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# Toxic and repellent effects of three botanical oils against adult *Aphis craccivora* Koch. (Homoptera: Aphididae) under laboratory conditions

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**Abstract**: In the present study, three botanical oils viz. neem, karanja and mahogany were tested against the adult bean aphid, *Aphis craccivora* Koch to evaluate the toxic and repellent effects under laboratory condition in the Department of Entomology, Hajee Mohammad Danesh Science and Technology University, Dinajpur during September 2016 to January 2017. The results indicated that all the botanical oils had different levels of toxic and repellent effects and was found effective for controlling the aphid. Among the botanicals, the highest (29.83%) mortality was found in mahogany oil whereas the lowest (22.96%) in karanja oil against the adult aphid. Mahogany oil also showed the highest (77. 33%) repellent effects i.e. class IV among the three tested botanical oils. On the contrary, karanja oil showed the lowest repellent (34.00%) effects, i.e. class II and neem oil belongs to the repellency class III. However, with the finding of this study, it may conclude that for eco-friendly management of *A. craccivora*, all the botanical oils showed promising toxic and repellent effects against the adult bean aphid but mahogany oil was the highest. So, mahogany oils may be considered at farmer's level as it is cheaper, easily available, processable, and usable for the management of bean aphid.

**Key words**: *Aphis craccivora*, botanical oils, mortality, repellent effect.

## Introduction

Country bean (*Lablab purpureus* L.) locally known as deshi sheem is a highly proteinaceous legume and a major winter vegetable in Bangladesh (Salim *et al.,* 2013). The amounts of crude protein of the country bean were estimated 21 to 38% and 20 to 28% in leaf and seed, respectively (Cook *et al.,* 2005). Besides, seeds have some micronutrients and minerals also (Kala *et al*., 2010; Shaahu *et al*., 2015). It is the atmospheric N-fixing legume and also a valuable green manure forming crop (Adebisi & Bosch, 2004). However, crops are threatened by several insect pests resulting huge economic losses (Thejaswi *et al.,* 2008; Oliveira *et al.,* 2014).

Among the insect pests of legumes crops, bean aphid, *Aphis craccivora* Koch (Homoptera: Aphididae) is most destructive and cosmopolitan pests (Capinera, 2001; Madahi *et al.,* 2013). There are about 4700 species of aphids in the world which cause enormous loss of vegetables (Blackman & Eastop, 2007; Alikhani *et al.,* 2010). Both the adults and nymphs sucks plant sap and cause up to 10-90% yield loss (Akhtar *et al.,* 2010; Razaq *et al.,* 2011). They damage all parts of the plants except roots and show symptoms as stunting growth, crinkling and curling of leaves, pods and indirectly by transmission of plant viruses (Saranya *et al.,* 2010; Sainsbury *et al.,* 2010; Kamphuis *et al.,* 2012). They also secrete honeydew causing the growth of sooty mould fungus, which inhibits photosynthesis (Kingler *et al.,* 2001; Smith & Boyko, 2007; Singh *et al*., 2014).

Aphid is a prolific breeder and randomly corrosive insecticides are used to control them for quick knockdown (Pavela *et al*., 2009). But chemical protection measures exert so many serious drawbacks (Karungi *et al.,* 2000; Lee *et al*., 2001; Ambethar, 2009). Their extensive and indiscriminate use causes ecological imbalance, resistance to pest, pest resurgence, outbreaks of secondary pests and also creates phytotoxicity, residues in foods and feeds, killing beneficial organisms in the ecosystem (Mahmud *et al*., 2002; Ashamo, 2004; Stapel *et al.,* 2000). Hence, researchers and scientists all over the world are now trying to adopt alternatives of insecticides to protect crop from insect pest (Rajappan *et al*., 2000; Isman, 2006). Use of natural products are an excellent source for controlling insect pests (Gorur *et al*., 2008; Franck *et al.,* 2009). Botanicals become promising to control pests by offering several advantages in comparing to insecticides (Attia *et al.,* 2013; Kedia *et al.,* 2015). Essential plant oils demonstrated high insecticidal activity against insect pests (Cheng *et al.,* 2003; Ahmedani *et al*., 2007; Khater, 2012). Therefore, the present study was undertaken to evaluate the effectiveness of three botanical oils viz. neem, karanja and mehogony for controlling bean aphids.



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## Materials and Methods

### Experimental site

The experiment was carried out in the laboratory conditions (25 ± 5$^o$C, 65-75% RH) of the Department of Entomology, Hajee Mohammad Danesh Science and Technology University (HSTU), Dinajpur, Bangladesh during the period from September 2016 to January 2017.

### Raising of host plant

Country bean used as host plant were raised in small plastic pots (11 cm x 14 cm) in the corridor of the laboratory. Plants were fertilized as needed and watering was done regularly. No pesticide was applied during the experiment.

### Collection and rearing of test insects

*A. craccivora* were collected from the infested bean plants of HSTU central farm. Twigs of bean harboring with *A. craccivora* colonies were brought to the laboratory (25 ± 5$^o$C, 70 ± 5% RH). Afterwards, aphids were gently removed from the bean twigs with the help of soft camel hair brush and released on the fresh bean twigs kept in rectangular jars (14 cm x 10.5 cm x 30 cm) in the laboratory. The jars were covered with a piece of cloth and fastened with rubber bands to prevent insect's getaway. Newly hatched crawlers were collected from the laboratory culture and were placed on to the fresh bean twigs for mass culture. These processes were continued upto the experimental requirements.

### Identification of adult *A. craccivora*

Traditionally, aphid species have been indentified based on their morphological characters (Emden & Harrington, 2007). Adults were identified by using the procedures as stated by Blackman & Eastop (2000).

### Tested botanical oils

Commercially available three plant oils namely neem (*Azadirachta indica*), mahogany (*Swietenia mahagoni* L.) and karanja oil (*Pongamia pinnata* L.) were collected from the local market of Dinajpur town. Four concentrations (0.5, 1.0, 1.5, and 2.0%) were prepared with distilled water containing 0.1% detergent as emulsifier.

### Direct toxicity test

Toxicity and repellent effects were performed in the ambient laboratory conditions against adult *A. craccivora*. Fresh young bean leaves were collected and dipped in assigned tested oils for 5-10 seconds containing four concentrations (0.5, 1.0, 1.5, and 2.0%) and then the leaves were air-cured for 15 m. Each leaf separately placed in each Petri dish (90 mm). Ten (10) aphids (both adults and nymphs separately) were released on each leaf twig by the help of a camel-hair brush. Three replications were done for each concentration. Adult and nymph mortalities were recorded at 24, 48 and 72 hour after treatment (HAT). Mortality data were corrected by Abbott's (1925) formula:

$$P = \frac{p' - C}{100 - C} \times 100$$

Where, $P$ = Percentage of corrected mortality

$P'$ = Observed mortality (%)

C = Mortality (%) at control.

### Repellency test

The repellency test was conducted according to the method as described by Talukder & Howse (1995). Petri dishes (120 mm) sized filter papers (Whatman No. 40) were cut in two half and 1.0 ml of each concentrate oils were applied to a half filter paper uniformly with a pipette. The treated half were then air-dried and attached with the untreated half with a cello-tape those could not interfere with the free movement of insect from one half to another. Distance between the filter paper segments remained sufficient to prevent seepage of test samples from one half of circle to another. Each filter paper was then placed in a Petri-dish and 5 pair adults and nymphs were released at the center of the filter paper. Three replications were done for each concentration for each plant extract. Number of insects on each treated and untreated portion was counted at two hour intervals up to the 10$^{th}$ hour.

The data were expressed as percentage repulsion (PR) by the following formula: [PR (%) = (Nc-50) × 2]. Where, Nc = the percentage of insects present in the control half. Positive (+) values expressed repellency while negative (-) attractancy. The average values were then categorized according to the following scale (McDonald *et al*. 1970).

| Class | Repellency (%) | Class | Repellency (%) |
|---|---|---|---|
| 0 | >0.01-0.1 | III | 40.1-60 |
| I | 0.1-20 | IV | 60.1-80 |
| II | 20.1-40 | V | 80.1-100 |

### Statistical Analysis

The data were analyzed using completely randomized design (CRD) through Mathematical and Statistical program (MSTAT-C). All graphs were done using MS-Excel software. The treatment mean values were compared by Duncan's New Multiple Range Test (DMRT). The median lethal values ($LD_{50}$) were determined by probit analysis.

## Results

### Direct toxic effect of oils on adult *A. craccivora*

The toxicity effects of three botanical oils, doses and interactions against the adult of *A. craccivora* are shown in the Tables 1-3. The highest (29.83%) adult mortality was recorded in mahogany oil while the lowest (22.96%) in karanja oil (Table 1). From the result it was observed that the order of toxicity of three oils were mahogany > neem > karanja. The highest mortality (51.63%) was found at the dose 2.0% (Table 2). Conversely, the lowest mortality (17.72%) was observed at the dose 0.5%. Average mortality was increased with increased dose and time. In the interaction of oils, doses and times, significantly the highest mortality (62.70%) was observed at 72 HAT in mahogany oil at 2.0% while the lowest at 0.0 (control) (2.23%) (Table 3). The $LD_{50}$ values of the tested oils against *A. craccivora* adult showed that mehogony oil possessed the lowest $LD_{50}$ values at 24, 48 and 72 HATs (Table 4). Conversely, karanja oil showed the highest $LD_{50}$ values at 24, 48 and 72 HATS. It was observed that the $LD_{50}$ values for mehogony oil were 2.75%, 1.73%, 1.04% at 24, 48 and 72 HATs respectively, among the tested botanicals (Table 4). The probit mortality data and log doses of the tested oils were presented in Figures 1.

**Table 1:** Direct toxic effect of different botanical oils against the adult of *A. craccivora* at different HAT* (oils × times)

| Treatments (Oils) | Adult mortality (% ) at different HAT | | | Average mortality (%) |
|---|---|---|---|---|
| | 24 | 48 | 72 | |
| Neem | 16.00 ab | 21.85 b | 34.37 b | 24.07 b |
| Karanja | 14.00 b | 23.26 ab | 31.63 b | 22.96 b |
| Mahogany | 19.33 a | 27.48 a | 42.67 a | 29.83 a |
| LSD | 4.01 | 5.187 | 4.480 | 3.750 |
| CV % | 32.68 | 28.74 | 16.58 | 19.63 |

*HAT= Hours after treatment. Within column values followed by different letter(s) are significantly different by DMRT at 5% level of probability.

**Table 2:** Toxicity effect of different doses of botanical oils against the adult of *A. craccivora* at different HAT* (oils × times)

| Treatments (Doses %) | Adult mortality (%) at different HAT | | | Average mortality (%) |
|---|---|---|---|---|
| | 24 | 48 | 72 | |
| 2.0 | 34.44 a | 51.48 a | 69.01 a | 51.63 a |
| 1.5 | 21.11 b | 33.09 b | 50.49 b | 34.90 b |
| 1.0 | 13.33 c | 18.27 c | 33.21 c | 21.60 c |
| 0.5 | 13.33 c | 14.81 c | 25.06 d | 17.72 c |
| Control | 0.00 d | 3.33 d | 3.33 e | 2.23 d |
| LSD | 5.175 | 6.696 | 5.783 | 4.841 |
| CV % | 32.68 | 28.74 | 16.58 | 19.63 |

*HAT= Hours after treatment. Within column values followed by different letter(s) are significantly different by DMRT at 5% level of probability.

**Table 3:** Direct toxic effect of botanical oils at different doses against the adult of *Aphis craccivora* at different HAT (oils × doses × times)

| Treatments (Oils) | Doses (%) | Adult mortality (%) at different HAT* | | | Average mortality (%) |
|---|---|---|---|---|---|
| | | 24 | 48 | 72 | |
| Neem | 2.0 | 33.33 b | 44.44 b | 68.89 b | 48.87 b |
| | 1.5 | 16.67 c | 27.41 c | 44.81 d | 29.63 c |
| | 1.0 | 13.33 c | 20.37 cd | 30.74 efg | 21.47 cde |
| | 0.5 | 16.67 c | 13.70 de | 24.07 g | 18.13 e |
| Karanja | 2.0 | 26.67 b | 48.15 b | 55.19 c | 43.33 b |
| | 1.5 | 16.67 c | 27.04 c | 41.11 de | 28.27 cd |
| | 1.0 | 13.33 c | 20.74 cd | 31.11 efg | 21.73 cde |
| | 0.5 | 13.33 c | 17.04 cd | 27.41 fg | 19.23 de |
| Mahogany | 2.0 | 43.33 a | 61.85 a | 82.96 a | 62.70 a |
| | 1.5 | 30.00 b | 44.81 b | 65.56 b | 46.80 b |
| | 1.0 | 13.33 c | 13.70 de | 37.78 def | 21.60 cde |
| | 0.5 | 10.00 c | 13.70 de | 23.70 g | 15.80 e |
| | 0.0(Control) | 0.00 d | 3.33 e | 3.33 h | 2.23 f |
| LSD | | 8.963 | 11.60 | 10.02 | 8.385 |
| CV (%) | | 32.68 | 28.74 | 16.58 | 19.63 |

*HAT= Hours after treatment, within column values followed by different letter(s) are significantly different by DMRT at 5% level of probability.

**Table 4:** Relative toxicity (probit analysis) of different oils against *A. craccivora* adults after 24, 48 and 72 hours

| Botanical oils | Adult used (n) | $LD_{50}$ (%) values | 95% fiducial limits | | $\chi^2$ values (df=2) |
|---|---|---|---|---|---|
| | | | Lower | Upper | |
| | | 24 HAT* | | | |
| Neem | 30 | 14.72 | 0.2425 | 893.84 | 2.57 |
| Karanja | 30 | 17.23 | 0.2347 | 1266.10 | 0.81 |
| Mahagony | 30 | 2.75 | 1.5167 | 4.98 | 1.25 |
| | | 48 HAT* | | | |
| Neem | 30 | 3.01 | 1.3544 | 6.6945 | 0.93 |
| Karanja | 30 | 2.99 | 1.2690 | 7.0778 | 1.77 |
| Mehogony | 30 | 1.73 | 1.3161 | 2.2943 | 4.19 |
| | | 72 HAT* | | | |
| Neem | 30 | 1.42 | 1.0481 | 1.9342 | 2.80 |
| Karanja | 30 | 2.04 | 0.9881 | 4.2299 | 1.00 |
| Mehogony | 30 | 1.04 | 0.8481 | 1.2872 | 2.20 |

*HAT = Hour after treatment.

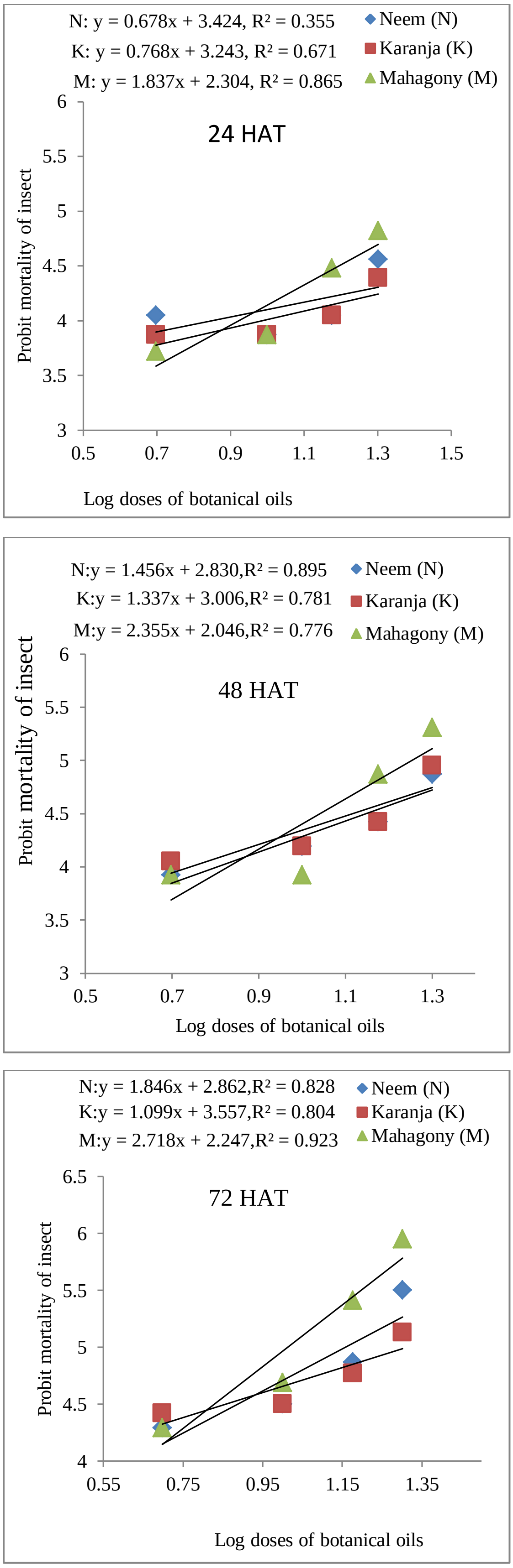


**Figure 1.** Relationship between probit mortality and log doses of different botanical oils on *A. craccivora* adult at various exposure times.

### Repellent effect of oils against *A. craccivora* adults

The repellent effect of three botanical oils on the adults of bean aphid are presented in Table 5. Among the three tested oils, mahagony and karanja showed the highest (77.33%) and the lowest (34.0%) repellency, respectively. Repellency percent decreased with the progress of time and increased with increasing doses of all oils. The repellency class of different oils at different concentration levels varied between II to V. Among the botanicals the highest repellency effect was found at 2.0 % mehogony oil (84.0%) whereas the lowest (26.67 %) at doses (1.0 and 1.5%) of karanja oil (Table 5).

## Discussion

Bean aphid, *A. craccivora* have become schedule pests of bean and causes major threat to its production in Bangladesh. Insecticides are solely relying to reduce its economic losses during production as a part of integrated management strategies against this notorious pest. However, the accumulation of toxic compounds in the green pods makes it vulnerable for human consumption as a delicious vegetable. Synthetic insecticides have been found effective in managing bean aphid to minimize yield losses but so many serious drawbacks. Results of the present study revealed that all the tested oils had toxic and repellent effects against the adult, *A. craccivora* under laboratory conditions. Among the botanicals, the highest (29.83%) mortality was found in mahogany oil whereas the lowest (22.96%) in karanja oil against the adults bean aphid. The mahogany oil offered promising toxicity by applying the highest dose of 2.0% against adult aphids. The results of the present findings are also in close proximity with those of Pinto *et al*. (2013). They concluded that botanical oils of neem (Neemseto ®), and cotton seed being achieved satisfactory control while 100% on the thiamethoxam-treated plants infested by *Aphis gossypii* Glover. The present results are similar with Patil & Chavan (2009). They cited that *Acacia concianna* extracts were the most toxic against sugarcane woolly aphid, *Ceratovacuna lanigera* Zehnter. Mortality of the tested extracts was concentrations and exposure time dependent under laboratory conditions. Lin *et al*. (2009) reported that sugar apple, (*Annona squamosa*) seed oil, an edible tropical fruit was also very effective in controlling the cotton aphid, *A. gossypii* Glover on melon leaves.

**Table 5:** Repellent effect of different botanical oils and doses against the adult *A. craccivora* at different HAT*

| Treatments (Oils) | Doses (%) | Repellency (%) at different HAT* 2 | 4 | 6 | 8 | 10 | Average | Class |
|---|---|---|---|---|---|---|---|---|
| Neem | 2.0 | 60.00 [bc] | 66.67 [ab] | 66.67 [ab] | 73.33 [ab] | 73.33 [ab] | 68.00 [ab] | IV |
| | 1.5 | 86.67 [ab] | 73.33 [ab] | 40.00 [a-c] | 66.67 [a-c] | 53.33 [bc] | 64.00 [ab] | IV |
| | 1.0 | 73.33 [a-c] | 73.33 [ab] | 40.00 [a-c] | 40.00 [b-d] | 53.33 [bc] | 56.00 [bc] | III |
| | 0.5 | 46.67 [cd] | 26.67 [c] | 33.33 [bc] | 23.33[d] | 26.67 [cd] | 31.33 [d] | II |
| Karanja | 2.0 | 33.33 [d] | 46.67 [bc] | 46.67 [a-c] | 46.67 [a-d] | 26.67 [cd] | 40.00 [cd] | II |
| | 1.5 | 26.67 [d] | 46.67 [bc] | 46.67 [a-c] | 33.33 [cd] | 33.33 [cd] | 37.33 [cd] | II |
| | 1.0 | 26.67 [d] | 26.67 [c] | 33.33 [bc] | 33.33 [cd] | 40.00 [cd] | 32.00 [d] | II |
| | 0.5 | 33.33 [d] | 26.67 [c] | 26.67 [c] | 26.67 [d] | 20.00 [d] | 26.67 [d] | II |
| Mehogony | 2.0 | 93.33 [a] | 86.67 [a] | 73.33 [a] | 80.00 [a] | 86.67 [a] | 84.00 [a] | V |
| | 1.5 | 73.33 [a-c] | 86.67 [a] | 73.33 [a] | 73.33 [ab] | 73.33 [ab] | 76.00 [ab] | IV |
| | 1.0 | 86.67 [ab] | 73.33 [ab] | 73.33 [a] | 66.67 [a-c] | 73.33 [ab] | 74.67[ab] | IV |
| | 0.5 | 66.67 [a-c] | 80.00 [ab] | 66.67 [ab] | 73.33 [ab] | 86.67 [a] | 74.67 [ab] | IV |
| LSD | | 25.12 | 31.78 | 34.17 | 30.89 | 26.35 | 20.72 | |
| CV | | 25.31 | 31.72 | 39.24 | 34.55 | 29.01 | 22.20 | |

*HAT = Hour after treatment. Mean followed by different letter(s) are significantly different by DMRT at 5% level of probability

Significant level of success in the suppression of aphid reported by various authors with extracts of different botanicals including neem and karanja leaf (Saikia *et al.,* 2000; Katsvangwa & Chigwaza, 2004; Rawleigh & Boyd, 2008; Biswas, 2013). The result of this study indicates that all levels of tested oil except 0.5% and 1.0% had shown sufficient aphicidal effect.

More current research illustrated that various plant products have been tried by several researchers with a good degree of success as protectants against several species of aphids. The results of the present study are comparable with various scholars where they mentioned that different extracts of Mexican marigold, sodom apples, garlic and ginger were promising against the aphids *Brevicoryne brassicaea* (Peris & Kiptoo, 2017); neem and moringa (*Moringa olifera*) to wheat aphid *Schizaphis graminum* (R.), *Rhopalosiphum padi* (L.) and *Sitobion avenae* (F.) (Shah *et al*., 2017); Orange peel *(Citrus sinensis*), Bitter goard (*Momordica dioica*), garlic (*Allium vineale*), mari gold; hot pepper (*Capsicum frutescens*) and tobacco (*Nicotiana tabacum*) to wheat aphid (Iqbal, 2011); garlic bulbs *(Allium sativum*), endod (*Phytolacca dodecandra*) and neem seeds (*Azadirachta indica*) to pea aphids, *Acyrthosiphon pisum* (Harris) (Megersa, 2016).

Among all the tested botanicals, mehogony, neem and karanja attributed significant repellency against *A. craccivora.* Varied repellency of different tested botanical oils indicated that the pest suppressing properties are not uniformly distributed. Ginger extract has been used to control aphids in various infested plants of Brassica family. It has repellant mode of action against the aphids but no effect on non-target organisms e.g. parasitoids. Suthisut *et al.* (2011) experienced that *A. conchigera* and *C. zedoaria* extracted essential oils at the highest concentration (314.56 × $10^{-3}$ µl/$cm^2$) repelled *S. zeamais* adults, with > 87% of the insects found on the untreated filter paper. Besides, aphid populations suppressed more effectively when it was applied in combined with neem and eucalyptus, *Eucalyptus globulus* (family Myrtaceae) leaves extract (Manzoor *et al*., 2015; Ali *et al*., 2015).

The biological activities of tested oils can be ascribed to several alkaloid contents as an insecticidal potency (Jastad *et al*., 2009; Alice *et al*., 2007). The alkaloids, terpinoids, steroids, glycosides as Cyclomahogenol, tannins, alkaloids, saponins and terpenoids as main phytoconstituents present in mehogony (Chakraborty & Basak, 1971; Sahgal *et al*., 2009; Hajra *et al*., 2011). Karanja oil contains karanjin another alkaloid (Prakash & Rao, 1996) having insect control properties against pulse beetle, *Callosobruchus chinensis* (Hossain *et al*., 2014). These chemical compounds might associate with deterrent, repellent and anti-feeding actions against *A. craccivora*, *B. brassicaea* (Homoptera: Ahididae) but seems harmless or tolerated by beneficial natural enemies as well (Katsvangwa & Chigwaza, 2004). Spray of botanicals particularly neem seed oil and neem seed extracts did not reduce the parasitism rate of *Diaeretiella rapae* (McIntosh) (Hymenoptera: Braconidae) on green peach aphid, *M*. *persicae* under laboratory and field conditions (Lowery & Isman, 1995). In fact, plant based formulation especially neem not only shows systemic and contact actions but also acts as repellant, antifeedent, sterilents, growth inhibitors and toxicological repellents against insect pests, which has been considered as a low-cost management tactic (Gahukar, 2000). Some oils have broad spectrum insecticidal activity against aphid, affecting insect nervous and defence systems (Hold *et al*., 2000; Isman, 2000; Ketoh, 2004). The biological activity of oils interferes with normal activities of insects resulting might to death (Schoonhoven, 1978).

## Conclusion

It is concluded that the botanical oils used in the present study had direct toxic effect on the adult and nymphs of *A. craccivora* which can be used as safe pesticides. Uses of these botanical oils have a great economic and environmental importance than the chemical pesticides. For this reason farmers can use botanical oils as insecticides in agricultural sector because of direct toxic and repellent effects against *A. craccivora* as well as other vegetable pests also. Among the three botanical oils mahogany showed the highest toxic and repellent effects against *Aphis craccivora*. The findings of the present investigation reveal the broad spectrum toxic properties of the tested indigenous botanical oils against the adult and nymph of *A. craccivora.* However, further studies need to be conducted to isolate and evaluate the compound with its mode of action of these oils.

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