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### RESEARCH ARTICLE

## Laboratory Assessment of Repellency and Insecticide Efficacy of Some Plant Extracts against Adults of Red Pumpkin Beetle (*Aulacophora foveicollis* Lucas)

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### ABSTRACT

Present studies were conducted to assess the repellency (3, 6, 9, 12 and 15 hour post-treatment) and insecticidal efficacy (24, 48, 72 and 96 hours post-treatment) of ethanolic-extracts of *Azadirachta (A.) indica*, *Eucalyptus (E.) camodulensis*, *Melia (M.) azadarech*, *Citrullus (C.) colocynthis* at 5, 10 and 15% concentrations against *Aulacophora (A.) foveicollis* Lucas (Chrysomelidae: Coleoptera) adults under laboratory conditions with the aim to explore alternatives to synthetic insecticides. *A. indica* and *M. azadarech* exhibited 76.7 and 69.1% repellency (repellency-class-IV), whereas *C. colocynthis* and *E. camaldulensis* demonstrated 36.9 and 28.0% repellency (repellency-class-II), respectively. Extract of *A. indica* at all concentrations and *M. azadarech* extract at 15 and 10% concentrations; whereas, that of both the extracts at 15 hours post-treatment interval performed better amplifying >70% repellency. LC<sub>50</sub> ranged between 6.3-28.0, 7.2-31.9, 7.8-33.5 and 12.7-47.4%; whereas, RC<sub>50</sub> fluctuated between 0.77-2.1, 5.9-7.4, 2.2-5.4 and 9.6-33.6% for *A. indica*, *M. azadarech*, *C. colocynthis* and *E. camaldulensis*, respectively, being higher for 24 hours and lower for 96 hours. Similarly, ET<sub>50</sub> reached 56.0-108.0, 57.5-109.9, 57.1-110.5 and 90.4-157.1 hours and RT<sub>50</sub> ranged between 5.6-8.6, 5.8-21.3, 5.6-34.2 and 82.4-121.9 hours for *A. indica*, *M. azadarech*, *C. colocynthis* and *E. camaldulensis*, respectively, being significantly longer for 5% and shorter for 15% concentration. It is suggested from these results that more repellent and insecticide action of *A. indica*, *M. azadarech*, *C. colocynthis* (1.5-2.0 times higher) than *E. camaldulensis* can be used in IPM program for this beetle.

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### INTRODUCTION

Red pumpkin beetle *Aulacophora (A.) foveicollis* Lucas (Chrysomelidae: Coleoptera), a widely distributed in South-East Asia, is the most destructive pest of cucurbit vegetables (Rahaman and Prodhan, 2007; Rahaman et al. 2008), which is reported to cause 80.63% damage on musk melon, 71.69% on long melon, 13.88% on ash gourd and 7.63% on snake gourd (Butani and Jotwani, 1984) especially in Indo-Pakistan subcontinent (Latif and Khan, 1952) and feeds on the flowers and leaves of the cucurbitaceous plants by making irregular holes and causing retardation of growth which delay maturation of crop. Its severe attack on the young seedlings of cucurbitaceous crops results in the death of plants (Waterhouse and Norris, 1987). It is noted that the

beetles prefer cotyledonous and young tender leaves and at the advent of the spring, the beetles defoliate the cucurbit seedlings to such an extent that sometimes the crop has to be re-sown for 3 to 4 times (Alam, 1969).

In Pakistan, farmers are completely dependent on the use of insecticides, like Cypergard (a mixture of cypermethrin and dimethoate), Cropgard (a mixture of cypermethrin with Cytralone [mephosfolan]), Sunmerin (cypermethrin), Mavrik (fluvalinate), Stinger (dimethoate) (Khan and Khattak, 1992), carbofuran (Sinha and Chakrabarti, 1983), Carbaryl (Khan and Jahangir, 2000), heptachlor, trichlorfon, (Makhdoomi and Ishaq, 1970), monocrotophos, chlorpyrifos (Lakshmi et al. 2005) to control *A. foveicollis*, however, the excessive use of synthetic pesticides promotes faster evolution of insecticide resistance, destruction of

natural enemies, contamination of food and production of residues etc. These threats have triggered to investigate effective alternative compounds. Exploration of plant derivatives as effective compounds has been a common pest management practice against various pests since long (Kim et al. 2004). Plant extracts have also been tried on this beetle. The lethal effect of 10% ethanolic extract of dried fruit of *Melia (M.) azedarach* on *A. foveicollis* was observed within 96 hours of exposure. The seed extract of *Azadirachta (A.) indica* showed good repellent properties against *A. foveicollis* (Chakravorty et al. 1969). A mixture of neem with karanja oil derived from the tree *Pongamia glabra*, reduced cucumber beetle population by 50-70% overnight. Ethanolic extracts of *A. indica*, *Annona squamosa*, *Convolvulus microphyllus* and *M. azedarach* were found significantly effective in repelling red pumpkin beetles (Tandon and Sirohi, 2009). The strong repellency of garlic and neem oil against cucumber beetle has also been reported (Ali et al., 2011).

In order to investigate more effective plant chemicals, the present studies were carried out under laboratory conditions on ethanolic plant extracts of *A. indica*, *M. azedarach*, *Eucalyptus (E.) camodulensis* and *Citrullus (C.) colocynthis* against *A. foveicollis*.

## MATERIALS AND METHODS

### Collection and rearing of red pumpkin beetle

The adults of *A. foveicollis* were collected from the cucurbit crops sown in the field and cultured on fresh squash leaves inside the glass cages (75×75×75 cm). The cages were placed in the laboratory maintained at 28±2°C, 70±5% RH and 12:12 D:L period. Before offering, the leaves were kept in a solution of tetracycline and water (1:9) for 10 minutes and then rinsed with water to remove residues. The leaves thus treated were first dried at room temperature (30±2°C) and then were offered for feeding. The dried and consumed squash leaves were removed and replaced with fresh and contaminated free leaves after two days interval.

### Preparation of plant extracts and their various dilutions

Seed and leaves of *A. indica* and *M. azedarach*, leaves of *E. camodulensis* and fruits of *C. citrullus* were collected, washed in water and dried under shade and were further dried in an oven at 50°C. These dried leaf materials were ground to the powder using a grinding mill. Extraction was carried out using Soxhlet apparatus with ethanol solvent. A sample of 20 g powder of each plant was taken in Soxhlet apparatus and 100 ml ethanol was added for digestion under boiling point at 45°C for 24 hours. The Ethanolic mixture was filtered and then evaporated under reduced pressure at 50°C in a rotatory evaporator to remove

solvent (Tandon and Sirohi, 2009). Four concentrations (0, 5, 10 and 15%) of each extract were prepared in emulsified water for repellency and mortality bioassay.

### Repellency bioassay

Leaf disc bioassay of *A. indica*, *M. azedarach*, *E. camodulensis* and *C. citrullus* extracts was carried out against adults of *A. foveicollis* under laboratory conditions following Tandon and Sirohi (2009) with some modifications. Discs of squash leaves were cut according to the size of large glass petri dish (41 cm diameter). Each leaf disc was cut into two equal halves. One half of each leaf disc was dipped into test solution (ethanolic extract of plant) and other one half into a solution of ethanol and emulsified distilled water (control) for five minutes and dried under an electric fan for fifteen minutes. Both halves of each leaf disc were then joined to full leaf disc by attaching extract treated and untreated (control) halves with a strip of cellotape. The treated leaf discs were placed in petri dishes over a moistened filter paper to avoid desiccation. Ten adults of *A. foveicollis* (5 days old) were released at the center of the leaf disc. Each petri dish was covered with perforated lids for proper ventilation. The lid was made perforated by cutting five circular holes (5 cm diameter) and then covering them with 5 mm wire mesh pasted with glue bond. The adults settled for feeding on treated (extract treated) and control (ethanol + emulsified water) halves were counted at post treatment intervals of 3, 6, 9, 12 and 15 hours. Whole experiment consisting of sixteen treatments (four plant extracts × four concentrations of each extract) was conducted under completely randomized designed and repeated thrice after an interval of a week under same laboratory conditions. Percent repellency at each post treatment interval was calculated by the following formula as described by Tandon and Sirohi (2009):

$$PR = \frac{(C - T)}{(C + T)} \times 100$$

Where PR = Percent repellency; C = number of adults on controlled half of leaf disc; T = number of adults on treated half of leaf disc

On the basis of percent repellency values, the extracts were classified as Class-0 (PR = <0.1%), class-I (PR = 0.1-20%), class-II (PR = 20.1-40%), class-III (PR = 40.1-60%), class-IV (PR = 60.1-80%) and class-V (PR = 80.1-100%) (Dales, 1996).

### Mortality bioassay

Leaf disc method was used for mortality bioassay and arrangement of treatments and replications were same as in repellency test. The mortality data was collected at post treatment intervals of 24, 48, 72 and 96 hours. The inactive adults from each treatment were separated and

placed in separate petri dishes having fresh and contamination free leaf discs for two days. After two days, these adults observed under microscope, if not active on probing, were considered as dead. The mortality data was transformed into percent corrected mortality according to Abbott formula (Abbott, 1925) prior to establish statistical analysis.

#### Statistical analysis

The data regarding percent repellency and mortality were subjected to ANOVA test to determine the parameters of significance and mean values for different treatments. The means of significant treatments were compared with Tukey's honestly significant difference, as performed by Danho et al. (2002). The mortality and repellency data were also subjected to probit analysis to determine  $LC_{50}$ ,  $RC_{50}$ ,  $ET_{50}$ ,  $RT_{50}$ , chi-square and confidence interval values and to regression analysis to establish linear regression equation, coefficient of correlation ( $r$ ), coefficient of determination ( $R^2$ ), ANOVA parameters and scatter diagram for each plant extract using the Minitab Statistical Program (Finney, 1971).

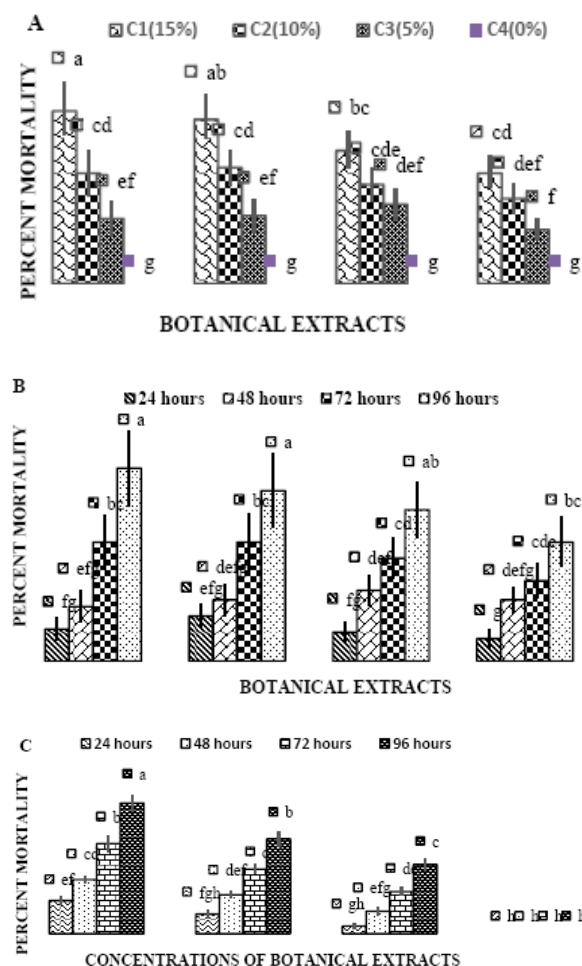
## RESULTS

The evaluated botanical extracts, their concentrations and post treatment intervals as well as their two factor interactions showed significant variations in the mortality of adult *A. foveicollis* (Table 1). Among botanical extracts, *A. indica*, *M. azadarech* and *C. colocynthis* had similar mortality rate ranging from 22.9-25.8% mortality, whereas, *E. camaldulensis* had 18.3% different from above extracts. The highest concentration and longest interval after exposure registered highest mortality in beetle (Fig. 1).

$LC_{50}$  values were time dependent and decreased with an increase in post treatment interval indicating that toxicity of the all evaluated botanical extracts increased with prolonging post treatment interval. Similarly,  $LC_{50}$  values of *A. indica*, *M. azadarech*, *C. colocynthis* and *E. camaldulensis* for each post treatment interval varied significantly as their respective 95% fiducial limits did not overlap (Table 2). The regression equations and their probability values against  $LC_{50}$  of each botanical extract explained a significant negative role ( $P < 0.05$ ) of each post treatment interval in percent mortality of adult *A. foveicollis*.  $LC_{50}$  values ranged from 6.3 to 28.0, 7.3 to 31.9, 7.8 to 33.5 and 12.7 to 47.4% for *A. indica*, *M. azadarech*, *C. colocynthis* and *E. camaldulensis*, respectively, being higher for 24 hours and lower for 96 hours of post treatment interval. Overall, *A. indica*, *M. azadarech*, *C. colocynthis* exhibited approximately equal  $LC_{50}$  values (6.3-7.8%) as well as toxicity response for adult *A. foveicollis* and were comparative 1.5-2.0 times more toxic than *E. camaldulensis* at post treatment interval of 96 hours (Table 2).

$ET_{50}$  of *A. indica*, *M. azadarech*, *C. colocynthis* and *E. camaldulensis* decreased with an increase in their concentrations and varied significantly for respective concentrations as their respective 95% fiducial limits did not overlap (Table 3).  $ET_{50}$  ranged from 56.0 to 108.0, 57.5 to 109.9, 57.1 to 110.5 and 90.4 to 157.1 hours for *A. indica*, *M. azadarech*, *C. colocynthis* and *E. camaldulensis*, respectively, being significantly longer for 5% and shorter for 15% concentration. *A. indica*, *M. azadarech*, *C. colocynthis* demonstrated approximately equal  $ET_{50}$  values (108.0-110.5 hours) for adult *A. foveicollis* and exhibited 1.5 times more  $ET_{50}$  values than *E. camaldulensis* at 5% concentration. The regression equations and their probability values against  $ET_{50}$  of each botanical extract expounded that each concentration had a significant negative impact ( $P < 0.05$ ) on the effective time ( $ET_{50}$ ) of each botanical extract against adult *A. foveicollis* (Table 3).

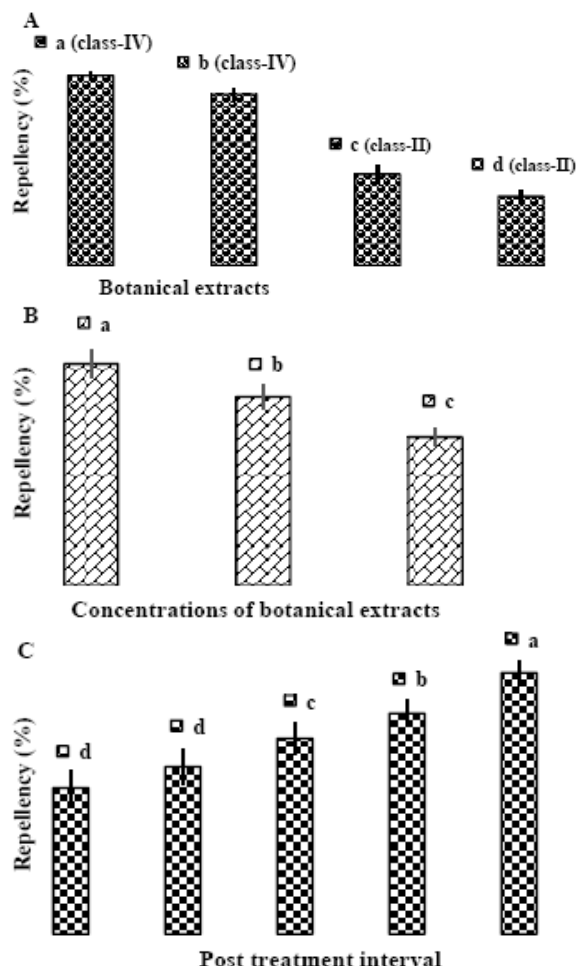
ANOVA parameters explained significant variation in repellency of adult *A. foveicollis* for botanical extracts, their concentrations and post treatment intervals as well as for all their two factor interactions except the interaction between concentrations and post treatment intervals (Table 1). Among botanical extracts, *A. indica*, repelled 76.7% whereas, *M. azadarech*, *C. colocynthis* and *E. camaldulensis* deterred 69.1, 36.9 and 28.0% of the released adults of *A. foveicollis*. Extracts of *A. indica* and *M. azadarech* belonged to repellency class-IV, whereas, extracts of *C. colocynthis* and *E. camaldulensis* fitted in repellency class-II (Fig. 2A). Repellency response of botanical against adult *A. foveicollis* increased with increasing concentration and exposure interval. Two factor interaction between botanical extracts and concentrations indicate that highest repellency was demonstrated by *A. indica* at 15% concentration (82.7%); whereas, *A. indica* at 10% and 5% and *M. azadarech* at 15% and 10% concentrations demonstrated 77.3, 70.0, 77.3 and 73.3% repellency of adult *A. foveicollis*, respectively. About 56.6% and 52.0% of the released adults of *A. foveicollis* were repelled by *M. azadarech* at 5% concentration and *C. colocynthis* at 15% concentration, respectively. However, *C. colocynthis* at 5% and 10% concentrations and *E. camaldulensis* at all tested concentration explained less than 50% repellency (18.7-38.6%) for adult *A. foveicollis*. These results reveal that application of *A. indica* extract at all tested concentrations and *M. azadarech* extract at 15% and 10% concentrations were comparatively better botanical-concentration combinations (Fig. 2). Two factor interaction between botanical extracts and exposure intervals indicate that repellency against adult *A. foveicollis* ranged from 68.9 to 86.7%, 61.1 to 77.8%, 15.5 to 64.4% and 10.0 to 48.9% for *A. indica*, *M. azadarech*, *C. colocynthis* and *E. camaldulensis* extracts, respectively, being higher for 15 hours and lower for 3 hours post treatment intervals.



**Fig. 1:** Percent mortality of *Aulacophora foveicollis* for interactions of different botanical extracts with concentrations (A) and post treatment intervals (B) and of different concentrations with post treatment intervals (C) (Means bars having different letters donot differ significantly at  $\alpha = 5\%$  and Error bars indicates standard error).

These results indicate that released adult *A. foveicollis* were firstly attracted to the treated food source and then, finding the food source unfit for consumption, started to move away from treated food. Percentage of repelled individuals of adult *A. foveicollis* gradually increased with increasing post treatment exposure period. After 15 hours of treatment application, *A. indica* extract deterred the most individuals of adult *A. foveicollis* (86.7%); whereas, *M. azadarech*, *C. colocynthis* and *E. camaldulensis* extracts deterred 77.8, 64.4 and 48.9% individuals of adult *A. foveicollis* (Fig. 3).

Probit analysis reveals that  $RC_{50}$  values of *A. indica*, *M. azadarech*, *C. colocynthis* and *E. camaldulensis* for each post treatment interval varied significantly as their respective 95% fiducial limits did not overlap (Table 4).



**Fig. 2:** Percent repellency of *Aulacophora foveicollis* for different botanical extracts (A), concentrations (B) and post treatment intervals (C) (Means bars having different letters donot differ significantly at  $\alpha = 5\%$  and Error bars indicates standard error).

The regression equations and their probability values against  $RC_{50}$  of each botanical extract explained a significant negative role ( $P < 0.05$ ) of concentrations in percent mortality of adult *A. foveicollis*.  $RC_{50}$  values ranged from 0.77 to 2.1, 5.9 to 7.4, 2.2 to 5.4 and 9.6 to 33.6% for *A. indica*, *M. azadarech*, *C. colocynthis* and *E. camaldulensis*, respectively, being higher for 24 hours and lower for 96 hours. All the tested botanical extracts exhibited less than 10%  $RC_{50}$  for adult *A. foveicollis* at post treatment interval of 96 hours; however,  $RC_{50}$  values of each botanical extract for adult *A. foveicollis* decreased with increasing post treatment interval. At maximum exposure interval (96 hours), *A. indica*, *M. azadarech* and *C. colocynthis* explained 12.5, 1.6 and 4.4 times lower  $RC_{50}$  than *E. camaldulensis* for adult *A. foveicollis*, respectively (Table 4).

**Table 1: ANOVA parameters of botanicals, concentrations, post treatment intervals and their interactions for mortality (Total df = 191) and repellency (Total df = 179) responses of *Aulacophora foveicollis***

Source of variation	Percent mortality			Repellency (%)		
	Df	F	P < 0.05	Df	F	P < 0.05
Botanicals (B)	3 <sup>a</sup> /128 <sup>b</sup>	9.287	0.00**	3 <sup>a</sup> /120 <sup>b</sup>	317.260	0.000**
Concentrations (C)	3 <sup>a</sup> /128 <sup>b</sup>	246.620	0.000**	2 <sup>a</sup> /120 <sup>b</sup>	79.779	0.000**
Post treatment Intervals (I)	3 <sup>a</sup> /128 <sup>b</sup>	154.190	0.000**	4 <sup>a</sup> /120 <sup>b</sup>	64.217	0.000**
B * C	9 <sup>a</sup> /128 <sup>b</sup>	3.094	0.002**	6 <sup>a</sup> /120 <sup>b</sup>	3.053	0.008**
B * I	9 <sup>a</sup> /128 <sup>b</sup>	3.079	0.002**	12 <sup>a</sup> /120 <sup>b</sup>	5.771	0.000**
C * I	9 <sup>a</sup> /128 <sup>b</sup>	18.998	0.000**	8 <sup>a</sup> /120 <sup>b</sup>	0.426	0.903 <sup>ns</sup>
B * C * I	27 <sup>a</sup> /128 <sup>b</sup>	0.520	0.975 <sup>ns</sup>	24 <sup>a</sup> /120 <sup>b</sup>	0.159	0.999 <sup>ns</sup>

<sup>a</sup> = Degree of freedom of independent factors and their interactions; <sup>b</sup> = Degree of freedom of error; \*\* = highly significant at probability level of 5%; <sup>ns</sup> = non-significant at probability level of 5%.

**Table 2: LC<sub>50</sub> values of different plant extracts at various exposure intervals against *A. foveicollis***

Plants	Post treatment intervals	LC <sub>50</sub> (SE)	95% Fiducial CI	Regression equation (Y = a + bx)	χ <sup>2</sup>	P
<i>A. indica</i>	24 hours	28.0 (7.4)	19.9 – 88.3	- 6.43 + 1.82x	0.004	0.001**
	48 hours	21.5 (3.6)	16.9 – 38.1	- 5.28 + 1.60x	0.201	0.000**
	72 hours	11.9 (0.97)	10.3 – 14.6	- 3.30 + 1.18x	0.504	0.000**
	96 hours	6.3 (0.57)	5.0 – 7.3	- 2.70 + 1.27x	2.171	0.000**
<i>M. azedarach</i>	24 hours	31.9 (11.4)	20.4 – 189.2	- 4.48 + 1.18x	0.113	0.002**
	48 hours	26.7 (7.7)	18.3 – 95.9	- 3.92 + 1.08x	0.273	0.001**
	72 hours	12.2 (1.0)	10.4 – 14.9	- 2.70 + 1.27x	2.171	0.000**
	96 hours	7.2 (0.65)	5.7 – 8.3	- 2.59 + 1.13x	2.917	0.000**
<i>C. colocynthis</i>	24 hours	33.5 (12.2)	21.5 – 227.8	- 6.0 + 1.60x	0.057	0.002**
	48 hours	33.3 (17.2)	18.5 – 403.9	- 2.66 + 0.65x	0.334	0.014**
	72 hours	18.7 (4.8)	13.3 – 70.0	- 2.36 + 0.68x	0.779	0.003**
	96 hours	7.8 (1.4)	3.7 – 10.7	- 1.42 + 0.51x	0.141	0.006**
<i>E. camaldulensis</i>	24 hours	47.4 (18.2)	24.3 – 135.6	- 5.82 + 1.41x	0.038	0.014**
	48 hours	41.7 (11.4)	21.3 – 128.1	- 3.11 + 0.74x	0.191	0.015**
	72 hours	35.3 (2.3)	18.3 – 39.0	- 2.38 + 0.57x	0.112	0.016**
	96 hours	12.7 (1.4)	10.4 – 17.5	- 2.66 + 0.91x	0.506	0.000**

**Table 3: ET<sub>50</sub> values of different plant extracts at various concentrations against *A. foveicollis***

Plants	Concentration	ET <sub>50</sub> (SE) (Hours)	95% Fiducial CI	Regression equation (Y = a + bx)	χ <sup>2</sup>	P
<i>A. indica</i>	5%	108.0 (7.2)	94.9 – 126.4	- 11.9 + 2.47x	1.618	0.000**
	10%	80.5 (3.9)	73.7 – 89.5	- 8.91 + 1.95x	4.772	0.000**
	15%	56.0 (2.5)	50.8 – 60.8	- 7.74 + 1.83x	9.672	0.000**
<i>M. azedarach</i>	5%	109.9 (14.9)	102.1 – 174.5	- 8.3 + 1.64x	1.618	0.000**
	10%	88.4 (7.8)	76.1 – 110.9	- 5.72 + 1.19x	3.078	0.000**
	15%	57.5 (2.9)	51.4 – 63.3	- 6.51 + 1.51x	10.397	0.000**
<i>C. colocynthis</i>	5%	110.5 (9.5)	99.2 – 135.8	- 6.89 + 1.43x	1.088	0.000**
	10%	95.5 (8.1)	82.9 – 118.6	- 8.56 + 1.75x	2.503	0.000**
	15%	75.1 (5.1)	66.1 – 87.5	- 5.80 + 1.26x	0.766	0.000**
<i>E. camaldulensis</i>	5%	157.1 (30.7)	118.9 – 300.3	- 7.92 + 1.49x	0.763	0.000**
	10%	119.1 (15.9)	96.9 – 175.9	- 6.44 + 1.27x	1.127	0.000**
	15%	90.4 (7.7)	78.3 – 112.2	- 6.18 + 1.29x	2.579	0.000**

Post treatment intervals at which 50% of the released adult *A. foveicollis* were repelled (RT<sub>50</sub>) varied significantly for respective concentrations as their respective 95% fiducial limits did not overlap and ranged from 5.6 to 8.6, 5.8 to 21.3, 5.6 to 34.2 and 82.4 to 121.9 hours for *A. indica*, *M. azadarech*, *C. colocynthis* and *E. camaldulensis*, respectively, being significantly longer for 5% and shorter for 15% concentration. Overall, *A. indica*, *M. azadarech* and *C. colocynthis* demonstrated better repellency as they repelled 50% of released individuals of adult *A. foveicollis* within shorter

period of post treatment interval at highest (15%) as well as at lowest (5%) concentrations. At lowest concentration (5%), *A. indica*, *M. azadarech* and *C. colocynthis* demonstrated 14-15 times less RT<sub>50</sub> than that of *E. camaldulensis*; however, they explained 14.2, 5.7 and 3.6 times less RT<sub>50</sub> than that of *E. camaldulensis*, respectively (Table 5). The regression equations and their probability values against RT<sub>50</sub> of each botanical extract reveal that concentration had a significant negative impact (P<0.05) on the RT<sub>50</sub> of each botanical extract against adult *A. foveicollis* (Table 5).

**Table 4: RC<sub>50</sub> values of different plant extracts at various exposure intervals against *A. foveicollis***

Plants	Post treatment intervals	RC <sub>50</sub> (SE)	95% Fiducial CI	Regression equation (Y = a + bx)	χ <sup>2</sup>	P
<i>A. indica</i>	24 hours	2.1 (0.15)	0.01 – 4.6	- 0.63 + 0.34x	0.027	0.035 *
	48 hours	1.9 (0.38)	0.00 – 4.3	- 0.63 + 0.38x	0.088	0.022 **
	72 hours	1.7 (0.11)	0.01 – 3.6	- 0.58 + 0.42x	0.008	0.009 **
	96 hours	0.77 (0.21)	0.00 – 2.2	- 0.25 + 0.44x	0.250	0.008 **
<i>M. azedarach</i>	24 hours	33.6 (12.1)	21.5 – 227.8	- 6.0 + 1.60x	0.057	0.002 **
	48 hours	33.7 (13.8)	20.4 – 332.8	- 3.93 + 1.01x	0.166	0.003 **
	72 hours	24.9 (7.8)	16.7 – 122.3	- 3.03 + 0.83x	0.042	0.002 **
	96 hours	9.6 (1.2)	7.2 – 12.8	- 1.94 + 0.69x	0.856	0.000 **
<i>C. colocynthis</i>	24 hours	5.4 (1.2)	2.2 – 7.3	- 1.36 + 0.59x	0.895	0.001 **
	48 hours	3.9 (1.3)	0.76 – 5.9	- 1.07 + 0.51x	0.559	0.002 **
	72 hours	3.3 (1.2)	0.5 – 5.2	- 0.97 + 0.51x	0.320	0.002 **
	96 hours	2.2 (0.96)	0.29 – 3.8	- 0.79 + 0.55x	0.006	0.001 **
<i>E. camaldulensis</i>	24 hours	27.4 (7.4)	19.2 – 85.6	- 5.04 + 1.41x	0.682	0.000 **
	48 hours	20.7 (3.5)	16.2 – 36.9	- 4.59 + 1.39x	0.448	0.000 **
	72 hours	16.3 (2.2)	13.2 – 24.7	- 3.48 + 1.11x	0.001	0.000 **
	96 hours	5.9 (0.76)	4.1 – 7.2	- 2.01 + 0.92x	1.53	0.000 **

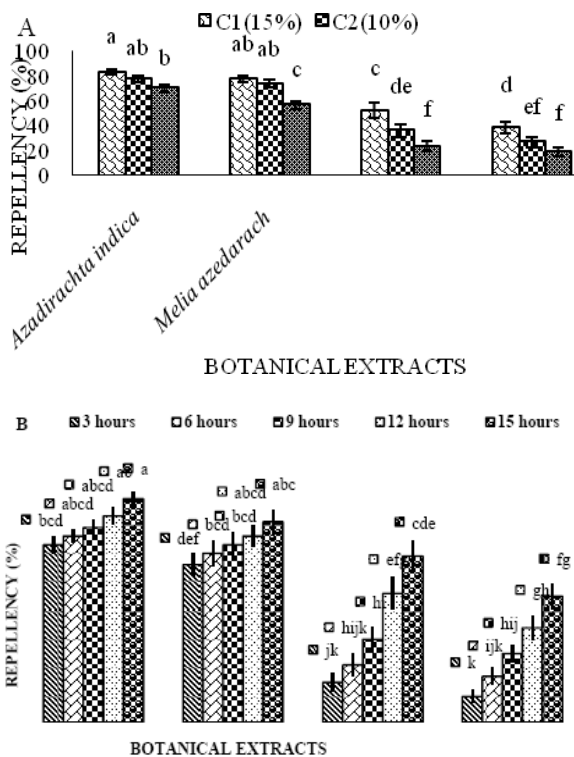
**Table 5: RT<sub>50</sub> values of different plant extracts at various concentrations against *A. foveicollis***

Plants	Concentration	RT <sub>50</sub> (SE) (Hours)	95% Fiducial CI	Regression equation (Y = a + bx)	χ <sup>2</sup>	P
<i>A. indica</i>	5%	8.6 (3.2)	0.00 – 21.4	- 0.99 + 0.29x	3.042	0.023 **
	10%	6.9 (2.8)	0.01 – 15.1	- 0.94 + 0.33x	2.385	0.007 **
	15%	5.6 (4.1)	0.56 – 14.8	- 1.23 + 0.44x	3.444	0.000 **
<i>M. azedarach</i>	5%	121.9 (12.8)	103.7 – 164.9	- 10.3 + 2.11x	1.347	0.000 **
	10%	111.7 (12.7)	93.5 – 153.8	- 6.94 + 1.39x	2.531	0.000 **
	15%	82.4 (6.3)	72.0 – 99.3	- 5.90 + 1.26x	4.977	0.000 **
<i>C. colocynthis</i>	5%	34.2 (8.8)	8.3 – 49.5	- 1.69 + 0.37x	0.954	0.006 **
	10%	14.1 (1.5)	2.4 – 21.3	- 8.56 + 1.75x	0.828	0.049 *
	15%	5.6 (2.8)	0.01 – 15.1	- 0.94 + 0.33x	2.385	0.007 **
<i>E. camaldulensis</i>	5%	113.1 (10.7)	97.4 – 146.9	- 9.34 + 1.89x	9.986	0.000 **
	10%	86.1 (5.6)	76.7 – 100.7	- 7.39 + 1.58x	7.491	0.000 **
	15%	58.1 (3.5)	50.9 – 65.4	- 5.44 + 1.25x	15.563	0.000 **

## DISCUSSION

Laboratory evaluation of the efficacy of new molecules is a prerequisite of their field evaluation. Keeping this fact in view, present study was conducted under laboratory conditions for evaluating the repellency and insecticidal efficacy of ethanolic extracts *A. indica*, *M. azedarach*, *C. colocynthis* and *E. camaldulensis* against *A. foveicollis* adults. The evaluated botanical extracts, their concentrations and post treatment intervals explained significant variations in the mortality of adult *A. foveicollis* in present study. This variation in mortality response of adult *A. foveicollis* against evaluated plant extracts is attributable to qualitative and quantitative variation in their chemical constitutions (Rizvi et al., 2012; Koubala et al., 2013). However, variations in mortality due to various concentrations and exposure intervals may have been due to varying degree of persistence and exposure chances of extracts at different concentrations and exposure intervals, respectively. Mortality and repellency response of adult *A. foveicollis* to all plant extracts increased with increasing concentrations and exposure interval. These

results suggest that mortality and repellency is directly proportional to ranges of concentrations and exposure intervals used in present study. Regression and correlation parameters demonstrated in present study also showed that concentration and exposure-interval played a significant contribution in mortality and repellency of *A. foveicollis*. All extracts at 15% and 10% concentrations had approximately 2-times and 1.5-times higher mortality in *A. foveicollis* adults, respectively, than 5% concentration (15.8-23.3%). The higher mortality of *A. foveicollis* adults by all extracts at higher concentration may be attributable to ingestion of more quantity of toxic molecules which exhibit more persistence and difficult metabolism in more concentrated extract as compared to diluted extract which persists for shorter period and is easy to be metabolized (Bashir et al., 2013). The high mortality at 96 hours is attributable to longer exposure of *A. foveicollis* adults to treated surface which increases the probability of ingestion of more treated surface and quantity of toxicants inside their digestive tract. The similar toxicity level and time of these plant extracts suggest a common mechanism of toxic action causing



**Fig. 3: Percent repellency of *A. foveicollis* for interactions of different botanical extracts with concentrations (A) and post treatment intervals (B) (Means bars having different letters donot differ significantly at  $\alpha = 5\%$  and Error bars indicates standard error).**

mortality of the beetle. Ali et al. (2011) reported that neem seed extract caused higher mortality and performed better than eucalyptus leaf extract against *A. foveicollis* adults. Osman et al. (2013) also documented better response of neem extract at 7.5% concentration against *A. foveicollis* after 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> spray. Similarly, the work of previous researchers demonstrated reasonable pesticidal efficacy of *A. indica*, *M. azedarach*, and *C. colocynthis*, extracts against insects and related arthropods (Italo et al., 2009; Kumral et al., 2010). Toxicity response of *M. azedarach* leaf extract demonstrated in present study against *A. foveicollis* adults were highly in consistent with the interpretations of various researchers who documented pesticidal activities of *M. azedarach* extracts against arthropod pests (Jazzar and Abou-Fakhkr, 2003; Italo et al., 2009). Some investigations on the pesticidal properties of *C. colocynthis* leaf extract against arthropod pests have been done and support the finding of present study; for example larvicidal activity of *C. colocynthis* leaf extract against mosquito larvae (Rahuman et al. 2008) and *Aphis craccivora* (Torkey et al. 2009). Regarding repellency, extracts of *A. indica* and *M. azadarech* exhibited class-IV- repellency (60.1-80%), whereas, extracts of *C.*

*colocynthis* and *E. camaldulensis* fitted in repellency class-II (20.1-40%) in present study. This variation in repellency is attributable to difference in type and chemical nature of the volatile secondary metabolites present in these plant extracts that need further investigation. The present findings are partially in agreement with those of Tandon and Sirohi (2009) who assessed the ethanolic extracts of *A. indica*, *Annona squamosa*, *Convolvulus microphyllus* and *M. azedarach* in a laboratory against *Raphidopalpa foveicollis* Lucas (Coleoptera: Chrysomelidae) and reported *A. indica* and *M. azedarach* as class-IV (60.1-80%) and class III (40.1-60%) repellents, respectively. The variation in results for *M. azedarach* may be due to difference in insect species (*R. foveicollis*) used in their experiment. A class-II repellency (20.1-40%) of *C. colocynthis* demonstrated in present study is also comparable with the results of Mullai and Jebanesan (2007) and Rehman et al. (2009) who reported repellency activity against *Culex quinquefasciatus* adults and 34.6% repellency against *Bactrocera zonata* Saunders (Diptera: Tephritidae), respectively. An increase in repellency response of botanicals against adult *A. foveicollis* with increasing concentration and exposure interval demonstrated in present study may have been due to highly concentrated solution at higher concentration as volatilization of concentrated compounds is slower and prolonged as compared to more diluted solution. These results also indicate that released *A. foveicollis* adults were firstly attracted to the food source and later on, finding the food source unfit for consumption, started to move away from treated food source.

The present investigations suggest that *A. indica*, *M. azadarech*, *C. colocynthis* had better repellency and insecticidal efficacy (1.5-2.0 times higher) than *E. camaldulensis* and can be used to develop natural molecules as repellents and insecticides that should be further evaluated in laboratory and field conditions.

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