

TOXICOLOGICAL AND BIOLOGICAL STUDIES
OF CERTAIN PLANT EXTRACTS ON *APHIS*
GOSSYPHII GLOVER

Hegazy, H. Wafaa; Roud, A. El-Doksh; S. A. Aref

Plant protect. Res. Inst. Sakha Agric. Res. Station Kafr El-Sheikh

ABSTRACT

The present study was directed to evaluate the residual effect of two plant extracts (black cummin and wormseed) on the cotton aphid, *Aphis gossypii* (Glover). Results indicated that the black cummin, as a plant extract was more toxic to adults of *Aphis gossypii* than wormseed with LC₅₀ values (140.42 and 1704 ppm.) respectively. The duration of different instars was longer than that of the untreated ones. Black cummin significantly increased the duration period of the first instar while no significant difference was found between wormseed and control. Plant extracts decreased the number of nymphs than the control (1.67 and 2.67 nymphs per individual for black cummin and wormseed respectively), while control showed 5.00 nymphs/individual. The total mortality was 60.01 and 40% during instars treated with black cummin and wormseed respectively. The duration of offspring was significantly decreased to 2.33 and 3.33 days for treated individuals with black cummin and wormseed compared with 5.67 days for the control. The results proved that black cummin was more effective extract on duration of offspring duration of *Aphis gossypii*.

INTRODUCTION

The cotton aphid, *Aphis gossypii* (Glover), has become, in recent years a deleterious pest infesting wide range of crops in Egypt. Therefore, laboratory screening of

insecticides against this insect was required to find an effective control program (Haydar *et al.* 1996).

Aphid is one of the most noxious insects for many crops. It causes leaf curling and stunting in plants and also transmits viral diseases. *A. gossypii* (Glover), is a common vector of viruses (Converse, 1993). Currently, aphid population is controlled by chemical insecticides which are potentially, hazardous, environmentally unsafe and insects frequently develop resistance to them. The development of alternative control methods is, therefore, urgently required. In green house, the problem of aphids becomes serious due to their reproduction in very short period (Converse, 1993).

In Egypt, *A. gossypii* (Glover) attack squash plants causing dangerous problem either directly or indirectly giving a reduction in the crop yield. The development of injurious population of the pest is associated with the increased use of insecticides which are ineffective against the pests and are known to create problems by destroying their natural enemies (Shaheen *et al.*, 1973).

Many authors concluded that plant extracts were effective against *A. gossypii* (Guirguis *et al.*, 1989, 1991; Matter *et al.*, 1993; Rahbe *et al.*, 1995; El-Gougary, 1998; El-Hariry *et al.*, 1998; Tunc and Shainkaya, 1998).

Abassy *et al.*, (1998) found that the acetone/ethanol extract was toxic to *A. gossypii* followed by lycerine, soosan oil and crude alkaloids. Also, synergism studies on *A. gossypii* indicated that lycerine, the principal alkaloid of soosan bulbs, strongly synergized cyanophos, reducing its LC₅₀ value from 120 to 48 p.p.m. Al-Shayji *et al.*, (1998) determined the insecticidal activities of five preparations of *Bacillus thuringiensis* (Bt) on adults and nymphs of *A. gossypii*. They found that both nymphs and adults were highly susceptible to Bactospeine and Dipel, whereas Thuricide 64 was the least toxic. El-Gougary (1998) found that the different solvent-extracts of *Atriplexhalimus*, (ether and petroleum-ether extracts) showed strong aphicidal activity against *A. gossypii* with LC₅₀ values of 0.059 and 0.085%, respectively.

Nassef (1998) indicated that vegetative oil (black cumin) % initial kill ranged (78.8-93.1) and average % reduction (61.3-73.1) on cotton and squash respectively against *A. gossypii*. Metwally *et al.*, (1999) indicated that natural oils gave a high percentage of initial mortality on the population of *A. gossypii*.

The present study was carried out to investigate the toxic effect of two plant extracts (black cumin and wormseed) against adults of *A. gossypii* (Glover). In addition the side effect of sublethal doses of the tested extracts on some biological aspects of *A. gossypii* was also studied.

MATERIALS AND METHODS

Aphid culture:

The aphid used in this study was identified as *A. gossypii* (Glover), (Homoptera : Aphididae). *Hibiscus esculentus* plants were used for aphid mass-rearing. Okra plants were put in cages and the aphids were always transferred weekly from old to young seedlings, by cutting the heavily infested leaves to transfer them to new plants. The aphid culture was kept at $25\pm 2^{\circ}\text{C}$ and 65 ± 5 R.H.

The tested plants :

Black cumin (*Negella sativum* Linn). and wormseed (*Artemisia cinnae* L.) were selected in the present study. The plants were obtained from the local market at Kafr El-Sheikh.

Plants extracted and investigated in the present study.

Scientific Name	Arabic Name	Common Name	Parts Used
Fam. <i>Ranunculaceae</i> <i>Negella sativum</i> Linn.	الحبة السوداء	Black cumin	Seeds
Fam. <i>Compositae</i> <i>Artemisia cinnae</i> L.	الشيح الخرساني	Wormseed	Leaves

Plant extract preparations :

A plant sample of wormseed leaves was thoroughly washed in detergent solution followed by rinsing in tap water. Plants were air dried at room temperature then in an

oven at 40°C and then they were grounded to fine powder. The powder was divided into batches each weighed 100 gm, which was macerated in 300ml of acetone/methanol (1:1 v/v) for 72 hours in dark bottle. During the maceration period sample was shaken for 6 hours using an electric shaker. Extract was filtered and placed in a refrigerator for 24 hrs, then was refiltered, dried over an-hydrous sodium sulphate and evaporated to dryness using a rotary evaporator. The residue was considered as active ingredient, weighted and dissolved in acetone to form the required concentrations. Black cumin seeds powder were extracted by absolute ethanol (5.0 gms/100.0 ml). Extraction run over night followed by filtration.

The dosage rates of the dissolved plant extracts were calculated as p. p.m., on the basis of the weight of solid residues (which considered as active ingredient).

Experimental techniques :

1. Leaf disc:

The pepper plants or okra plants were cultured under the greenhouse conditions. When young plants bearing only 4 primary leaves, they were considered to be suitable to carry on the desired experimental tests.

The toxic effect of the tested extracts were evaluated by leaf disc-dip technique according to Siegler (1947). Leaf disc experiments were conducted at $25\pm 2^{\circ}\text{C}$. Ten adult females of *A. gossypii* were transferred to each disc, these discs were dipped in different concentrations of each plant extract. The discs were placed on moist filter paper, which rested on moist cotton wool bad contained in petri dishes and kept under controlled conditions of $25\pm 2^{\circ}\text{C}$ & 65 ± 5 R.H. Each treatment was replicated four times. Mortality counts were made 24 hours after treatment.

2. Dipping of whole plants:

To assay the residual effect of the tested compounds on biological aspects of aphid such as, reproduction, developmental stages, mortality of different instars and

offspring; the method of Smith *et al.*, (1963) was used. The plants were infested with 8 adults each. These plants were dipped in the required concentration of plant extracts, Aphids (newly born) were transferred on the treated plants. Five individuals of the first instar were transferred on each plant. Each treatment was replicated four times. First instar was allowed to complete its development on treated plants and the duration of each instar as well as the number of dead individuals were recorded. When nymphs reached the adult stage, newly born nymphs were counted. The experiment were conducted under the controlled condition of $25\pm 2^{\circ}\text{C}$ & 65 ± 5 R.H.

RESULTS AND DISCUSSION

1. Toxicity of tested extracts against *A. gossypii* adults:

Two plant extracts (black cumin and wormseed) were tested for their toxicity to adult stage of laboratory strain of *A. gossypii* by the leaf disc technique, Probit regression lines were statistically analyzed according to Finney (1952) supplying information on confidence limits of LC_{50} and slope values.

Based on LC_{50} values (Table 1), the results indicated that the black cumin as a plant extract was more toxic than wormseed to adult females of *A. gossypii* with LC_{50} values of 140.42 and 1704 p.p.m. respectively. The slope values of log concentration probit lines showed in Table (1) indicated that black cumin has higher slope value (1.68), while wormseed has slop value of (1.58).

Plant extracts were found effective against *A. gossypii* by many investigators (Dhandapani *et al.*, 1985; Guirguis *et al.*, 1989; Guirguis *et al.*, 1991; Lowery, *et al.*, 1993; Matter *et al.*, 1993; Rahbe *et al.*, 1995; Hutton *et al.*, 1996 and Tunc and Sahinkay, 1998). Nassef 1998 obtained vegetative oil (black cumin) % initial kill ranged from 78.8 to 93.1 and average% reduction of 61,3 and 73.1 on cotton and squash respectively against *A. gossypii*. Metwally *et al.*, (1999) indicated that natural oils gave a high percentage of initial mortality on the population of *A. gossypii*.

Table (1): Toxicity of two plant extracts against *A. gossypii* adults

Plan extracts	LC ₅₀ p.p.m	Confidence Limits of LC ₅₀		Slope value
		Lower	Upper	
Black cumin	140.42	99.42	186.73	1.68
Wormseed	1704.0	1228	2230	1.58

2. Effect of two plant extracts on biology, of *A. gossypii*.

2.1. Effect of tested extracts on nymphs developmental time, percentage of mortality of different instars and offspring of *A. gossypii*.

The tested extracts against *A. gossypii* caused a prolongation of development as well as a reduction in offspring whether in newly emerged nymphs, the duration of different instars was longer than the untreated ones, Table (2).

Black cumin significantly increased the duration period of the first instar while no significant differences were found between wormseed and control. The durations values were 2.25 and 2.00 days for black cumin and wormseed respectively, compared with 1.33 days for the control. The duration of the second instar was longer for black cumin and wormseed (1.83 and 1.50 days) compared with 1.17 days in the control. The duration of the third instars was longer for black cumin and wormseed extracts (2.2 and 1.8 days) compared with 1.7 days in the control. The duration of the fourth instar was longer for black cumin and wormseed (1.67 and 1.67 days) compared with 1.33 days in the control.

Plant extracts (black cumin and wormseed) decreased the number of nymphs than the control (1.67 and 2.67 nymphs per individual for black cumin and wormseed respectively), while control showed 5.00 nymphs/individuals. The total mortality was 60.01 and 40% during instars treated with black cumin and wormseed respectively. These results are in agreement with that recorded by (Sheila *et al.*, 1991; Kerns and Gaylor, 1992 and Hollingsworth *et al.*,

1997). Nikolov and Draganov (1998) found that high insecticidal activity from some extracts (Various infusions were made from hot pepper fruits at a concentration of 1%), which caused 86-93% aphids mortalities on cucumber and pepper.

Abd El-Wahab (2003) stated that castor oil application appeared to be the most effective against the population of *A. gossypii* followed by petroleum ether, hexane and water extracts (leaves extracts of castor plants).

Table (2): Effect of sub-lethal concentrations of extracts on nymph developmental time, percentage of mortality of different instars and offspring of *A. gossypii*.

Plant extracts	Developmental stages and percentage of mortality									
	Offspring / individual [*]	1 st instar	Mortality %	2 nd instar	Mortality %	3 rd instar	Mortality %	4 th instar	Mortality %	Offspring/individual ^{**}
Black cumin	2.33±0.58 b	2.50±0.50 a	40	1.83±0.29	6.67	2.2±0.29	6.67	1.67±0.29	6.67	1.67±0.58
Wormseed	3.00±1b	2.00±0.50 ab	20	1.50±0.50	13.33	1.8±0.29	0.0	1.67±0.29	6.67	2.67±0.58
Control	4.67±0.58 a	1.33±0.29 b	6.67	1.17±0.29	0.0	1.7±0.29	0.0	1.33±0.29	0.0	5.00
L.S.D.										
5%	1.49	0.88	-	-	-	-	-	-	-	1.49
1%	2.26	1.33	-	-	-	-	-	-	-	2.26

Means followed by a common letter are not significantly different at the 5% level by Duncan's multiple range test (1955).

* after treatment

** after generation

2.2. Effect of extracts on duration of offspring and newly emerged nymphs of *A. gossypii*.

The duration of offspring was significantly decreased to 2.33 and 3.33 days of treated individuals with black cumin and wormseed compared with 5.67 days for the control, respectively.

The results in Table (3) proved that black cumin extract was the most effective followed by wormseed extract on duration of offspring of *A. gossypii*.

The present results are in agreement with the findings of Bachatly *et al.*, (1995) who found that the duration of the

different instars were less prolonged with sorrel extract. Prolongation as well as reduction of offspring numbers produced significantly fewer nymphs than those of the control.

Table (3): Effect of sublethal concentrations of plant extracts on duration of nymphal stage and offspring of newly emerged nymphs of *A. gossypii*.

Treatment	Nymphal stage	Offspring
Black cumin	8.17 ±0.58a	2.33± 0.58 b
Wormseed	7.00±0.50b	3.33±0.58b
Control	5.50±0.50c	5.67±0.58a
L.S.D.		
5%	1.05	1.15
1%	1.60	17.5

Means followed by a common letter are not significantly different at the level by DMRT.

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الملخص العربي

دراسات تكسوكولوجية وبيولوجية لبعض المستخلصات النباتية على حشرة المن

وفاء حسن حجازي ، روض الدكش ، صفوت عارف
معهد بحوث وقاية النباتات محطة للبحوث لزراعية بسخا
كفر الشيخ

لقد وجهت الدراسة الحالية لتقييم النشاط الإبادي لإثنان من المستخلصات النباتية (بنور حبة البركة ، والشيح الخرساني) ضد حشرة المن. ودلت النتائج على أن مستخلص حبة البركة - كان أكثر سمية على الطور الكامل للمن من مستخلص الشيح الخرساني- حيث بلغت قيم LC_{50} 17.04 ، 140.42 جزء في المليون على الترتيب وكانت فترة حياة الحوريات للأعمار المختلفة طويلة بالمقارنة بالغير معاملاً وأن مستخلص حبة البركة كان له تأثيراً معنوياً وعالياً على إطالة فترة حياة الحوريات للأعمار الأولى بينما لا يوجد فروق معنوية بين مستخلص الشيح الخرساني والكنترول. المستخلصات النباتية (حبة البركة- الشيح الخرساني) أدت إلى انخفاض أعداد الولادات الناتجة من الأطوار الكاملة بالمقارنة بالكنترول حيث كانت 1.67 و 2.67 لكل من

مستخلص حبة البركة والشيخ الخرساني مقارنة بالكنترول حيث كان ٥ . وأيضا نسبة الموت حيث كانت ٦٠,٠١ ، ٤٠ % لكل من حبة البركة والشيخ الخرساني وكانت فترة النسل تقل معنويا لكل من مستخلص حبة البركة والشيخ الخرساني حيث كانت ٢,٣٣ ، ٣,٣٣ يوماً على الترتيب مقارنة بالكنترول الذي كان ٥,٦٧ يوماً وأثبتت النتائج أن مستخلص حبة البركة أشد تأثيراً على فترة النسل لحشرة المن يليه مستخلص الشيخ الخرساني.