexane, dichloromethane and ethanol extract of *P. hexapetalum* against the stored pest, an experimental system consisting of 7 airtight plastic box (3 plant extract treatments, 3 control and 1 centre box). Aeration was provided on the cap of the each box. Each box was connected to the centre box by means of 10 cm tube with a diameter of 2.5mm. Twenty pairs of adult insects were introduced into the centre box. Different concentration of 200ppm, 400ppm and 600ppm of plant extracts were mixed separately with the 100 chickpea and kept undisturbed for 5 minutes for the solvent to evaporate completely. The similar setup was replicated for five times and the same procedure was carried out for the remaining two extracts: dichloromethane extract and ethanol extract of *P. hexapetallum.* Neem azal was used as a control. After 24, 48 and 72 hrs, the contents of beetles at each treated and control was counted and the repellency (%) was calculated by Lwande's method [11].

$$\text{Repellency (\%)} = \frac{C - E}{T} X100$$

Where C is the insect numbers in the negative control jar,
E is the insect numbers in extract treated jar and
T is the number of total insects.
C, E and T were the mean data of 5 replicates.

2.5.2 Oviposition deterrent activity of different solvent extracts of *Pterolobium hexapetalum* against *Callosobruchus maculatus*: To perform this activity 20 black gram seeds treated with different concentrations of 200, 400 and 600 ppm were introduced into the small plastic box and aeration was provided on the cap of the each box. For each concentration five replicates were maintained. Neem azal was used as a control. Also, make sure that the treated seeds are coated evenly and dried well before introducing in to the plastic box. Then, 5 pairs of newly emerged *C. maculatus* were introduced into the box and closed. After 24, 48 and 72 hrs, the numbers of eggs laid on the control and on the treated grain were recorded and the percentage of oviposition deterrence was calculated by Abbott's corrected formula [12].

$$\text{POD} = \frac{Ts - Cs}{Cs} X100$$

Where POD is the percentage of oviposition deterrence
C is the insect numbers in the negative control jar,
T is the insect numbers in extract treated jar and
C and T were the mean data of 5 replicates.

2.5.3 Ovicidal activity of different solvent extract of *Pterolobium hexapetalum* against the eggs (24 h old) of *Callosobruchus maculatus:* The freshly laid eggs of *C.maculatus* were immersed in the different plant extracts of different concentrations (200, 400 and 600ppm) and the seeds were immersed for 10 seconds. Then, they were allowed to dry for few minutes and introduced separately for different concentrations into the medium sized boxes. The exposure period was 72 h. The final mortality counts were made after 11 days with the help of a hand lens. Unhatched eggs with black spots inside were considered as dead and readings were recorded. The data obtained from the present experiment was subjected to the following formula to derive the ovicidal activity of the selected plant extracts. Percentage mortality was calculated, and data were corrected for natural mortality in controls using the Abbott (1925) formula [12]. The corrected mortality was then subjected to probit analysis to estimate LC_{50} and LC_{90} values [13]. Analysis of variance (ANOVA-Two Way) was used to determine the effect of solvent extract concentrations on ovicidal activity. Following a significant ANOVA, differences amongst means were established using Least Significant Difference (LSD) test at 0.05% level.

$$\%\text{Ovicidal Activity} = \frac{\text{No. of eggs hatched}}{\text{Total no. of eggs treated}} X\ 100$$

$$\text{Corrected Mortality} = \frac{\%\text{ of mortality in treated group} - \%\text{ of } mortality \text{ in control group}}{100 - \%\text{ of mortality in control group}} X100$$

3. Results

3.1 Phytochemical screening of hexane, DCM and ethanol extracts of *P. hexapetalum*: The phytochemical screening of *P. hexapetallum* of different solvent extracts was assessed and the results pertaining to the experiments are shown in table 1. Hexane extract showed the presence of alkaloid, terpenoids, carbohydrate, anthoquinone and coumarins. Similarly, dichloromethane extract exhibited the presence of alkaloid, phenols, flavonoids, tannins, terpenoids, carbohydrate,

anthoquinone, coumarins and protein. Whereas, the presence of alkaloids, phenols, flavonoids, anthroquinones, coumarins, terpenoids and steroids were noticed in ethanol extract.

Table 1: Qualitative analysis of phytochemical in different solvent extracts of *Pterolobium hexapetallum*

S. No	Phytochemical groups	Extracts tested		
		Hexane extract	Dichloromethane extract	Ethanol extract
1	Alkaloids	+	+	+
2	Phenols	-	+	+
3	Flavanoids	-	+	+
4	Tannins	-	+	-
5	Terpenoids	+	+	+
6	Saponins	-	-	-
7	Carbohydrate	+	+	+
8	Glycosides	-	-	+
9	Anthoquinone	+	+	+
10	Coumarins	+	+	+
11	Steroids	-	-	+
12	Protein	-	+	-
13	Acid	-	-	+

(+): Presence; (-): Absence

Table 2: Oviposition deterrent activity of hexane extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*

Concentrations tested (ppm)	Exposure periods (in Hrs)		
	24 Hrs	48Hrs	72Hrs
	Oviposition deterrent activity (%)		
200	$14.00\pm0.00^{a(21.97)}$	$18.6\pm0.55^{a\ (25.55)}$	$26.00\pm0.00^{a\ (30.66)}$
400	$23.00\pm0.71^{b\ (28.66)}$	$32.6\pm0.55^{b\ (34.82)}$	$42.00\pm0.00^{b\ (40.4)}$
600	$38.00\pm0.00^{c\ (38.06)}$	$44.00\pm0.71^{c\ (41.55)}$	$66.00\pm1.00^{\ (54.33)}$
Neem azal	$76.66^{d\ (61.07)}$	$83.66^{d\ (66.11)}$	$92.66^{d\ (74.21)}$

Values expressed are mean of five replications (n=250). Parentheses hold angular transformed values. Different alphabet in the column shows statistical significance at *P<0.05*% level; LSD, Duncan Multiple Range Test (DMRT).

3.2 Oviposition deterrent activity of hexane extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*

The oviposition deterrent activity of hexane extract of *P. hexapetalum* was tested with different concentrations such as 200, 400 and 600ppm with different exposure periods such as 24, 48 and 72 hrs against the eggs of *C. maculatus*. The data pertaining to the above experiments are shown in table 2. The data revealed that the oviposition deterrent activity of the plant extract are directly proportional to the concentration and exposure periods, *i.e.*, the oviposition deterrent activity was observed increased with the increase in concentration and exposure periods. Perusal of the data clearly indicates that at 24hrs exposure periods, hexane extract of the selected plant produced the maximum oviposition deterrent effect at 600ppm concentration with 38.00% oviposition deterrent activity of, later, it was followed by 400 and 200ppm concentrations of the plant extract with an activity of 23.00 and 14.00% respectively. Though the activities noted against each concentration varied, the oviposition deterrent activity showed statistically significant values *p<0.05%;* ANOVA, Duncan Multiple Range Test (DMRT) – table 2.

In the same way, seeds treated with the hexane extract of *P. hexapetalum* and exposed to 48hrs of exposure period, produced the maximum oviposition deterrent effect at highest concentration (600ppm) with an oviposition deterrent activity of 44.00% followed by 400 and 200ppm concentrations of the plant extract with an activity of 32.6 and 18.6% respectively. The activities noted against each concentration varied and differed statistically (*p<0.05*%; ANOVA, DMRT – table 2). Besides, *C. maculatus* exposed to hexane extract of *P. hexapetalum* at 72hrs exposure periods produced the maximum oviposition deterrent activity of 66.00% at 600ppm concentration followed by 42.00%, 26.00% activity at 400 and 200ppm concentrations respectively.

Table 3: Oviposition deterrent activity of Dichloromethane extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*

Concentrations tested (ppm)	Exposure periods (in Hrs)		
	24 Hrs	48Hrs	72Hrs
	Oviposition deterrent activity (%)		
200	$10.6\pm1.00^{a\ (19)}$	$25.2\pm1.30^{a\ (30.13)}$	$33.0\pm0.00^{a\ (35.06)}$
400	$30.8\pm0.84^{b\ (33.71)}$	$37.2\pm0.45^{b\ (37.58)}$	$45.2\pm0.84^{b\ (42.25)}$
600	$36.8\pm0.45^{c\ (37.35)}$	$44.6\pm0.55^{c\ (41.9)}$	$72.6\pm0.89^{c\ (58.44)}$
Neem azal	$76.66^{d\ (61.07)}$	$83.66^{d\ (66.11)}$	$92.66^{d\ (74.21)}$

Values expressed are mean of five replications (n=250). Parentheses hold angular transformed values. Different alphabet in the column shows statistical significance at *P<0.05*% level; LSD, DMRT.

3.3 Oviposition deterrent activity of dichloromethane extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*

The oviposition deterrent activity of dichloromethane (DCM) extract of *P. hexapetalum* was tested with different

concentrations such as 200, 400 and 600ppm with different exposure periods such as 24, 48 and 72 hrs against the eggs of *C. maculatus*. The data pertaining to the above experiments are shown in table 3. The data revealed that the oviposition deterrent activity of the plant extract are directly proportional to the concentration and exposure periods, *i. e*., the oviposition deterrent activity was observed increased with the increase in concentration and exposure periods.

Perusal of the data clearly shows that at 24hrs exposure periods, DCM extract of the selected plant produced the maximum oviposition deterrent effect at 600ppm concentration with 36.8% oviposition deterrent activity of, later, it was followed by 400 and 200ppm concentrations of the plant extract with an activity of 30.8% and 10.6% respectively. Though the activities noted against each concentration varied, the oviposition deterrent activity showed statistically significant values *p<0.05%;* ANOVA, DMRT – table 3.

In the same way, seeds treated with the DCM extract of *P. hexapetalum* and exposed to 48hrs of exposure period, showed the maximum oviposition deterrent effect at highest concentration (600ppm) with an oviposition deterrent activity of 44.6% followed by 400 and 200ppm concentrations of the plant extract with an activity of 37.2% and 25.2% respectively. The activities noted against each concentration varied and differed statistically (*p<0.05*%; ANOVA, DMRT – table 3).

Besides, *C. maculatus* exposed to DCM extract of *P. hexapetalum* at 72hrs exposure periods produced the maximum oviposition deterrent activity of 72.6% at 600ppm concentration followed by 45.2, 33.00% activity at 400 and 200ppm concentrations respectively.

Table 4: Oviposition deterrent activity of ethanol extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*

Concentrations tested (ppm)	Exposure periods (in Hrs)		
	24 Hrs	48Hrs	72Hrs
	Oviposition deterrent activity (%)		
200	22.4 ± 0.55^{a} (28.25)	30.2 ± 0.45^{a} (33.34)	40.2 ± 1.10^{a} (39.35)
400	33.2 ± 0.84^{b} (35.18)	38.6 ± 1.14^{b} (38.41)	48.4 ± 0.55^{b} (44.08)
600	36.8 ± 1.48^{c} (37.35)	48.6 ± 1.14^{c} (44.2)	78.2 ± 0.45^{c} (62.17)
Neem azal	76.66^{d} (61.07)	83.66^{d} (66.11)	92.66^{d} (74.21)

Values expressed are mean of five replications (n=250). Parentheses hold angular transformed values. Different alphabet in the column shows statistical significance at *P<0.05*% level; LSD, DMRT.

3.4 Oviposition deterrent activity of ethanol extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*

The oviposition deterrent activity of ethanol extract of *P. hexapetalum* was tested with different concentrations such as 200, 400 and 600ppm with different exposure periods such as 24, 48 and 72 hrs against the eggs of *C. maculatus*. The data pertaining to the above experiments are shown in table 4. The data revealed that the oviposition deterrent activity of the plant extract is directly proportional to the concentration and exposure periods, *i. e*., the oviposition deterrent activity was observed increased with the increase in concentration and exposure periods.

Perusal of the data clearly shows that at 24hrs exposure periods, ethanol extract of the selected plant produced the maximum oviposition deterrent effect at 600ppm concentration with 36.8% oviposition deterrent activity. Of, later, it was followed by 400 and 200ppm concentrations of the plant extract with an activity of 33.2 and 22.4% respectively. Though the activities noted against each concentration varied, the oviposition deterrent activity showed statistically significant values *p<0.05%;* ANOVA, DMRT – table 4.

In the same way, seeds treated with the ethanol extract of *P. hexapetalum* and exposed to 48hrs of exposure period, produced the maximum oviposition deterrent effect at highest concentration (600ppm) with an oviposition deterrent activity of 48.6% followed by 400 and 200ppm concentrations of the plant extract with an activity of 38.6 and 30.2% respectively. The activities noted against each concentration varied and differed statistically (*p<0.05*%; ANOVA, DMRT – table 4).

Besides, *C. maculatus* exposed to ethanol extract of *P. hexapetalum* at 72hrs exposure periods produced the maximum oviposition deterrent activity of 78.2% at 600ppm concentration followed by 48.4, 40.2% activity at 400 and 200ppm concentrations respectively.

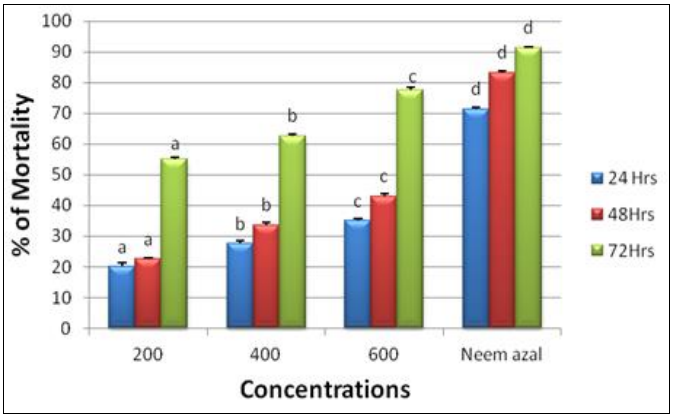

Fig 1: Repellent activity of hexane extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*

3.5 Repellent activity of hexane extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*

The repellent effect of hexane extract of *P. hexapetalum* was tested with different concentrations such as 200, 400 and 600ppm with different exposure periods such as 24, 48 and 72 hrs against the *C. maculatus* adults. The data pertaining to the above experiments are shown in figure 1. It was observed that the repellent activity of the plant extract is directly proportional to the concentration and exposure periods, *i. e*., the repellent activity was observed increased with the increase in the concentration and exposure periods.

Analysis of the data pertaining to the repellent activity, clearly indicates that at 24hrs exposure periods, hexane extract of the selected plant produced the maximum repellent effect at 600ppm concentration with 33.50% repellent activity, later, it was followed by 400 and 200ppm concentrations of the plant extract with an activity of 29.00, and 23.50% respectively. Repellency noted against each concentration varied and also there were significant differences between the observed means (*p<0.05%;* ANOVA, DMRT).

Furthermore, experimental adults treated with the hexane extract of *P. hexapetalum* exposed to 48hrs, exhibited the maximum repellency at highest concentration (600ppm) with the repellent activity of 48.00% followed by 400 and 600ppm concentrations of the plant extract with an activity of 38.50 and 27.00% respectively. These activities noted were found differed statistically (*p<0.05*%; ANOVA, DMRT).

In the same way, *C. maculatus* exposed to hexane extract of *P. hexapetalum* to 72hrs exposure periods produced the maximum repellent activity of 75.00% at 600ppm concentration followed by 67.00 and 43% activity at 400 and 200ppm concentrations respectively. The repellent activity of *P. hexapetalum* (72hrs) are found to be statistically significant at *p<0.05%;* ANOVA, DMRT.

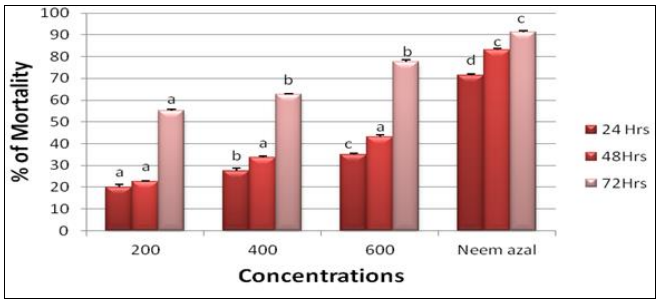

Fig 2: Repellent activity of Dichloromethane extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*.

3.6 Repellent activity of Dichloromethane extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*.

The repellent effect of DCM extract of *P. hexapetalum* was tested with different concentrations such as 200, 400 and 600ppm with different exposure periods such as 24, 48 and 72 hrs against the *C. maculatus* adults. The data pertaining to the above experiments are shown in figure 2. It was observed that the repellent activity of the plant extract is directly proportional to the concentration and exposure periods, *i. e.*, the repellent activity was observed increased with the increase in the concentration and exposure periods.

Analysis of the data pertaining to the repellent activity clearly indicates that at 24hrs exposure periods, DCM extract of the selected plant produced the maximum repellent effect at 600ppm concentration with 35% repellent activity, later, it was followed by 400 and 200ppm concentrations of the plant extract with an activity of 27.5 and 20% respectively. Repellency noted against each concentration varied and also there were significant differences between the observed means (*p<0.05%;* ANOVA, DMRT).

Furthermore, experimental adults treated with the DCM extract of *P. hexapetalum* exposed to 48hrs, exhibited the maximum repellency at highest concentration (600ppm) with the repellent activity of 43% followed by 400 and 600ppm concentrations of the plant extract with an activity of 33.50 and 22.5% respectively. These activities noted were found differed statistically (*p<0.05*%; ANOVA, DMRT).

In the same way, *C. maculatus* exposed to DCM extract of *P. hexapetalum* to 72hrs exposure periods produced the maximum repellent activity of 77.5% at 600ppm concentration followed by 62.5 and 55% activity at 400 and 200ppm concentrations respectively. The repellent activity of *P. hexapetalum* (72hrs) were found to be statistically significant at *p<0.05%;* ANOVA, DMRT.

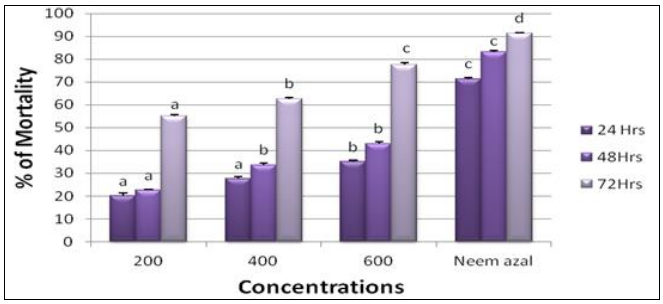

Fig 3: Repellent activity of ethanol extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*

3.7 Repellent activity of ethanol extract of *Pterolobium hexapetalum* tested against *Callosobruchus maculatus*

The repellent effect of ethanol extract of *P. hexapetalum* was tested with different concentrations such as 200, 400 and 600ppm with different exposure periods such as 24, 48 and 72 hrs against the *C. maculatus* adults. The data pertaining to the above experiments are shown in figure 3. It was observed that the ovicidal activity of the plant extract is directly proportional to the concentration and exposure periods, *i. e.*, the repellent activity was observed increased with the increase in the concentration and exposure periods.

Analysis of the data pertaining to the repellent activity, clearly indicates that at 24hrs exposure periods, hexane extract of the selected plant produced the maximum repellent effect at 600ppm concentration with 60.00% repellent activity, later, it was followed by 400 and 200ppm concentrations of the plant extract with an activity of 52.50, and 47.50% respectively. Repellency noted against each concentration varied and also there were significant differences between the observed means (*p<0.05%;* ANOVA, DMRT).

Furthermore, experimental adults treated with the ethanol extract of *P. hexapetalum* exposed to 48hrs, exhibited the maximum repellency at highest concentration (600ppm) with the repellent activity of 65.00% followed by 400 and 600ppm concentrations of the plant extract with an activity of 60.00 and 50.00% respectively. These activities noted were found differed statistically (*p<0.05*%; ANOVA, DMRT).

In the same way, *C. maculatus* exposed to ethanol extract of *P. hexapetalum* to 72hrs exposure periods produced the maximum repellent activity of 85.00% at 600ppm concentration followed by 70.00 and 63% activity at 400 and 200ppm concentrations respectively. The repellent activity of *P. hexapetalum* (72hrs) were found to be statistically significant at *p<0.05%;* ANOVA, DMRT.

Table 5: Lethal concentrations values for ovicidal activity of Hexane extract of *Pterolobium hexapetallum* tested against the stored pest – *Callosobruchus maculatus*.

Exposure periods (Hrs)	Concentrations (ppm)	% of Ovicidal activity	LC_{50}	95 % Fiducial Limit		LC_{90}	95% fiducial limit		χ^2
				LCL	UCL		LCL	UCL	
24	200	25.2±0.84[a]	685.57	543.41	1294.03	1644.18	1138.76	4238.33	0.10
	400	36.4±0.71[b]							
	600	44.8±0.55[c]							
48	200	30.8±0.71[a]	500.20	402.19	707.95	1373.23	1000.30	2859.81	1.21
	400	48.6±0.84[b]							
	600	53.6±0.45[c]							
72	200	50.2±1.10[a]	203.22	445.75	334.65	1299.63	898.38	4146.48	0.03
	400	58.4±0.55[b]							
	600	68.2±0.45[c]							

The value represents mean ± S. D. of five replications. *mortality of the larvae observed after 24h, 48h & 72 of the exposure period, WHO (2005). LC_{50}=Lethal Concentration brings out 50% Mortality and LC_{90} = Lethal Concentration brings out 90% mortality. LCL = Lower Confidence Limit; UCL = Upper Confidence Limit; Values in a column with a different superscript alphabet are significantly different at $P < 0.05$ (MANOVA; LSD -Tukey's Test).

3.8 Lethal concentrations values for ovicidal activity of Hexane extract of *Pterolobium hexapetallum* tested against the stored pest – *Callosobruchus maculatus*.

Ovicidal activity of hexane extract of *P. hexapetallum* was tested against the freshly laid eggs on the chickpea are shown in table 5. It was observed that for 24 hours, 25.2±0.84, 36.4±0.71 and 44.8±0.55 % of ovicidal mortality among the experimental eggs when treated with 200, 400 and 600ppm concentration of the hexane extract of *P. hexapetallum* respectively. The LC_{50} value for the data was 685.57ppm and their confidence limits ranged from 543.41ppm (LCL) to 1294.03ppm (UCL). Furthermore, the LC_{90} value for the data was found to be 1644.18ppm and their confidence limits ranged from 1138.76ppm (LCL) to 4238.33ppm (UCL). The calculated chi-square value was 0.10. Statistical analysis of the data was found significant between the observed egg mortality and the tested concentrations ($p < 0.05$ (ANOVA; LSD -Tukey test; table 5)

Similarly it was observed that for 48 hours, 30.8, 48.6 and 53.6 % of ovicidal mortality among the experimental eggs when treated with 200, 400 and 600ppm concentration of the hexane extract of *P. hexapetallum* respectively. The LC_{50} value for the data was 500.20ppm and their confidence limits ranged from 402.19ppm (LCL) tob 707.95ppm (UCL). Furthermore, the LC_{90} value for the data was found to be 1373.23 ppm and their confidence limits ranged from 1000.30ppm (LCL) to 2859.81ppm (UCL). The calculated chi-square value was 1.21. Statistical analysis of the data was found significant between the observed egg mortality and the tested concentrations ($p < 0.05$ (ANOVA ; LSD -Tukey test; table 5)

Likewise it was observed that for 72 hours, 50.2, 58.4 and 68.2 % of ovicidal mortality among the experimental eggs when treated with 200, 400 and 600ppm concentration of the hexane extract of *P. hexapetallum* respectively. The LC_{50} value for the data was 203.22ppm and their confidence limits ranged from 445.75ppm (LCL) to 334.65ppm (UCL). Furthermore, the LC_{90} value for the data was found to be 1299.63ppm and their confidence limits ranged from 898.38ppm (LCL) to 4146.48ppm (UCL). The calculated chi-square value was 0.03. Statistical analysis of the data was found significant between the observed egg mortality and the tested concentrations ($p < 0.05$ (ANOVA; LSD -Tukey test; table 5)

Table 6: Lethal concentrations values for ovicidal activity of DCM extract of *Pterolobium hexapetallum* tested against the stored pest – *Callosobruchus maculatus*

Exposure periods (Hrs)	Concentrations (ppm)	% of Ovicidal activity	LC_{50}	95 % Fiducial Limit		LC_{90}	95% fiducial limit		χ^2
				LCL	UCL		LCL	UCL	
24	200	35.4±1.30[a]	492.65	375.16	794.94	1529.08	1052.30	4323.42	0.05
	400	46.4±1.10[b]							
	600	54.8±1.00[c]							
48	200	40.8±0.84[a]	323.37	137.11	423.28	1203.88	889.42	2458.80	1.05
	400	58.6±0.71[b]							
	600	63.6±0.45[c]							
72	200	60.2±0.89[a]	79.77	5986.32	172.55	1223.43	808.52	11634.40	0.10
	400	69.4±0.84[b]							
	600	74.2±0.55[c]							

The value represents mean ± S. D. of five replications. *mortality of the larvae observed after 24h,48h & 72 of the exposure period, WHO (2005). LC_{50}=Lethal Concentration brings out 50% Mortality and LC_{90} = Lethal Concentration brings out 90% mortality. LCL = Lower Confidence Limit; UCL = Upper Confidence Limit; Values in a column with a different superscript alphabet are significantly different at $P < 0.05$ (MANOVA; LSD -Tukey's Test).

3.9 Lethal concentrations values for ovicidal activity of DCM extract of *Pterolobium hexapetallum* tested against the stored pest – *Callosobruchus maculatus*.

Ovicidal activity of DCM extract of *P. hexapetallum* was

tested against the freshly laid eggs on the chickpea are shown in table 6. It was observed that for 24 hours, 35.4, 46.4 and 54.8 % of ovicidal mortality among the experimental eggs when treated with 200, 400 and 600ppm concentration of the hexane extract of *P. hexapetallum* respectively. The LC_{50} value for the data was 492.65ppm and their confidence limits ranged from 375.16ppm (LCL) to 794.94ppm (UCL). Furthermore, the LC_{90} value for the data was found to be 1529.08ppm and their confidence limits ranged from 1052.30ppm (LCL) to 4323.42ppm (UCL). The calculated chi-square value was 0. 05. Statistical analysis of the data was found significant between the observed egg mortality and the tested concentrations ($p < 0.05$ (ANOVA; LSD -Tukey test; table 5)

Similarly it was observed that for 48 hours, 40.8, 58.6 and 63.6 % of ovicidal mortality among the experimental eggs when treated with 200, 400 and 600ppm concentration of the hexane extract of *P. hexapetallum* respectively. The LC_{50} value for the data was 323.37ppm and their confidence limits ranged from 137.11ppm (LCL) to 423.28ppm (UCL). Furthermore, the LC_{90} value for the data was found to be 1203.88ppm and their confidence limits ranged from 889.42ppm (LCL) to 2458.80ppm (UCL). The calculated chi-square value was 1.05. Statistical analysis of the data was found significant between the observed egg mortality and the tested concentrations ($p < 0.05$ (ANOVA ; LSD -Tukey test; table 5)

Similarly it was observed that for 72 hours, 60.2, 69.4 and 74.2 % of ovicidal mortality among the experimental eggs when treated with 200, 400 and 600ppm concentration of the hexane extract of *P. hexapetallum* respectively. The LC_{50} value for the data was 79.77ppm and their confidence limits ranged from 5986.32ppm (LCL) to 172.55ppm (UCL). Furthermore, the LC_{90} value for the data was found to be 1223.43ppm and their confidence limits ranged from 808.52ppm (LCL) to 11634.40ppm (UCL). The calculated chi-square value was 0.10. Statistical analysis of the data was found significant between the observed egg mortality and the tested concentrations ($p < 0.05$ (ANOVA ; LSD -Tukey test; table 5)

Table 7: Lethal concentrations values for ovicidal activity of Ethanol extract of *Pterolobium hexapetallum* tested against the stored pest – *Callosobruchus maculatus*

Exposure periods (Hrs)	Concentrations (ppm)	% of Ovicidal activity	LC_{50}	95 % Fiducial Limit		LC_{90}	95% fiducial limit		χ^2
				LCL	UCL		LCL	UCL	
24	200	45.4±0.89[a]	298.81	52.30	419.44	1368.45	947.15	4066.06	0.003
	400	54.6±0.84[b]							
	600	64.2±0.71[c]							
48	200	50.2±0.84[a]	192.35	245.10	312.73	1146.38	833.95	2650.88	.010
	400	61.4±0.84[b]							
	600	70.6±0.55[c]							
72	200	60.4±1.10[a]	59.05	325.74	190.71	772.64	618.68	1238.98	.048
	400	72.2±0.84[b]							
	600	83.8±0.45[c]							

The value represents mean ± S. D. of five replications. *mortality of the larvae observed after 24h,48h & 72 of the exposure period, WHO (2005). LC_{50}=Lethal Concentration brings out 50% Mortality and LC_{90} = Lethal Concentration brings out 90% mortality. LCL = Lower Confidence Limit; UCL = Upper Confidence Limit; Values in a column with a different superscript alphabet are significantly different at $P < 0.05$ (ANOVA; LSD -Tukey's Test).

3.10 Lethal concentrations values for ovicidal activity of ethanol extract of *Pterolobium hexapetallum* tested against the stored pest – *Callosobruchus maculatus*.

Ovicidal activity of ethanol extract of *P. hexapetallum* was tested against the freshly laid eggs on the chickpea are shown in table 6. It was observed that for 24 hours, 45.4, 54.6 and 64.2 % of ovicidal mortality among the experimental eggs when treated with 200, 400 and 600ppm concentration of the hexane extract of *P. hexapetallum* respectively. The LC_{50} value for the data was 298.81ppm and their confidence limits ranged from 52.30ppm (LCL) to 419.44ppm (UCL). Furthermore, the LC_{90} value for the data was found to be 1368.45ppm and their confidence limits ranged from 947.15ppm (LCL) to 4066.06ppm (UCL). The calculated chi-square value was 0. 003. Statistical analysis of the data was found significant between the observed egg mortality and the tested concentrations ($p < 0.05$ (ANOVA ; LSD -Tukey test; table 5)

Similarly it was observed that for 48 hours, 50.2, 61.4 and 70.6 % of ovicidal mortality among the experimental eggs when treated with 200, 400 and 600ppm concentration of the hexane extract of *P. hexapetallum* respectively. The LC_{50} value for the data was 192.35ppm and their confidence limits ranged from 245.10ppm (LCL) to 312.73ppm (UCL). Furthermore, the LC_{90} value for the data was found to be 1146.38ppm and their confidence limits ranged from 833.95ppm (LCL) to 2650.88ppm (UCL). The calculated chi-square value was 0.10. Statistical analysis of the data was found significant between the observed egg mortality and the tested concentrations ($p < 0.05$ (ANOVA ; LSD -Tukey test; table 5)

Similarly it was observed that for 72 hours, 60.4, 72.2 and 83.8 % of ovicidal mortality among the experimental eggs when treated with 200, 400 and 600ppm concentration of the hexane extract of *P. hexapetallum* respectively. The LC_{50} value for the data was 59.05ppm and their confidence limits ranged from 325.74ppm (LCL) to 190.71ppm (UCL). Furthermore, the LC_{90} value for the data was found to be 772.64ppm and their confidence limits ranged from 618.68ppm (LCL) to 1238.98ppm (UCL). The calculated chi-

square value was 0.048. Statistical analysis of the data was found significant between the observed egg mortality and the tested concentrations ($p < 0.05$ (ANOVA ; LSD -Tukey test; table 5)

4. Discussion

The results of this study showed that the egg deposition on the seeds and repellent activity of the bruchids varied for different concentrations. As the concentration increases the eclosion were found to be decreased, because, the insects lose its potency of laying their eggs on the seed. It can be attributed due to the influence of application of plant extracts on the seed coat which intervened the physiology of pest's ovarioles which in turn interferes with the maturation of egg and processing of yolk materials inside the eggs. Wigglesworth reported that the physiology of ovary may be altered with the external factors like any compound having aroma smell [14]. Our findings corroborating with the earlier findings of several authors [15-16]. Generally, phytochemicals exert spectrum of activities including antifeedant, insect growth regulating activity IGR and chitinase inhibitor activities [17-19]. As the plant extracts used in this invention was having the strong toxic aroma to disturb its activity, it started repelling out from the highest concentration. Therefore, this plant extracts showed vital pesticidal activity in general and repellent activity in particular against the candidate pest species, at the concentration of 600pm with the ethanol extract. It may be called for the reports of several authors who have reported their study to support our present findings [20-22].

To avoid the usage of synthetic insecticides, essential oils or plant extraction from aromatic plants plays a vital role for insecticidal activity in the last decade [23]. Dhanasekaran reported that the volatile oil extracted from *Anethum sowa* showed as the best biopesticides products for controlling the emergence of the adults pulse beetle [24]. Similarly, the Citrus Sp also showed insecticidal activity against *C.maculatus* [25], *Azadirachta indica* [26], *Adhatoda vasica* and *Chenopodium ambrosioides* [27]. Female bruchid plays an important role in selecting the seeds for laying their eggs [28]. The peel oils from *Citrus aurantiifolia* and *Citrus reticulate* also showed similar insecticidal activity against the stored pest *Sitophilus zeamais* [29]. A successful insect control would be possible after having thorough knowledge about the life cycle of the pest. The main aim of the pest control is to control the insects in any form at its any one of the life stages. Henceforth, this present study throws more light on control of pulse beetle *C. maculatus* using plant based product.

5. Conclusion

Though there are several chemical pesticides available now a day to control the infestation of bruchids at field and storage, but their application could elicit various unwanted consequences on the people who consumed those pulses. Furthermore, those chemicals can deteriorate the quality of the pulses. Thus, as a part of integrated pest management green pesticide could serve as an important strategy which will be an alternate to the chemical pesticide in the near future. This present study on repellent and oviposition deterrent properties of *P. hexapetalum* led us further exploration of possible utilization of selected plant species for the successful control of *C. maculatus*.

6. Acknowledgement

Authors express their sincere gratitude to University Grants Commission, New Delhi for their financial assistance (Foundation Project: Reference No. 42-583/2013 (SR)), The Principal, and Professor and Head, Department of Advanced Zoology and Biotechnology, Govt. Arts College (Autonomous), Nandanam, Chennai-35 for the laboratory facilities provided.

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