INSECTICIDAL ACTIVITY OF ETHANOL AND AQUEOUS EXTRACTS OF MEDICINAL PLANTS AGAINST GREEN PEACH APHID (*MYZUS PERSICAE)*

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Abstract. The green peach aphid *Myzus persicae* (Homoptera: Aphididae) is one of the most common and harmful pests. Plant extracts contain many active compounds for defense against pests, and an extensive search for effective and ecological pest control alternatives, particularly those derived from botanicals, is currently underway. This study evaluated the effectiveness of five botanicals from *Citrullus colocynthis, Nicotiana tobaccum* L., and seeds of *Trachyspermum ammi, Azadirachta indica,* and *Withania coagulans* against *Myzus persicae*. Botanical oils were extracted using ethanol oil extraction (EOE) from powder and extraction through boiling (ETB) of powder and tested for insecticidal efficacy in a laboratory and a greenhouse. The highest mortality (81.7%) was caused by *T. ammi*, followed by *C. colocynthis* (76.5%), *N. tobaccum* (63.0%), *A. indica* (56.5%), and *W. cagulans* (50.0%). EOE resulted in a higher mortality at 48-h intervals, and fumigation resulted in the highest mortality at 24-h intervals, with the EOE extracts effectively reducing female aphid fecundity. EOE also was more effective against aphids in cucumber than ETB in greenhouse. Taken together, the EOE extracts were more effective than ETB, and all plant extracts possessed significant insecticidal properties, with potential as botanical insecticides for use in integrated pest management programs.
Keywords: *essential oils, mortality, contact toxicity, repellency, fecundity*

Introduction

The green peach aphid *Myzus persicae* (Hemiptera: Aphididae) is one of the most prominent and destructive pests. Green peach aphids feed on over 800 species of plants, including ornamental plants (Van Driesche et al., 2008), vegetables (Hofsvang and Hågvar, 1979; Freuler et al., 2003), fruits (Kim and Kim, 2004), and weeds, but have also been found to attack barley, rye, and winter wheat, as well as on potatoes in northern climates (Davis and Radcliffe, 2008). This aphid is extremely polyphagous and transmits more than 100 plant viruses. It has a wide range of genetic variations in color, life cycle, host-plant relationships, and methods of resisting insecticides (Blackman and Eastop, 2007; Davis and Radcliffe, 2008). Because of its high reproductive rate and short generation time, the green peach aphid is able to adapt to insecticides and develop resistance to active compounds rapidly (Machial, 2010).

Insecticides, such as organophosphates, pyrethroids, and modern chemical insecticides, have been shown to be effective in controlling insects (Umrao et al., 2013). However, due to the widespread use of these insecticides, many pests have developed insecticide resistance (Ahmad and Arif, 2009; Jan et al., 2015). Conventional insecticides have long been used to manage insect pests. Although the dangers and drawbacks of synthetic insecticides were not anticipated when they were first introduced, their use has led to disruptions in the natural balance via their effects on natural enemies, pollinators and other wild life, as well as the evolution of tolerance and the reappearance of treated

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populations (Vitousek et al., 1997). According to the World Health Organization, pesticide contamination kills 200,000 people globally each year. Insecticidal toxicity, a high risk for disease, residual toxicity, biomagnifications, use limits, and poor environmental impacts have led to a rise in the use of botanical insecticides (Feng and Zheng, 2007; Khater, 2012; Tomé et al., 2013). In addition to biological regulation, pheromones, and plant-derived chemicals, new pest management techniques are urgently needed. Despite the fact that pesticide resistance is widespread, certain mechanisms of insecticide resistance have been identified. The first report of pesticide resistance in *M. persicae* species dates back to 1955 (Anthon, 1955). Resistance to a key class of pesticides includes organophosphates, carbamates, cyclodienes, pyrethroids, and neonicotinoids has now made this species the most significant and robustly resistant species on aphid have been found (Bass et al., 2014).

Controlling harmful pests with synthetic chemical insecticides is crucial for new farming practices, improving crop yield as a result. However, the indiscriminate use of synthetic pesticides for agricultural production and safety has been a source of carcinogenesis, reproductive issues, and mutagenesis in humans via contamination through touch, inhalation, and dietary exposure (Kazem and Shereif, 2010; Lu et al., 2018). These circumstances have prompted a search for efficient and environmentally friendly pest control alternatives, especially those extracted from natural plant resources (Fetoh and Asiry, 2012). Many insecticides originating from botanical sources are readily available, inexpensive, open to farmers, safer for humans, with negligible residual effects, as well as being target-specific and less harmful to vertebrates, pollinators (Batta, 2004; Isman, 2006; Koul et al., 2008).

Due to these challenges, researchers and farmers have been exploring alternative insect control options under the integrated pest management program (IPM). IPM integrates all available resources to control insect species that inflict economic damage. Mechanical control, cultural traditions, biological agents, and botanical extract practices are also part of IPM. In terms of the severity and nature of insect attacks, these are optional. Alkaloids, terpenoids, flavonoids, phenols, glycosides, sitosterols, and tannins are only some of the secondary metabolites present in plants (Gulzar et al., 2019). While plant compounds may be hazardous to variety of insect species when consumed, antifeeding action disrupting (molt inhibitory or anti-molting) (Ulrichs et al., 2008). Plant extracts and botanical pesticides have become increasingly popular in agricultural fields in recent years because of their low cost, lack of residual effects, environmentally friendly nature, and toxic effects on important pests, including aphids, jassids, whiteflies, trips, and mites (Stumpf and Nauen, 2001). Since they are safe for beneficial species, target-specific, and compatible with biological control agents, neem oils and extracts are considered the best choice for insect pest management in vegetables (Tang et al., 2002). Tobacco, sweetsop, and garlic extracts, in particular, have been found to be effective biopesticides for controlling cowpea insect pests (Ahmed et al., 2009). *Citrullus colocynthis*, a botanical insecticide, has also attracted the interest of researchers, and the efficacy of its extracts and isolated compounds has been tested against commercially significant insect pests. Several pests were found to be repelled by this plant's antifeedant, deterrent, and infertility effects (Seenivasan et al., 2004). Studies on the effects of *Citrullus colocynthis* fruit, leaf, stem, and root extracts of *Rhopalosiphum padi* L. found that the stem extract was more effective against this pest than the other component extracts (Khalid, 2015). The antimicrobial effects of *Withania coagulans* and *Withania munifera* leaves, roots, and fleshy buds were also tested, and were found to have an inhibitory effect on bacterial

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and *Fusarium* wilt, as well as being effective against bacterial Fusarium wilt in tomatoes (Najeeb et al., 2019; Khan et al., 2019). *Anopheles stephensi* larvae are repelled by *Trachyspermum ammi* shoots and leaves from the Blue Mint Bush (*Ziziphora clinopodioides*). Previous studies have also used ajwain oil at various concentrations and exposure periods to fumigate adult *Oryzaephilus surinamensis*, *Rhyzopertha dominica*, and *Tribolium confusum* (Habashi et al., 2011; Torabi et al., 2017). In the present study, the insecticidal effects of different botanicals were evaluated to identify safe alternatives for the management of insect pests. Botanical oils were extracted using EOE and ETB and tested for insecticidal efficacy under both laboratory and greenhouse conditions.

Material and methods

Preparation of plant extracts

The five botanicals used were *Azadirachta indica*, *Withania coagulans Trachyspermum ammi*, and leaves of *Nicotiana tobaccum* L. *Citrullus colocynthis*. Botanical oil was extracted using two methods: ethanol oil extraction from EOE and ETB. Either the seeds or leaves of each botanical plant were washed before being dried in an oven at 45°C for 48 h. The dried material was then ground and pushed through a sieve with a 40-mm mesh. The EOE (ethanol oil extraction from powder), the powder was separated with 95 percent ethanol (1 g of powder in 5 ml of ethanol), and the mixture was stored in bottles at room temperature (20–25°C) for 7 days. To ensure the proper mixing and dissolution of the powder in ethanol, the bottles were shaken twice daily. After filtering the mixture with filter paper, the residuals were dissolved using the same method solution with a ratio. In a rotary evaporator, the first and second solvents were mixed and dried until all of the liquid was evaporated. The oil was collected in a brown collection bottle and stored at 4°C. Then, 0.3 mL of dimethyl sulfoxide (DMSO) with 1% Tween-20 was added to 5 mL of the crude extract solution. The ETB dried powder content was boiled separately for 30–40 min in 2 liters of water, and then sieved through muslin cloth. Each mixture was stored in its own bottle at 4°C. For the control solution, 0.3 mL of DMSO, 1%Tween-20, and 5 mL of double-distilled water were mixed together. The essential oil was extracted using the technique reported by Su et al. (2009).

Laboratory conditions

The aim of the laboratory experiment was to validate the efficacy of various plant extracts and their effects on aphid mortality. The experiments were performed at Langfang Research station laboratory of Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China. The toxicity of each treatment was investigated through fumigation, contact, and fecundity experiments. Complete randomized design CRD with five replications was used for the laboratory experiments.

Test 1: Fumigant toxicity

For this experiment, 0.5 ml of each essential oil in solution was applied onto 9-cm-diameter filter paper using a micropipette after being soaked separately. Thereafter, the dried filter paper was kept on the underside of the petri dishes. Thirty aphids (2^{nd} and 3^{rd} instar) were placed in 9-cm petri dishes, which were sealed with polythene strips. The petri dishes were incubated at 25 ± 2°C, 50% RH, and a photoperiod of 10:14 h L:D. Throughout the bioassay test, the petri dishes were examined at 2, 6, 12, and 24 h to assess

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fumigant toxicity against each extract aphid, observed in terms of the percentage of mortality.

Test 2: Contact toxicity and fecundity

Contact toxicity tests were performed to confirm the effectiveness of the different extracts. During this test, both types of extractions were applied using CRD with five replications. In each petri dish, twenty-five field-collected aphids (2nd and 3rd instar) were separated using cabbage leaves that were soaked with each extraction solution (10 ml/100 ml water). Once they had settled, botanical extracts were applied using a fine hand sprayer (Flip & Spray^{TM} Bottles; Thomas Scientific, USA), using two spray applications to completely cover the Petri dish area. Excess solution was removed using a tissue paper. A lid with four holes drilled was used to cover the petri dishes while allowing for ventilation. Lastly, the petri dishes were incubated at 25 ± 2°C, 50% RH, and a photoperiod of 10:14 h L: D. The petri dishes were examined consecutively at 6, 12, 24, and 48 h starting from extract application.

To determine the fecundity of *Myzus persicae*, the extracts were subjected to the same process described above. In each petri dish (9-cm diameter) ten adult aphids were placed on cabbage leaves after 24 h. Surviving adults were transferred into new petri dishes with fresh food to assess the fecundity of the mature aphids. Data on fecundity were recorded from each treatment on a daily basis for five days.

Greenhouse study

The glasshouse experiment was carried out to assess the effectiveness of several essential oils against green peach aphid on cucumber cultivated in net houses. Cucumber seedlings were cultivated in germination trays and transplanted during the last week of April 2019, keeping a row to row and plant to plant spacing of 60 cm x 30 cm. The seedlings were placed in raised beds 30 cm by following recommended cultural and management practices. The cucumber was cultivated using a randomized complete block design (RCBD) with six treatments and two methods (EOE, ETB) for each treatment performed in triplicate. Each sub-plot size measured as 10 × 10 m. The treatments were as follows: T1 (*Citrullus colocynthis*), T2 (*Trachyspermum ammi*), T3 (*Nicotiana tobaccum* L), T4 (*Azadirachta indica*), T5 (*Withania cagulans*), and T6 (control). The extracts were sprayed using a Knapsack sprayer. After each treatment, the sprayer was rinsed twice to prevent contamination. The aphid population was counted on 10 randomly selected plants from each subplot. Aphid populations were randomly examined on 10 randomly selected plants from a sub-plot, and each selected plant was examined. Pre-treatment observations were recorded 24 h prior to application, and post-treatment aphid counting was conducted at 1-, 2-, 4-, and 7-day intervals. The environmental conditions inside greenhouses followed seasonal trends (April: 24 ± 0.4 °C, 40.2 ± 1.9% RH; May: 25.3 ± 0.4 °C, 45.6 ± 1.6% RH; June: 26.2 ± 0.4 °C, 48.5 ± 1.9% RH; July: 26.5 ± 0.5 °C, 50.8 ± 1.5% RH).

Data analysis

The materials and techniques were evaluated based on the mortality, reduction, and fecundity percentage and corrected using Abbott's formula (*Eq.1*) (Abbott, 1925).

$$Corrected\ \%\ mortality\ =\ \frac{(1-n\ in\ T\ after\ treatment)}{n\ in\ Co\ after\ treatment} \times\ 100 \qquad \text{(Eq.1)}$$

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where *n* is the insect population, *T* is "treated", and *Co* is the control. The mortality, reduction, and fecundity data were subjected to factorial analysis of variance (ANOVA) using Minitab 16.1 software. To analyze the influence of treatments on mortality, reduction, and fecundity of the treated green peach aphid *M. persicae*, the means were compared using Tukey's HSD test at a 5% level of significance.

Results

Effect of botanicals on the mortality rate of green peach aphid through fumigation toxicity

The effectiveness of fumigation of each treatment was determined by counting the mortality of *Myzus persicae* after treatment with each extract. Each botanical extract had adverse effects on the pest. Mortality was found to increase with the exposure period. Mortality data are presented in *Table 1*, with significant differences at 2 h (F=69.1; df=4; P < 0.000), 6 h (F=27.4; df=4; P < 0.001), 12 h (F=24.9; df=4; P < 0.000), and 24 h (F=2.7 ; df=4; P < 0.012). The highest mortality was reported after 24 h of exposure. The interval wise mortality caused by *Trachyspermum ammi* was 81.7% and 65.20% (EOE) and 75.45% and 57.92% (ETB= after 24 h and 12 h of exposure, respectively, followed by *Citrullus colocynthis* (76.5% and 60.0% (EOE) and 67.73% and 45.0% (ETB)), *Nicotiana tobaccum* L. (63.0% and 51.60% (EOE) and 60.0% and 36.25% (ETB)), *Azadirachta indica* (56.5% and 32.20% (EOE) and 49.09% and 31.67% (ETB)), and *Withania cagulans* (50.0% and 30.40% (EOE) and 46.36% and 28.33% (ETB)) after 24 h and 12 h of exposure. Extracts obtained using the EOE method resulted in a higher mortality than those obtained via the ETB method.

Table 1. *Fumigant toxicity effect of botanicals extracts against aphid M. persicae in laboratory*

Treatments	Method	Pre-treatment	Post-treatment population and mortality percentage (%)[a]			
			2 h	6 h	12 h	24 h
Citrullus colocynthis	**EOE**	30	21.5 (28.33) f	15.2 (43.70) fg	10 (60.0) cd	5.4 (76.5) c
	ETB	30	24.2 (19.33) d	18.7 (25.20) e	13.2 (45.0) c	7.1 (67.73) c
Trachyspermum ammi	**EOE**	30	18.4 (38.67) g	13.3 (50.74) g	8.7 (65.20) d	4.2 (81.7) cd
	ETB	30	23.4 (22.00) e	16.1 (35.60) f	10.1 (57.92) cd	5.4 (75.45) c
Nicotiana tobaccum L	**EOE**	30	20.1 (33.00) b	15.7 (41.85) f	12.1 (51.60) c	8.5 (63.0) c
	ETB	30	26.1 (13.00) c	20.3 (18.80) d	15.3 (36.25) bc	8.8 (60.00) c
Azadirachta indica	**EOE**	30	25.4 (15.33) cd	21.1 (21.85) d	16.2 (35.20) b	10 (56.5) b
	ETB	30	27.4 (8.67) b	22.3 (10.80) cd	16.4 (31.67) b	11.2 (49.09) b
Withania coagulans	**EOE**	30	26.2 (12.67) c	22.4 (17.04) cd	17.4 (30.40) b	11.5 (50.0) b
	ETB	30	28 (6.67) b	22.8 (8.80) c	17.2 (28.33) b	11.8 (46.36) b
Control	**EOE**	30	30 a	27 a	25 a	23 a
	ETB	30	30 a	25 b	24 a	22 a
Significance level	-		F=69.1, df 4 P < 0.000	F=27.4, df 4 P < 0.001	F=24.9, df 4 P < 0.000	F=2.7, df 4 P < 0.012

EOE, ethanol oil extraction from powder; ETB, extraction through boiling of powder. [(%)] Percentage reduction due to the post application of treatments at a particular time mentioned in parentheses. [a]Means followed by same letters withen colum are non-significantly different from each other, (LSD; P=0.05)

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Effect of botanicals on mortality rate of green peach aphid through fumigation toxicity contact toxicity

Table 2 shows the mortality (percentage) of *Myzus persicae* due to contact toxicity. The maximum mortality was recorded after 24 h. According to the results, the highest mortality was 71.19% and 85.71% (EOE) and 67.54% and 75% (ETB), caused by the crude extract of *Trachyspermum ammi* at 24 h and 48 h, respectively, followed by *Citrullus colocynthis* with 65.25% and 71.43% (EOE) and 48.25% and 58.33% (ETB) at 24 h and 48 h, respectively, *Nicotiana tobaccum* L. with 52.54% and 66.33% (EOE) and 45.61% and 54.76% (ETB) at 24 h and 48 h, and *Azadirachta indica* with 25.0% and 32.28% (EOE) and 18.0% and 26.85% (ETB) at 6 h and 12 h. The same trend of mortality was observed for *Withania cagulans* at 6 h to 48 h of exposure, with 19.0%, 25.71%, 38.14%, and 46.94% (EOE), and 12.0%, 21.30%, 28.95, and 32.14% (ETB), respectively. The EOE and ETB extracts were found to be effective compared to the control, and the highest mortality in the aphid population was observed up to 24 h after exposure. The extracts obtained through ETB did not perform as well as the extracts obtained through ETB.

Table 2. *Contact toxicity effect of botanicals extracts against aphid M. persicae on cabbage leaves in laboratory*

Treatments	Method	Pre-treatment	Post-treatment population and mortality percentage (%)			
			6 h	12 h	24 h	48 h
Citrullus colocynthis	EOE	25	15.25 (39.00) e	13.75 (47.62) c	10.25 (65.25) de	7 (71.43) de
	ETB	25	18.25 (27.00) d	16.5 (38.89) bc	14.75 (48.25) d	8.75 (58.33) d
Trachyspermum ammi	EOE	25	14.5 (41.7) e	12.3 (54.29) c	8.5 (71.19) e	3.5 (85.71) g
	ETB	25	16.75(33.00)de	13.75 (49.07) c	9.25 (67.54) e	5.25 (75.00) f
Nicotiana tobaccum L	EOE	25	19.5 (22.00) d	16.25(38.10)bc	14 (52.54) d	8.25 (66.33) d
	ETB	25	20.75 (18.00) c	18.75(30.56)bc	15.5 (45.61) cd	9.5 (54.76) d
Azadirachta indica	EOE	25	18.75 (25.00) d	17.75(32.28)bc	16.5 (44.07) c	11.5 (53.06) d
	ETB	25	20.5 (18.00) c	19.75 (26.85) b	18.75 (34.21) bc	11.75 (44.05) d
Withania coagulans	EOE	25	20.25 (19.00) c	19.5 (25.71) b	18.25 (38.14) bc	13 (46.94) c
	ETB	25	22 (12.00) b	21.25 (21.30) b	20.25 (28.95) b	14.25 (32.14) c
Control	EOE	25	25 a	26.25 a	29.5 a	24.5 a
	ETB	25	25 a	27 a	28.5 a	21 b
Significance level	-	-	F=4.49, df 4 P < 0.010	F=2.44, df 4 P < 0.084	F=33.05, df 4 P < 0.000	F=6.14, df 4 P < 0.002

EOE, ethanol oil extraction from powder; *ETB,* extraction through boiling of powder. [(%)] Percentage reduction due to the post application of treatments at a particular time mentioned in parentheses. [a]Means followed by same letters withen colum are non-significantly different from each other, (LSD; P=0.05)

Effect of botanicals on fecundity rate of the green peach aphid

The impact of botanical extracts on fecundity revealed that *Withania coagulans* was the least effective. Aphid gave birth to a non-significant number of offspring (21.6 and 35 EOE and 22.3 and 32 ETB nymphs per female) in *W. coagulans*-treated petri dishes and the control (*Fig. 1*). During the same time period, green peach aphids treated with *Nicotiana tobaccum* L. and *Azadirachta indica* extract also gave birth to offspring (19.6 and 17.3 EOE and 18.6 and 18 ETB nymphs per female). The *Citrullus colocynthis* EOE and ETB extract had the lowest fecundity, with females producing only 9.3 and 13.3

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nymphs in 5 days. Similarly, during the same incubation period, adult aphids were treated with *Trachyspermum ammi* EOE, and the ETB botanicals were ranked the second lowest. In terms of fecundity, these females produced 12.6 and 16.6 offspring, respectively. Significant differences in fecundity were observed between all botanical extracts (F=2.10; df=4; P < 0.123). The results revealed that *C. colocynthis* performed well in reducing the fecundity of *M. persicae*.

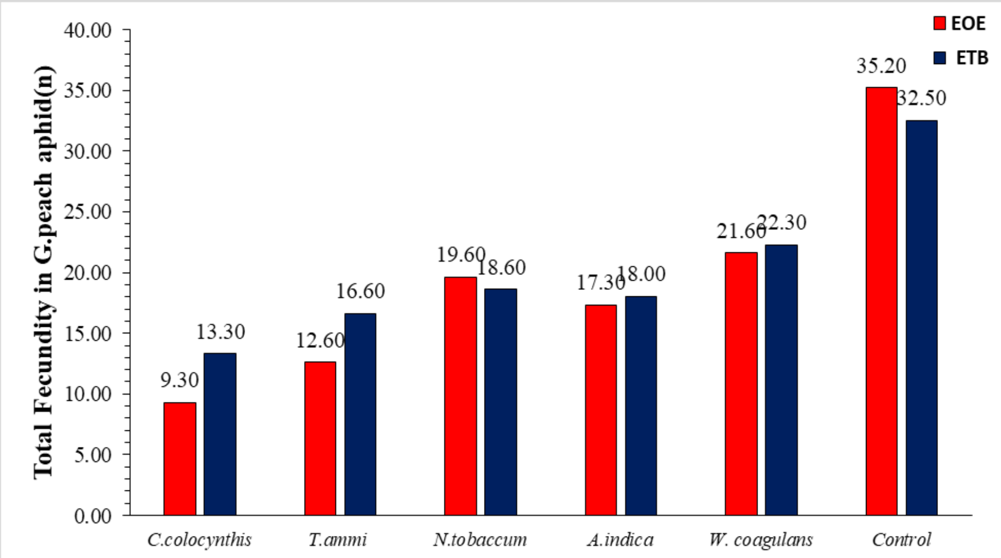

Figure 1. *Total fecundity (nymphs produced) in green peach aphids treated with EOE and ETB botanical extracts*

Population reduction of M. persicae in greenhouse

All treatments were effective under greenhouse conditions. The aphid populations were clearly affected by both extraction methods. The efficacy at 24 h post-treatment (*Table 3*) revealed that *Citrullus colocynthis* showed a maximum reduction in the *Myzus persicae* population of 50.48% (EOE) and 41.05% (ETB), followed by *Trachyspermum ammi* with 41.23% (EOE) and 31.20%, (ETB), Tobacco Desi with 41.23% (EOE) and 31.20% (ETB), neem seed with 35.05% (EOE) and 22.53% (ETB), and *Withania coagulans* with 24.23% (EOE) and 15.49% (ETB). *Citrullus colocynthis* performed well at 3-day intervals after treatment and resulted in a maximum of 70.9% (EOE) and 63.30% (ETB) in the aphid population, followed by *Trachyspermum ammi* with an efficacy of 62.30% (EOE) and 49.48% (ETB), *Nicotiana tobaccum* L. with 63.30% (EOE) and 50.79% (ETB), neem seed with 48.4% (EOE) and 40.29% (ETB), and *Withania coagulans* with 45.56% (EOE) and 34.93% (ETB). After seven days of treatment, *Citrullus colocynthis* was consistently the most effective in reducing the populations of aphids, with a reduction of 79.42% (EOE) and 68.50% (ETB), followed by *Trachyspermum ammi* with 73.91% (EOE) and 60.19% (ETB), Tobacco Desi with 67.38% (EOE) and 57.9% (ETB), *Azadirachta indica* with 53.77% (EOE) and 46.89% (ETB), and *Withania coagulans* with 50.0% (EOE) and 42.78% (ETB). According to the average output of all biopesticides (Fig. 1), the *Citrullus colocynthis* extract was the most successful against aphids, followed by *Trachyspermum ammi*, *Nicotiana tobaccum* L., *Azadirachta indica*, and *Withania coagulans*.

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Table 3. *Effect of foliar application of botanicals extracts against aphid M. persicae on cucumber plants in greenhouse*

Treatments	Method	Pre-treatment	Post-treatment population and reduction percentage (%)			
			24 h	**48 h**	**96 h**	**168 h**
Citrullus colocynthis	**EOE**	60	38.3 (50.48) fg	32.2 (61.41) g	25.7 (70.99) f	19.5 (79.42) h
	ETB	73.43	45.6 (41.05) e	39.34 (52.86) ef	32.52 (63.30) e	29.85 (68.50) f
Trachyspermum ammi	**EOE**	52.7	45.46 (41.23) e	41.35 (50.45) e	33.4 (62.30) e	24.72 (73.91)g
	ETB	68.9	53.22 (31.20) cd	48.53 (41.85) d	44.76 (49.48) d	37.72 (60.19) e
Nicotiana tobaccum L	**EOE**	66.4	43.7 (43.50) f	37.83 (54.67) f	32.52 (63.30) e	30.91 (67.38) f
	ETB	63.35	54.27 (29.84) c	46.9 (43.80) d	43.6 (50.79) d	39.8 (57.99) e
Azadirachta indica	**EOE**	70.6	50.24 (35.05) d	47.3 (43.32) d	45.65 (48.48) d	43.8 (53.77) d
	ETB	83.6	59.92 (22.53) b	54.1 (35.17) c	52.9 (40.29) c	50.32 (46.89)b
Withania coagulans	**EOE**	81.5	58.37 (24.54) bc	53.45 (35.95) c	48.23 (45.56) d	47.35 (50.03) c
	ETB	75.5	65.37 (15.49) b	61.72 (26.04) b	57.65 (34.93) b	54.22 (42.78)b
Control		63.2	77.35 a	83.45 a	88.6 a	94.75 a
Significance level			F=15.27, df 4 P < 0.000	F=3.94, df 4 P < 0.018	F=12.47, df 4 P < 0.003	F=29.66, df 4 P < 0.035

EOE, ethanol oil extraction from powder; *ETB*, extraction through boiling of powder. **(%)** Percentage reduction due to the post application of treatments at a particular time mentioned in parentheses. [a]Means followed by same letters withen colum are non-significantly different from each other, (LSD; P=0.05)

Discussion

Due to the problems associated with the use of synthetic chemicals for pest control, there is an urgent need to incorporate natural products, primarily those derived from plant sources, for use in pesticides against insects. This is especially true for *Myzus persicae*, which is increasingly causing damage in vegetable and fruit-growing regions around the world. Because of their strong efficacy against various insect pests, plant extracts and essential oils are widely used for pest control. The plants chosen in this study, namely *Citrullus colocynthis*, *Trachyspermum ammi*, *Nicotiana tobaccum* L., *Azadirachta indica*, and *Withania coagulans*, are to some degree natural insect repellents and promising resources to combat infestations by the green peach aphid. In this study, the phytochemical constituents of the extracts of the above-mentioned plants were identified, and their insecticidal activity was assessed using residual and contact toxicity methods. Essential oils or bio-pesticide extraction from plant parts is dependent on the plant material and solvent used. In our results, ethanol was used to obtain a high polarity and extract yields during solvent extraction, as previously reported (Awang et al., 2017). Ethanol was found to yield more from *Melastoma malabathricum* leaves compared to other solvents. Furthermore, using ethanol as an extraction solvent, a relatively high extract yield from *C. colocynthis* and *C. sativa* has been previously reported (Maqsood et al., 2020).

The fumigant activity of plant volatiles can be exploited as a control of the phytophagous insects of greenhouse crops. However, due to the volatile and unstable nature of these compounds, this idea has yet to gain commercial ground. The essential oils of anise and cumin have been reported to be toxic against cotton aphids (*Aphis gossypii* Glover) (Hemiptera: Aphididae) (Isman, 2000). In the present study, the mortality of *Myzus persicae* was determined using EOE and ETB from different plants under laboratory conditions by fumigation toxicity. All of the botanical extracts were

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found to be effective and resulted in mortality at 2 h against aphids and increased with the exposure period. The present study is in line with previous studies. In a laboratory, the bioassay inside small air-tight dishes, vapors of anise essential oil (EO), or its major compound (E)-anethole, were effective against the bird cherry-oat aphid *Rhopalosiphum padi* L. (Pascual et al., 2017). In the red flour beetle adults, *Tribolium castaneum*, (E)-anethole in combination with 1,8-cineole provided the best results in terms of fumigant toxicity (Koul et al., 2007). Various concentrations and exposure times of ajwain (*Trachyspermum ammi*) oil have been found to have fumigant activity against adults of *Oryzaephilus surinamensis*, *Rhyzopertha dominica*, and *Tribolium confusum* (Habashi et al., 2011).

In the present study, the results of contact toxicity under laboratory conditions further revealed that treatment with *Trachyspermum ammi* had the highest mortality and was the most effective against aphids, followed by *C. colocynthis*, *Nicotiana tobaccum* L., *Azadirachta indica*, and *Withania coagulans*. Additionally, extracts obtained by extraction via the ETB of the leaves and seeds of various plants were found to perform less well than extracts obtained by the EOE method. Similarly, Maqsood et al. (2020) found that, after 48 h of exposure, treatment with *Brevicoryne brassicae* resulted in the highest mortality rate (83.33 ± 1.29%), followed by *Citrullus colocynthis* and *C. indica*, with mortality rates of 81.67 ± 3.41% and 81.67% ± 1.67%, respectively. With regards to the essential oil of *Myristica fragrans* extracted using ethanol, the maximum concentration (10 mg/mL) was found to result in a high mortality in the nymphs of whiteflies (72.50 ± 4.23%), while the lowest concentration resulted in the lowest nymph mortality (Wagan et al., 2017). The Trifolio company's neem oil, which contained 0.04% azadirachtin and an equivalent amount of 3-tigloyl-azadirachtol, caused approximately 90% mortality of *Schistocerca gregaria* within 2 days (Zia, 2009). The ethanol-essential oil of *Gardenia jasminoides* and its components were tested in a contact toxicity procedure against the nymphal stage of the whitefly (Wagan et al., 2018). The action of aqueous extracts of tobacco, tagetes, and tephrosia of *Callosobruchus spp.* were also evaluated against insect pests (Kawuki et al., 2005). Similarly, in laboratory conditions, Vekaria and Patel (2000) tested tobacco leaf extracts against aphids.

Fecundity is an important aspect of insect populations, which can be influenced by plant extracts, resulting in reduced fecundity. In the present study, differences in fecundity were observed after *Myzus persicae* was exposed to a range of extracts. Aphids treated with the extracts were found to reproduce at a slower rate than untreated aphids; treatment with *Citrullus colocynthis* extract resulted in the lowest fecundity, while treatment with *Trachyspermum ammi* botanicals resulted in the second lowest fecundity during the same incubation period. In a previous study, the longevity and fecundity of cotton aphids were both found to be harmed by treatment with neem and eucalyptus extracts (Bayhan et al., 2006). Similarly, Pavela et al. (2004) observed the lowest fecundity in cabbage aphids after exposure to systemic neem extract. The combination of entomopathogenic fungi and botanical extracts was also found to have a negative impact on fecundity. In a similar study, when aphids were treated with a binary mixture of *Beauveria bassiana* and eucalyptus extract, the lowest fecundity (7 nymphs per female) was observed (Ali et al., 2017). In another study, aphids exposed to neem seed extracts and citrus aphids yielded a smaller number of nymphs. The negative effects of the fungal infections of *Beauveria bassiana* and *Metarhizium anisopliae* on the fecundity, fertility, and egg-laying of Russian wheat aphids have also been previously explored (Wang and Knudsen, 1993; Tang et al., 2002).

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Research has demonstrated that *Cassia sophera* and *Ageratum conyzoides* extracts induced mortality in *B. brassicae* comparable to the synthetic insecticide emamectin benzoate (Amoabeng et al., 2013). *Elettaria cardamomum,* another medicinal plant, was found to be effective against this pest (Jahan et al., 2016). According to reports, *B. brassicae* is sensitive to 1,8-cineole, a chemical compound found in *Laurus nobilis* caused a 52% reduction in reproduction (Kahan et al., 2008). In the present study, compared to the average output of all biopesticides, *Citrullus colocynthis* extract was the most successful against aphids, followed by *Trachyspermum ammi*, *Nicotiana tobaccum* L., *Azadirachta indica*, and *Withania coagulans*. In line with the present study, leaf extracts were found to exert the greatest pesticide action against *B. brassicae*; however, extracts from the leaves and fruits of *C. colocynthis* have also been found to be repellant against *Aphis craccivora* (Farghaly et al., 2009). Cucurbitacin E glycosides isolated from *C. colocynthis* extract have previously shown effective insecticidal activity against *Aphis craccivora* (Farghaly et al., 2009). Moreover, *Nicotiana tabacum* L. was found to be highly effective against insect pests of cabbage, wheat, peas, grammes, and other crops (Lal and Verma, 2006). The efficacy and control level of tobacco (variety IT86D-719) against insect pests of cowpea and their effects on yield were determined by Ahmed et al. (2009). These extracts have potential for use in novel eco-friendly sprays that are commercially available for use by small farmers. In fact, some have already been introduced by farmers in several regions, adding to the urgency of their evaluation. The use of natural bio-insecticides has been shown to be both efficient and environmentally beneficial, resulting in a reduction of the pesticide load on biodiversity. Furthermore, essential oils extracted from natural plants are widely believed to be healthy for humans and the environment, making them a potential source of new botanical insecticides.

Conclusion

In conclusion, the results presented in this study indicate that the EOE method of extraction was more effective against aphids than the ETB method. The highest activity in the fumigant toxicity, contact toxicity, fecundity, and greenhouse experiments were associated with extracts obtained using this method for the maximum duration. However, further studies are needed to fractionate and isolate the active compounds responsible for aphid mortality, as well as to examine their risks and compatibility, to improve aphid eco-friendly management.

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