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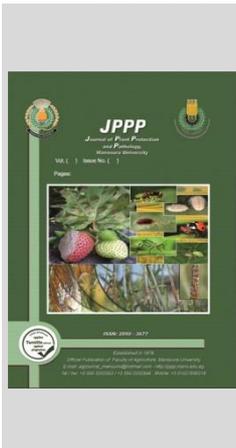
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Toxic and Repellent Effects of Three Oils as Seed Treatment against *Aphis craccivora* Koch. under Laboratory Conditions

El-Solimany, E. A. and M. A. Abolfadel*



Plant Protection Research Institute, Agricultural Research Centre, Dokki, Giza, Egypt.



ABSTRACT

The present study was carried out in the laboratory to investigate the repellent and toxic effects of faba bean seed treatment by camphor, mint, and basil oils on *Aphis craccivora*, and also, the effect of using dry and sprouting seeds was investigated. Data indicated that using sprouting seeds increased the repellent and toxic effects of oils compared with using dry seeds. After 1 hour, the highest repellent effect was recorded on sprouting seed treatment with basil oil (73.68%), followed insignificantly by dry seed treatment with basil oil (71.43%), sprouting seed treatment with mint oil (70.00%), and sprouting seed treatment with camphor oil (67.74%). However, after 24 hours, sprouting seed treatment with mint oil recorded the highest repellent effect (76.74%), followed insignificantly by dry seed treatment with basil oil (75.61%), sprouting seed treatment with basil oil (75.61%), and sprouting seed treatment with camphor oil (70.00%). No mortality percentages were observed by 1 day from treatment. After 2 days, the mortality percentage was almost low to moderate, however, a sharp increase was observed after 3 days, then mortality slightly increased after the fourth and fifth days in all treatments. Sprouting seed treatment with basil oil was the most effective against the adults of *A. craccivora* with mean mortality of 63.74%, followed insignificantly by sprouting seed treatment with camphor oil (63.02%) and with mint oil (61.36%).

Keywords: camphor, mint, basil, faba bean, *Aphis craccivora*.

INTRODUCTION

Faba bean (*Vicia faba* L.) is one of the most important legume vegetables in Egypt. It is generally used as a vegetable, green or dried, fresh or canned human food in the local market or to be exported. The most important and dangerous insect pest attacking faba bean is *A. craccivora* (Mousa and Metwally, 2014 and Singh and Singh, 2017).

Seed dressing provide targeted control of insect pests, it offers protection at the establishment phase and can often delay or remove the need to apply foliar sprays, however, it depends almost on the use of synthetic insecticides (Hassan *et al.*, 2018). The negative effects of chemical insecticides on non-target animals and humans, caused by residues left on plant crops can negatively affect human health (Bale *et al.*, 2008). Essential oils define as secondary metabolic products of a plant that are volatile, natural, and complex compounds. Essential oils can act as toxins, antifeedants, and repellents for many aphid species (Mossa, 2016). Many investigators reported the insecticidal effect of essential oils against *A. craccivora* (Dutra *et al.*, 2020 and Sayed *et al.*, 2020). In earlier research, Abdul Kareem *et al.* (1989) suggested the use of neem for rice before sowing to control two homopterous pests. Nowadays, there has been a growing interest by many authors in the use of essential oils as an alternative to synthetic pesticides for seed dressing. Suradkar and Ukey (2014) studied the effect of seed treatment by neem, karanj, mahua and eucalyptus oils in controlling thrips population on tomato. So, the present work aimed to examine the seed treatment of camphor, mint and basil oils and their toxic and repellent effects on *Aphis craccivora*, also, the effect of using dry and sprouting seeds was investigated.

MATERIALS AND METHODS

All experiments were carried out in the laboratory of Plant protection, Shandweel Agricultural Research Station, Sohag Governorate under room conditions where the ambient temperature ranged between 11 and 25 °C, and RH ranged from 44% to 60%.

1. – *Aphis craccivora* culture:

Aphis craccivora was reared in the Laboratory of Plant protection at Agricultural Research Station of Shandaweel, Sohag Governorate. Aphids were obtained from infested faba bean plants at the Farm of Agricultural Research Station of Shandaweel. Seeds of faba bean were raised in small plastic pots (11 cm x 14 cm) which were kept in a wooden screen cage (1×1×1m) covered with an anti-aphid screen under laboratory conditions. After that, the collected aphids were placed on the growing seedlings. On new un-infested faba bean seedlings, the artificial infestations by aphids were successively repeated.

2. - Oils and plants:

The three essential oils were chosen based on the market availability, and also on the previous assays that showed potential toxicity to *A. craccivora*. The selected oils were supplied as ready-made oil from El-Captain Company for Extraction of Natural oils, Plants and Cosmetics, Cairo, Egypt. The essential oils of camphor (*Cinnamomum camphora* L.: Myrtaceae), mint (*Mentha piperita* L.: Labiatae), and basil (*Ocimum basilicum* L.: Lamiaceae) were used. The seeds of faba bean (variety weam) were obtained from The Agricultural Research Center at Giza.

* Corresponding author.

E-mail address: abuelfadelmohamed400@gmail.com

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3. - Seed treatment:

Two methods were used to treat seeds by the three oils, the first one was the use of dry seeds, however, the second one was the use of sprouting seeds as modification from a systemic method. Faba bean was sprouted for 36 hours, and then each dry and sprouting seed was treated with the three oils at the rate of 60 ml/ 1 Kg. seeds. Then ten seeds were sown in plastic pots (30 cm in diameter and 55 cm high) with soil taken from the field. The plants were left to grow under natural environmental conditions. Toxic and repellent experiments were started when the plants were two weeks old.

4. - The repellent effect:

The free choice test was used to evaluate whether the seed treatment with oils as dry and sprouted affected the response of *A. craccivora* behavior (repellent effect). From each treatment, one leaflet was put on a Petri dish (15 cm diameter with wetted filter paper) against an untreated one, then twenty adults were released in the center of each Petri dish. The numbers of aphid settled on treated and untreated leaflets were counted after 1 and 24 hours. This experiment was arranged in a complete randomized design with five replicates. The data were expressed as a percentage of repulsion (PR) using the following formula:

$$PR (\%) = (Nc-50) \times 2$$

Where, Nc = the percentage of *A. craccivora* found in the control leaflets.

The mean values were categorized as 0: > 0.01 to < 0.1%, I: 0.1 to 20%, II: 20.1 to 40%, III: 40.1 to 60%, IV: 60.1 to 80% and V: 80.1 to 100% according to the scale of McDonald *et al.*, (1970).

5. - The toxic effect:

After 15 days from seeds sowing, fresh young faba bean leaves were collected, each leaf separately placed in

each Petri dish (1 cm height × 9 cm diameter) which contained a wetted filter paper. Ten aphid adults were released on each Petri dish with the help of a camel-hair brush. The six treatments were arranged in a complete randomized design with five replicates. Adult mortality was recorded at 1, 2, 3, 4, and 5 days after treatment. Mortality data were corrected by Abbott's (1925) formula:

$$\text{Corrected \% mortality} = ((X - Y) / (100 - Y)) \times 100$$

Where: X: % mortality in treatment and Y: % mortality in control.

6. - Statistical analysis:

Data obtained were analyzed as a complete randomized design. Means values were distinguished using the Duncan Multiple Range Test at P = 5% (Snedecor and Cochran, 1971).

RESULTS AND DISCUSSION

1. - Repellent effect:

Data in Tables (1) and (2) show the effect of the seed treatment method and oil on the acceptance of *A. craccivora* for faba bean leaves after 1 hour and 24 hours, respectively. It is clear that the differences between the six seed treatments were significant, and the three oils proved a repellent effect (≥ 50%) against *A. craccivora* adults after 1 and 24 hours.

After 1 hour, the highest repellent effect was observed on sprouting seed treatment with basil oil (73.68%), followed insignificantly by dry seed treatment with basil oil (71.43%), sprouting seed treatment with mint oil (70.00%), and sprouting seed treatment with camphor oil (67.74%), however, the lowest repellent effect was observed on dry seed treatment with camphor oil (50.00%). After 1 hour, the repellency class of dry seed treatment with camphor oil was III, however, the rest treatments were in the repellency class of IV.

Table 1. The effect of two seed treatment methods and three essential oils on repellent of *A. craccivora* on faba bean after 1 hour.

Seed treatment	Oil	Non responded%	Responded%		Repellent %	Class
			Treatment	Control		
Dry seeds	Camphor	44.00	14.00	42.00	50.00 c**	III
	Mint	36.00	12.00	52.00	62.50 b	IV
	Basil	30.00	10.00	60.00	71.43 ab	IV
Sprouting seeds	Camphor	38.00	10.00	52.00	67.74 ab	IV
	Mint	20.00	12.00	68.00	70.00 ab	IV
	Basil	24.00	10.00	66.00	73.68 a	IV
F. value		-----	-----	-----	9.293*	-----

(*): The F value is significant at P ≤ 0.05

(**): Means followed by different letters within the column are significantly different from each other (P < 0.05) by Duncan, s Multiple Range Test. III: 40.1 to 60% and IV: 60.1 to 80%

For the repellent effect after 24 hours, the six treatments were arranged in two significantly groups (Table 2), the first and the highest consisted of dry seed treatment with basil oil (75.61%) and sprouting seed treatment with camphor oil (70.00%), with mint oil (76.74%) and with basil oil

(75.61%), however, the dry seed treatment with camphor oil (57.58%) and with mint oil (60.00%) found on the second and the lowest group. All treatments of the first group were in the repellency class of IV, however, all treatments of the second group were in the repellency class of III.

Table 2. Effect of interaction between two seed treatment methods and three essential oils on repellent of *A. craccivora* on faba bean after 24 hours.

Seed treatment	Oil	Non responded%	Responded%		Repellent %	Class
			Treatment	Control		
Dry seeds	Camphor	34.00	14.00	52.00	57.58 b**	III
	Mint	20.00	16.00	64.00	60.00 b	III
	Basil	18.00	10.00	72.00	75.61 a	IV
Sprouting seeds	Camphor	20.00	12.00	68.00	70.00 a	IV
	Mint	14.00	10.00	76.00	76.74 a	IV
	Basil	18.00	10.00	72.00	75.61 a	IV
F. value		-----	-----	-----	10.944*	-----

(*): The F value is significant at P ≤ 0.05

(**): Means followed by different letters within the column are significantly different from each other (P < 0.05) by Duncan, s Multiple Range Test. III: 40.1 to 60% and IV: 60.1 to 80%

In general, data indicated that the use of sprouting seeds increased the repellent effect of the three oils compared with using dry seeds after 1 and 24 hours. For oils, their repellency is arranged as basil oil > mint oil > camphor oil.

In the same line as the present results, Sammour et al. (2011) indicated that basil oil had an antifeedant effect on the nymphal stage of *A. craccivora*, Wubie et al. (2014) concluded that mint has repellent activity against *Brevicoryne brassicae* aphid, also in close proximity with those of Yasmin et al. (2017) who indicated that the highest (77.33%) and the lowest (34.0%) repellency of *A. craccivora* was found on mahogany and karanja oils, respectively.

For the two seed treatment methods and the three oils proved, the repellent activity increased with increasing time; however, the number of non-responded aphids decreased with time, the same results were obtained in previous studies by El-Solimany (2020) on *A. craccivora* and Youssef and Khorchid (2020) on *Sesamia cretica* larvae.

2. - Toxicity effect:

The data in Table (3) show the efficiency of camphor, mint, and basil oils as seed dressing by two methods (dry and sprouting seeds) on *A. craccivora* adults after 1, 2, 3, 4, and 5 days, also, the mean effect was presented in Fig. (1).

Table 3. The effect of two seed treatment methods and three essential oils on mortality of *A. craccivora* on faba bean.

Seed treatment	Oil	Mortality% after				
		1 day	2 days	3 days	4 days	5 days
Dry seeds	Camphor	0.00	18.00 d**	56.25 c	74.47 c	91.11 b
	Mint	0.00	26.00 c	77.08 ab	82.98 b	95.56 ab
	Basil	0.00	36.00 b	72.92 b	82.98 b	97.78 a
Sprouting seeds	Camphor	0.00	36.00 b	83.33 a	95.74 a	100.00 a
	Mint	0.00	34.00 b	79.17 ab	93.62 a	100.00 a
	Basil	0.00	48.00 a	77.08 ab	93.62 a	100.00 a
F. value		-----	19.425*	20.677*	10.400*	4.400*

(*): The F value is significant at $P \leq 0.05$

(**): Means followed by different letters within the column are significantly different from each other ($P < 0.05$) by Duncan, s Multiple Range Test.

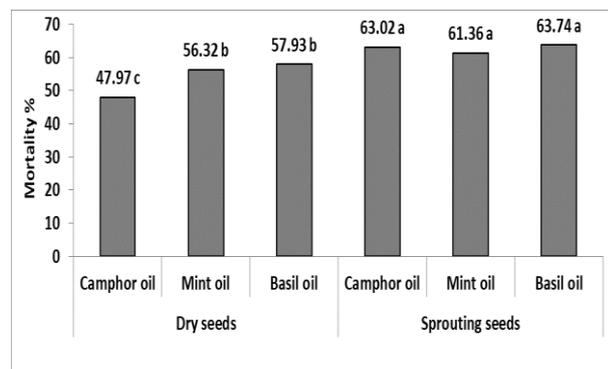


Figure 1. The effect of two seed treatment methods and three essential oils on mortality of *Aphis craccivora* on faba bean. F. value= 45.029 Different letters next to each bar indicate significant differences at $p < 0.05$ by Duncan's Multiple Range Test.

It is clear that no mortality percentage was observed within 1 day from treatment. After 2 days, the mortality percentage was almost low to moderate, however, a sharp increase was observed after 3 days, then mortality slightly increased after the last two days in all treatments. The differences between the six seed treatments were significant after 2, 3, 4, and 5 days.

After 2 days, the highest and the lowest mortality percentages were recorded on sprouting seed treatment with basil oil and dry seed treatment with camphor oil, respectively, by 48.00% and 18.00%, respectively.

After 3 days, the highest mortality percentage was recorded on sprouting seed treatment with camphor oil by 83.33%, followed insignificantly by dry seed treatment with mint oil (77.08%), sprouting seed treatment with mint oil (79.17%), and with basil oil (77.08%), while, the lowest mortality percentage was recorded on dry seed treatment with camphor oil with 56.25%.

After 4 days, the six treatments were arranged into three significantly groups, the first and the highest one contained sprouting seed treatment with camphor oil

(95.74%), mint oil (93.62%), and with basil oil (93.62%), followed by dry seed treatment with mint oil (82.98%) and with basil oil (82.98%) in the second one, while the last and the lowest one consisted of dry seed treatment with camphor oil (74.47%).

Sprouting seed treatment with camphor, mint, and basil oils recorded 100.00% mortality in adults of *A. craccivora* after 5 days, followed insignificantly by dry seed treatment with basil oil (97.78%) and with mint oil (95.11%), however, the lowest mortality was recorded on dry seed treatment with camphor oil (91.11%).

Depending on the mean effect, the six combinations between two seed treatment methods and three oils were arranged into three significantly groups (Fig. 1). The first and the highest one contained sprouting seed treatment with camphor oil (63.02%), with mint oil (61.36%) and with basil oil (63.74%). The second one consisted of dry seed treatment with mint oil (56.32%) and with basil oil (57.93%). While dry seed treatment with camphor oil (47.97%) was the last and the lowest one.

For seed treatments, Suradkar and Ukey (2014) revealed that seed treatment by neem, karanj, mahua and eucalyptus oils can be effective control method against thrips on tomato. Also, Abdel-Aziz et al. (2015) reported that the soil treatments with oils of rosacide, sagix and cura have toxic effect against *A. craccivora* especially at the highest concentrations, treatment with the previous oils at 0.1% caused 66.67, 53.33 and 49.98 % mortality, respectively.

For oils, in previous studies, Adly and Bakr (2016) found that camphor fumigant can be suggested as a control agent against *A. craccivora*. Mesbah et al. (2016) found that camphor oil and red basil oil reduced the infestation of *A. gossypii* on cucumber by 60.17% and 56.94%, respectively, after 3 days. Aziz et al. (2018) indicated that the mortality percentage of *A. craccivora* adults ranged between 35.1% to 77.6% for neem oil and between 21.3% to 57.6 for cinnamic oil after seven days, depending on concentration.

The foregoing results suggested that the seed treatment by camphor, mint, and basil oils was effective on

A. craccivora. The use of basil oil on sprouting faba bean as seed treatment holds great promise for application against *A. craccivora* control as the eco-friendly method.

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السمية والتأثير الطارد لثلاثة زيوت كمعاملة بذرة علي من الفول تحت ظروف المعمل عصمت احمد السليماني و محمد عبدالمعطي احمد ابو الفضل معهد بحوث وقاية النباتات – مركز البحوث الزراعية.

الملخص

أجريت هذه الدراسة في المعمل لبحث التأثير الطارد والسمية لمعاملة بذور الفول الرومي بزيت الكافور وزيت النعناع وزيت الريحان علي من الفول. كذلك تم دراسة تأثير استخدام البذور الجافة والمستنبتة. أوضحت النتائج أن استخدام البذور المستنبتة زاد من التأثير الطارد والسمية للزيوت المستخدمة مقارنة باستخدام البذور الجافة. وجد أنه بعد ساعة سجل أعلى تأثير طارد عند استخدام زيت الريحان علي البذور المستنبتة (73,68%) يليه بشكل غير معنوي زيت الريحان علي البذور الجافة (71,43%). زيت النعناع علي البذور المستنبتة (70,00%) و زيت الكافور علي البذور المستنبتة (67,74%). بينما بعد 24 ساعة سجل أعلى تأثير طارد عند استخدام زيت النعناع علي البذور المستنبتة (76,74%) يليه بشكل غير معنوي زيت الريحان علي البذور المستنبتة (70,61%) وزيت الريحان علي البذور الجافة (70,61%) وزيت الكافور علي البذور المستنبتة (70,00%). لم يلاحظ أي نسب للموت بعد يوم من المعاملة. كانت نسبة الموت علي الأغلب منخفضة إلي متوسطة بعد يومين من المعاملة. بينما تم ملاحظة زيادة حادة في نسبة الموت بعد 3 أيام. بعد ذلك زادت نسبة الموت بشكل طفيف خلال اليوم الرابع واليوم الخامس في كل المعاملات. استخدام زيت الريحان علي البذور المستنبتة حقق أعلى لنسبة موت في من الفول بمتوسط 63,74% يليه بشكل معنوي زيت الكافور علي البذور المستنبتة (63,02%) وزيت النعناع علي البذور المستنبتة (61,36%).