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Effect of different plant extracts and nanoparticles on *Thrips tabaci* (Lind.) (Thysanoptera: Thripidae) under field conditions and their allelopathic potential on the onion, *Allium cepa* L. using bioassays and RAPD analysis



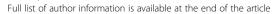
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Abstract

The present study was conducted to investigate the toxicity of Aerosil 200® (fumed silica nanoparticles) and leaf extracts of four plants, Cinnamomum camphora, Matricaria chamomilla, Mentha arvensis, and Trigonella foenumgraecum against Thrips tabaci (Lind.) (Thysanoptera: Thripidae) in onion fields, as well their allelopathic effects on onions; moreover, the chlorophyll, phenol, and protein contents were determined in onions. This study was performed in completely randomized plots. After a growth period of 1 month, bioassay investigations and molecular polymorphism in *T. tabaci* by RAPD-PCR were performed, and total chlorophyll, phenol, and protein concentrations were investigated in onion plants posttreatment as well. The initial reduction% of the T. tabaci population in onion fields after application of a high concentration of nanoparticles (Aerosil 200° (4 ml/l)) and 8000 ppm concentrations of the four plant extracts were 83.66, 81.08, 86.92, 74.49, and 91.38%, respectively, whereas their persistence effects were 73.18, 67.78, 71.46, 66.94, and 78.29%, respectively. Furthermore, the total chlorophyll contents in onions treated with the nanoparticles and four plant extracts were 1.35, 1.17, 1.09, 1.07, and 1.18 mg/g, respectively; additionally, the concentrations of phenols were 4.65, 3.15, 3.15, 2.85, and 3.70 mg/g in onions treated with C. camphora, M. chamomilla, M. arvensis, T. foenum-graecum, and Aerosil 200°, respectively. The C. camphora extract was the most potent, as it increased the protein content in the onion plants, while the Aerosil 200° decreased the protein content in onions. In addition, DNA-RAPD showed that the polymorphism percentages were 73, 71, and 67% when treated with high concentrations of *C. camphora* and *M. arvensis* extracts and Aerosol 200°, respectively. T. foenum-graecum and M. chamomilla extracts induced the least polymorphism (17 and 16%, respectively). Overall, this study indicated that these plant extracts as well as the nanoparticles in Aerosil 200° could be used to reduce onion infestations of *T. tabaci* in the field environment.

Keywords: Thrips tabaci, Onion, RAPD-PCR, Nanoparticles, Plant extracts, Allelopathy, Aerosil 200®

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Background

The onion (*Allium cepa* L.) is an important vegetable crop cultivated worldwide. Egypt is considered the largest dehydrated onion source in Africa and the Middle East. Iris yellow spot virus (IYSV) transmission via *Thrips tabaci* (Lind.) (Thysanoptera: Thripidae) has been reported in many countries, including Egypt (Abdelkhalek et al. 2019). Furthermore, onion plants are considered one of the main origins of dietary polyphenols worldwide (Knekt et al. 1996), and their consumption leads to the protection of DNA against oxidation. Polyphenols in dietary plants showed very important antioxidant activities (Singh et al. 2009). Additionally, they protect organisms from/against cellular damage (Martínez et al. 2007).

Devastating losses of annual onion crops occur due to disease transmission and can range from nonsignificant amounts of loss up to more than 50% of the total crop, depending upon many factors. Iris yellow spot virus is the most important disease transmitted by *T. tabaci* (Abdelkhalek et al. 2019). Disease symptoms of Iris yellow spot virus are similar to those of *T. tabaci* infestation and appear as eye-like, chlorotic, or diamond-shaped lesions (Pappu et al. 2008). *T. tabaci* is among the major insect pests of onion crops and cause large amounts of damage to the crop by consuming the plant sap and/or by transmitting other diseases, including IYSV (Gent et al. 2006). Control strategies of *T. tabaci* include chemical insecticide treatments (Jensen 2005).

Heavy application and indiscriminate uses of chemical pesticides in insect pest management has led to the advancement of resistance in all classes of insecticides (Kranthi et al. 2002). On the other hand, using natural plant oils are encouraged in order to decrease the application of chemical pesticides and their residues (Murray et al. 2011).

Allelopathy (largely of secondary metabolites) refers to the harmful or beneficial effects of a plant on another plant through biochemical release from the plant parts, and the release of such chemicals may attract or repel insects. Additionally, allelopathy can affect many plant bionomics, including growth, plant succession, and plant productivity (Singh et al. 2001). Allelochemicals are the secondary metabolites of allelopathic organisms (Hanan M. 2016). Recently, plant extracts have been used for pest control as alternatives to chemical pesticides (Qari and Abdel-Fattah 2017). Several molecular techniques have been used to determine the ability of these plant extracts to reduce insect infestation. Random amplification polymorphism DNA PCR (RAPD-PCR) could be a good technique for the rapid identification of genetic changes on the molecular level (Qari 2010). RAPD-PCR has also been used to determine variations in the *T. tabaci* genome (Ramakrishnan et al. 2019). Moreover, plants offer a different and alternative source of insect biocontrol agents, as they contain a wide range of bioactive materials, and many of such compounds are selective or have no harmful effects on nontargeted organisms as well as the environment, unlike synthetic insecticides. Additionally, some of these bioactive materials have been shown to have repellent, antifeedant, and toxic effects and induce enzymatic changes in insects (Adakole and Adevemi 2012).

This study was carried out to evaluate the effect of plant extracts and Aerosil 200° nanoparticles in reducing onion infestations of *T. tabaci* in the field using bioassays and molecular tools.

Materials and methods

Plant extracts and tested material preparation

The effects of the leaf extracts of four plants, namely, camphora, Matricaria chamomilla, Cinnamomum Mentha arvensis, and Trigonella foenum-graecum, on the infestation of *T. tabaci* in an onion field, as well as their allelopathic effects on the onion plants, were studied. According to Qari (2008), powdered leaves of each plant were soaked in liquid nitrogen and 1000 ml distilled water for 72 h at room temperature for aqueous extraction and then kept in a dark at 4 °C. The solutions were filtered and evaporated via rotary evaporation until the extracts were jelly-like in appearance and reach a final concentration of 1 g/l (1000 ppm). Two different concentrations were calculated and prepared (4000 and 8000 ppm) to be used in the bioassay.

On the other hand, nanoparticles (ca. 6.6–13.3 nm: Aerosil 200° as a silica oxide nanopowder, 99.8% SiO₂) was obtained from a commercial company in Egypt for scientific services (Tiba Company). The shape and size of the nanoparticles were verified by electron microscopy and prepared at two concentrations, 4 ml and 2 ml/l in order to be use in the bioassays.

Experimental design

Studies which evaluate the effects of different plant extracts, as well as Aerosil 200° nanoparticles, on T. tabaci in onion cultivation fields and their allelopathic effects on the onions were carried out at Kaha Agricultural Research Station Farm, Egypt, in March 2018. The onion cultivar Giza 20 was obtained from the Agricultural Research Center, Giza, Egypt. A backpack sprayer was used as for foliar spray. Onions were grown in completely randomized plots in the field; each treatment was repeated four times, and each replicate area was 4 m × 4 m. After 2 months of onion cultivation, the bioassay investigations were performed; concentrations of 4000 ppm (8 ml/2 l/64 m for each treatment) and 8000 ppm (16 ml/2 l/64 m for each treatment) were applied for the plant extracts, while the implemented concentrations of Aerosil 200° were 2 ml/l (4 ml/2 l/64 m) and 4 ml/l (8

ml/2 l/64 m). All tested materials were prepared and applied as foliar sprays on onions in the morning. Pretreatment counting in each plot was carried out, then the application performed and the reduction% of *T. tabaci* adults were counted after 1, 3, 7, and 10 days and corrected according to Henderson and Tilton (1955).

Onion parameter analyses

All biochemical studies were carried out on new onion leaves after applications of both concentrations of the botanical extracts (4000 ppm and 8000 ppm) and Aerosil 200° (2 and 4 ml/l).

Total chlorophyll

The assessment of total chlorophyll content was estimated in 85% acetone leaf extracts according to Metzner et al. (1965). First, the leaves were centrifuged at 14000 rpm, for 20 min then the absorbance was read spectrophotometrically at 663, 652, 646, and 470 nm. Finally, the concentrations of chlorophyll were calculated according to the following equation (Metzner et al. 1965): total chlorophyll (μ g/ml) = A652 × 27.8.

Determination of phenols

One gram of onion leaves was ground in liquid nitrogen, extracted with 80% methanol (v/v) and heated at 85 °C for 2 h. After cooling, phenol determination in the extracted leaves was estimated according to Denre (2014).

Protein content

According to Qari (2016), the protein content of each treated and untreated onion sample was estimated.

RAPD-PCR protocol

Sampling of T. tabaci and DNA extraction

Adult insects of T. tabaci were collected from different treated onions by shaking the treated plants onto a white sheet to gather the insects. The samples of T. tabaci were crushed and stored in TE buffer at -20 °C until DNA extraction. DNA was extracted according to the procedure described Oliveira et al. (2010). The insects were ground in liquid nitrogen, dissolved in 4% CTAB extraction buffer, mixed with 4 μL β-mercaptoethanol, and then heated for 30 min at 65 °C. After centrifugation at 14000 rpm for 5 min, the supernatant was treated with cold isopropanol (v/v) for 20-30 min then centrifuged again at 14000 rpm for 5 min. DNA samples were precipitated, and the resulting precipitate was washed twice with 70% alcohol and resuspended in 0.1 ml TE buffer. The quality of the isolated DNA was quantified with a fluorometer and 0.8% agarose gel stained with 0.2 μg/ml ethidium bromide (EB) visualized under UV light.

RAPD-PCR analysis

The RAPD-PCR procedure was carried out on a final volume of 25 µl reaction mixture containing 20 ng genomic DNA, 1.5 µl PCR buffer (50 mM KCl, 12 mM MgCl₂, 100 mM Tris-Cl, pH 9.0), 7.92 µl sterile deionized water, 1.8 μl dNTPs (0.1 mM each), 1.8 μl decamer primers (Operon Tech) as shown in Table 1 (4 mM), and 1.5 U Tag DNA polymerase. PCR was conducted in a Perkin-Elmer TC480 thermocycler as follows: one cycle at 94 °C for 2 min; 45 cycles at 94 °C for 10 s, 36 °C for 30 s, 72 °C for 1 min; and a final extension at 72 °C for 2 min. The PCR products were separated by electrophoresis in 1% agarose gel, stained with EB, and visuunder UV and documented using Gel Documentation System software (Bio Rad, USA). Each primer yielding productive bands was repeated twice. Band sizes were specified by a DNA ladder.

Statistical analysis

Analysis of variance ANOVA was implemented with SPSS software. The percentage of DNA stability was estimated according to the following equation: $100 - (100 \text{ a} \div \text{n})$, according to Luceri et al. (2000), where n is the number of total bands in the control and a is the RAPD-PCR polymorphic bands detected in each treatment sample. The average for each treatment was used for evaluation. The reduction% was calculated according to Henderson and Tilton (1955) with the following equation:

$$Reduction\% = \left(1 - \frac{\text{No.in control before treatment x no.in treatment after treatment}}{\text{No.in control after treatment x no.in treatment before treatment}}\right)$$

Results and discussion

Average number of T. tabaci after treatment

The results presented in Table 2 showed that the average number of T. tabaci adults ranged from 46.9 ± 4.4 to 28.55 ± 5.9 in the untreated control, while it ranged from 2.95 ± 0.9 to 9.55 ± 1.7 , 8.45 ± 3.1 to 12.9 ± 4.2 , 4.75 ± 1.9 to 11.95 ± 4.4 , and 6.45 ± 3.7 to 12.65 ± 3.9 when 8000 ppm C. camphora, M. chamomilla, M. arvensis, and T.

Table 1 List of RAPD-PCR primer sequences

No	Primer name	Sequences 5' → 3'
1	OPA/08	GTGACGTAGG
2	OPA/11	CAATCGCCGT
3	OPA/16	AGCCAGCGAA
4	OPA/19	CAAACGTCGG
5	OPAI/21	CACGCGAACC
6	OPAL/20	AGGAGTCGGA
7	OPAB/09	GGGCGACTAC
8	OPH/11	CTTCCGCAGT

Table 2 Average number and reduction% of *Thrips tabaci* after application of plant extracts and Aerosil 200° in an onion field compared to that of the control field

Compound	Aerosil 200	O.	Trigonella foenum- graecum		Mentha arvensis		Matricaria chamomilla	1	Cinnamomo camphora	Untreated control	
concentration	2ml/l	4ml/l	4000 ppm	8000 ppm	4000 ppm	8000 ppm	4000 ppm	8000ppm	4000 ppm	8000 ppm	
Pre-treatment	41.65±5.2		41.05±4.8		43.75±3.6		39.9±3.5		41.25±4.2		46.9±4.4
After 24 hrs	13.95±3.4	5.65±1.8	15.5±3.6	6.45±3.7	14.75±3.0	4.75±1.9	17.35±3.6	8.45±3.1	11.1±2.4	2.95±0.9	38.95±4.1
Reduction %	59.67	83.66	54.53	81.08	59.40	86.92	47.64	74.49	67.59	91.38	-
After 3 days	8.2±1.8	3.65±1.4	10.95±3.9	3.9±2.1	10.9±4.5	3.45±2.3	14.1±3.5	5.35±3.1	5.7±2.3	2±1.1	34.95±7.7
Reduction %	73.58	88.24	64.20	87.25	66.56	89.41	52.75	82.00	81.45	93.49	-
After 7 days	11.3±3.1	6.35±1.9	12.2±2.9	8.85±3.9	12.5±5.3	8.55±1.6	14.65±3.9	7.25±3.3	11.9±4.6	5.5±1.8	30.4±6.7
Reduction %	58.14	76.478	54.14	66.73	55.92	69.85	43.35	71.96	55.49	79.42	-
After 10 days	17.1±2.4	11.45±2.6	18.65±3.4	12.65±3.9	17.25±5.2	11.95±4.4	22.45±5.0	12.9±4.2	13.5±3.2	9.55±1.7	28.55±5.9
Reduction %	32.55	54.83	25.36	49.37	35.22	55.12	28.15	46.88	46.32	61.96	-

Average number ± SD (standard deviation)

foenum-graecum extracts were used, respectively. Moreover, the average number of T. tabaci ranged from 5.65 ± 1.8 to 11.45 ± 2.6 over a period of 1-10 days after application of 4 ml of Aerosil 200. Furthermore, the average number of T. tabaci adults ranged from 11.1 ± 2.4 to 13.5 ± 3.2 , 17.35 ± 3.6 to 22.45 ± 5.0 , 14.75 ± 3.0 to 17.25 ± 5.2 , 15.5 ± 3.6 to 18.65 ± 3.4 , and 13.95 ± 3.4 to 17.1 ± 2.4 when 4000 ppm C. camphora, M. chamomilla, M. arvensis, and T. foenum-graecum leaf extracts and Aerosil 200 were applied, respectively. Overall, it was found that the efficacy of the different plant extracts and Aerosil 200 varied due to the nature of the extract and the concentration used.

Reduction percentages of *T. tabaci* after application of botanical extracts and Aerosil 200°

The data in Table 2 show that the reduction% of *T. tabaci* individuals 10 days after application was 61.96–46.32, 46.88–28.15, 55.12–35.22, 49.37–25.36, and 54.83–32.55% when high and low concentrations of *C. camphora, M. chamomilla, M. arvensis*, and *T. foenum-graecum* leaf extracts and Aerosil 200 were applied, respectively, while the initial reduction% was 91.38–67.59, 74.49–47.64, 86.92–59.40, 81.08–54.53, and 83.66–59.67% after 1 day when high and low concentrations of

the extracts and Aerosil 200° were used, respectively. The most efficient essential oils were those obtained from *Mentha pulegium*, with estimated LC_{50} and LC_{90} values of 3.1 and 3.8 mg l^{-1} air, while those of *Thymus mastichina* were 3.6 and 4.6 mg l^{-1} air, respectively (Stepanycheva et al. 2019).

Comparison based on initial and persistence reduction, potency level, and control index of Aerosil 200° and four phytochemical extracts on *T. tabaci* in onion fields

The data shown in Table 3 indicate that the initial reduction% (after 24 h) of *T. tabaci* in onion fields after application of the highest concentration of Aerosil 200 and *T. foenum-graecum, M. arvensis, M. chamomilla*, and *C. camphora* was 83.66, 81.08, 86.92, 74.49, and 91.38%, respectively, while their persistence was 73.18, 67.78, 71.46, 66.94, and 78.29%, respectively.

Regarding the relative potency level of the initial reduction, the fold changes are shown in Table 4, it was found that the potency levels of *C. camphora*, *M. arvensis*, Aerosil 200°, and *T. foenum-graecum* were respectively 1.22, 1.16, 1.12, and 1.08 times greater than that of the *M. chamomilla* extract, while the potency levels based on the persistence of the different treatments descended as follows:

Table 3 Studies on the initial (Init.) and persistence (Pers.) reduction values of the high concentrations, the potency level and the susceptibility index of Aerosil 200° and four plant extracts applied to *Thrips tabaci* in an onion field

Compound	Initial and	d Pers. reduction	Potency	y level		Suscept	Susceptibility index			
	Initial	Persistence*	Initial	Persistence	General mean	Initial	Persistence	General mean		
Aerosil 200®	83.66	73.18	1.12	1.09	1.10	91	93	92		
Trigonella foenum-graecum	81.08	67.78	1.08	1.01	1.03	88	81	87		
Mentha arvensis	86.92	71.46	1.16	1.06	1.09	95	91	92		
Matricaria chamomilla	74.49	66.94	1.00	1.00	1	81	81	84		
Cinnamomum camphora	91.38	78.29	1.22	1.16	1.18	100	100	100		

^{*}Persistence: average number of Thrips tabaci in treatments within interval time 3, 7, and 10 days

Table 4 Effect of Cinnamomum camphora, Matricaria chamomilla, Mentha arvensis, and Trigonella foenum-graecum leaf extracts and Aerosil 200° on the total chlorophyll, phenol, and protein concentrations (mg/g) in onion plants after 10 days of spray application

Compound	Chlorophylls (mg/g)	Phenol (mg/g)	Protein (mg/g)
Control	1.08 ^c	2.15 ^d	50.88 ^d
C. camphora 4000 ppm	1.20 ^b	4.65 ^b	67.50 ^b
C. camphora 8000 ppm	1.35 ^a	6.75 ^a	77.40 ^a
M. chamomilla 4000 ppm	1.09 ^c	3.15 ^c	58.65 ^c
M. chamomilla 8000 ppm	1.17 ^b	4.78 ^b	64.40 ^b
M. arvensis 4000 ppm	0.93 ^e	3.15 ^c	54.45 ^c
M. arvensis 8000 ppm	1.09 ^c	4.21 ^b	60.35 ^b
T. foenum-graecum 4000 ppm	1.01 ^d	2.85 ^d	57.75 ^c
T. foenum-graecum 8000 ppm	1.07 ^c	4.10 ^b	63.15 ^b
Aerosil 200® 2 ml/l	1.11 ^c	3.70 ^c	47.47 ^e
Aerosil 200® 4 ml/l	1.18 ^b	5.80 ^a	35.25 ^e
LSD (Least significant difference)	0.6	1.25	4.25

^aHighly significant at P < 0.01

C. camphora, Aerosil 200°, M. arvensis, T. foenum-graecum, and M. chamomilla.

According to Sun (1950), the susceptibility index can describe and differentiate between treatments with a given arbitrary index value of 100 units for the most effective treatment. In this study, the most effective treatment was that with the *C. camphora* extract, so it considered the standard toxicant and given arbitrary index value of 100 units, and the reduction effect of tested compounds descended in the following order: *C. camphora*, *M. arvensis*, Aerosil 200°, *T. foenum-graecum*, and *M. chamomilla*. These results are in accordance with those of Shehawy et al. (2013), who concluded that plant extracts showed high reduction efficacy against *T. tabaci*.

Effect of plant extracts on the chlorophyll, phenol, and total protein contents in onion plants Chlorophyll concentration

As shown in Table 4, there was a significant difference observed in the total chlorophyll concentration of onion leaves after 10 days of application for all treatments. The total chlorophyll concentration in the control was 1.08, while it was 1.20, 1.09, 1.09, 0.93, 1.01, and 1.11 mg/g dry weight, when the lowest concentrations of *C. camphora, M. chamomilla, M. arvensis, T. foenum-graecum,* and Aerosil 200° were applied, respectively, and that after application of the highest concentrations was 1.35, 1.17, 1.09, 1.07, and 1.18 mg/g dry weight. Overall, it can be concluded that there was a direct relation between the application of *C. camphora* and *M. chamomilla* with the total chlorophyll concentration in the onions, i.e., the greatest applied extract concentration was the greatest total chlorophyll concentration compared to that of the

control. However, no significant differences in the chlorophyll content were observed when the lowest concentrations of *M. chamomilla, M. arvensis*, and Aerosil 200° were used compared to that of the control. Leaf senescence including chlorophyll loss (leaf yellowing) decreases the amount of photosynthesis occurring (Munne-Bosch 2007). Variations in the chlorophyll concentration of the plant may be due to the presence of some interactions of allelochemicals present in the plant extracts used in this study (Talukder et al. 2015).

Phenol concentration

The data in Table 4 show that the phenol concentration was 2.15, 6.75, 4.78, 4.21, and 4.10 mg/g dry weight, in onions after application of the lowest concentration of C. camphora, M. chamomilla, M. arvensis, T. foenumgraecum, and Aerosil 200°, respectively. And that after application of the highest concentrations was 4.65, 3.15, 3.15, 2.85, and 3.70 mg/g dry weight, respectively, after 10 days. It could be concluded that treatment with the highest concentrations of C. camphora and Aerosil 200° correlated with a positive allelopathic effect (leading to increased phenol accumulation) compared to that of the control. However, application of *M. arvensis* and Aerosil 200° exhibited a negative allelopathic effect and led to a low concentration of phenols in the onions. Phenols in plants are involved in diverse processes, such as UV protection, plant pigmentation, plant nitrogen fixation, and disease resistance in the plant. Additionally, phenols, including flavonoids, are considered antioxidants and are medically important Abreu and Munne'-Bosch S. (2009). A complex allelopathic effect may inhibit growth of other plant species. Allelopathy is plant competition; it involves biochemical interactions, and it can involve

^bSignificant at P < 0.5

 $^{^{}c,d,e}$ Non-significant at P < 0.5 (there is no significant difference between data which given the same letter in the same column)

Table 5 Bands generated and polymorphism percentages obtained from RAPD-PCR primers of *T. tabaci* after exposure to high concentrations of plant extracts and Aerosol 200° at 4 ml/l

Primer	No. bands generated						Polymorphic bands compared to control				Average of	Polymorphism %				Average of		
	C	М	F	Ca	Ch	N	М	F	Ca	Ch	N	Polymorphic bands	M	F	Ca	Ch	N	polymorphism within primer
OPA/08	6	5	6	6	4	6	4	1	5	1	5	3.2	80	16	83	25	83	57.4%
OPA/11	12	10	9	7	12	12	4	5	7	2	8	5.2	40	55	100	16	66	55.4%
OPA/16	8	7	7	10	7	8	6	1	8	1	7	4.6	85	14	80	10	87	55.2%
OPA/19	9	8	9	5	9	9	6	0	5	0	6	3.4	75	0	100	0	66	48.2%
OPAI/21	10	5	8	8	9	5	5	3	5	2	3	3.6	100	37	62	22	60	56.2%
OPAL/20	11	10	11	9	11	11	7	0	6	4	7	4.8	70	0	66	36	63	47.0%
OPAB/09	8	8	8	7	8	8	5	1	5	0	4	3.0	62	12	71	0	50	39.0%
OPH/11	14	13	14	14	12	13	8	3	10	3	8	6.4	61	21	71	25	61	47.8%
Average o	of pol	ymor	phism	า %									71	17	73	16	67	

C, control; N, Aerosol 200°; Ca, Cinnamomum camphora; Ch Matricaria chamomilla; M, Mentha arvensis; F, Trigonella foenum-graecum

both "inhibitory" and "stimulatory" responses from the same compound (Leicach et al. 2009).

Total protein

The data illustrated in Table 4 show that the concentration of total proteins, measured after application of high concentrations of *C. camphora*, *M. chamomilla*, *M. arvensis*, *T. foenum-graecum*, and Aerosil 200° on the onions was 77.4, 64.4, 60.35, 63.25, and 35.25 mg/g dry weight, respectively, whereas the total protein contents was 67.5, 58.65, 54.45, 57.75, and 47.47 mg/g dry weight, after application at low concentrations. The application of *C. camphora* at 8000 ppm increased the protein concentration the most after 10 days of application, followed by *M. chamomilla*, *M. arvensis*, *T. foenum-graecum*, and *M. arvensis*. Application of the nanoparticles in Aerosil 200° decreased the concentration of total protein, indicating that Aerosil

200° had a negative allelopathic effect on the onions. However, the four plant leaf extracts exhibited beneficial allelopathic effects on the onion plant protein content. Overall, it could be concluded that there was a strong inverse relation between insect infestation and the chemical constituents of the onion plants after the treatments.

Allelopathy can affect many occurrences of plant bionomics, including growth, plant succession, and plant productivity (Singh et al. 2001).

Molecular markers

All eight RAPD-PCR primers used in the present study gave sufficient PCR banding patterns as shown in Table 5 and Fig. 1. DNA analysis based on the molecular marker technique is an effective method for studying the genetic diversity of insect pests (Loxdale and Lushai 1998). The main advantage of PCR-based systems is the development

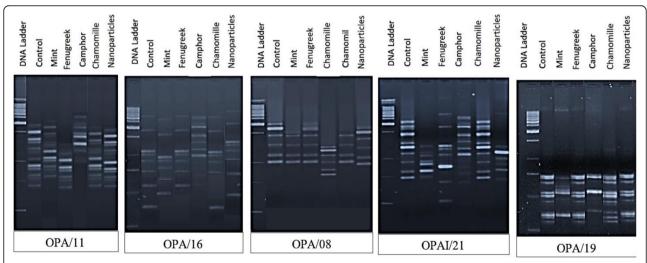


Fig. 1 Molecular polymorphism banding patterns of RAPD-PCR primers of *T. tabaci* after exposure to high concentrations of plant extracts and Aerosil 200° at 4 ml/l for 10 days (1 kb DNA ladder)

of single and multiloci fragments, and all eight RAPD primers used in this study produced scorable PCR products by amplifying the template DNA of *T. tabaci* with *Taq* polymerase.

The results illustrated in Table 5 indicate that primer OPA/08 gave 6, 5, 6, 6, 4, and 6 generated bands in the control, *M. arvensis, T. foenum-graecum, C. camphora, M. chamomilla*, and Aerosil 200° groups, respectively. Moreover, OPA/11 gave 12, 10, 9, 7, 12, and 12 generated bands in the control, *M. arvensis, T. foenum-graecum, C. camphora, M. chamomilla*, and Aerosil 200° groups, respectively, and OPA/16, OPA/19, OPAI/21, OPAL/20, OPAB/09, and OPH/11, gave generated bands in the control (8, 9, 10, 11, 8, and 14), *M. arvensis* (7, 8, 5, 10, 8, and 13), *T. foenum-graecum* (7, 9, 8, 11, 8, and 14), *C. camphora* (10,5, 8, 9, 7, and 14), *M. chamomilla* (7, 9, 9, 11, 8, and 12), and Aerosil 200° (8, 9, 5, 11, 8, and 13) groups.

Generally, the average number of polymorphic bands of the eight tested primers showed that the best harvested bands produced in RAPD-PCR were those of the primer OPH/11, which gave 6.4 bands, followed by those of OPA/11 and OPAL/20, which gave 5.2 and 4.8 bands, respectively. The lowest average number of polymorphic bands was produced by the primers OPAB/09, OPA/08, and OPA 19, which were 3, 3.2, and 3.4 bands, respectively (Table 5). Such a wide variation in the number of bands produced by these arbitrary primers may be attributed to the differences in the binding sites throughout the genome of *T. tabaci*.

Moreover, the polymorphism percentages in the present study indicated genetic changes in *T. tabaci* due to exposure to the different plant extracts as well as to the nanoparticles (Aerosil 200°). The results presented in Table 5 show that the polymorphism percentages were 73, 71, and 67% when the insects were treated with the highest concentration of the *C. camphora* and *M. arvensis* extracts and Aerosil 200°, respectively, whereas the *T. foenum-graecum* and *M. chamomilla* extracts induced the least polymorphism, yielding percentages of 17 and 16%, respectively, when compared to that of the control.

The RAPD marker system distinguished the different impacts of different plant extractions and Aerosil 200° as expected since they were affected on vitality of *T. tabaci*. This result was corroborated by the reduction percentage of onion infestation. This might be the host-induced genetic variation as observed in *Thrips* (Brunner et al. 2004).

Many studies have shown that there is a strong correlation between the genetic variation in the *Thrips* genome and changes in the DNA bands produced with RAPD (Jenser et al. 2001). The present study showed the ability of several plant extracts (*C. camphora*, *M. arvensis*(and nanoparticles to cause genetic changes in *Thrips*.

This result is consistent with that of many studies that suggested the ability of the *Thrips* genus to induce genetic modifications for pesticide resistance (Tsukahara et al. 2003), which may explain the global distribution of *Thrips*.

Conclusion

Overall, it can be concluded that using the aqueous leaf extracts, as well as the fumed silica nanoparticles had significant reduction effects on *T. tabaci* populations in an onion field. On the other hand, all of the plant extracts had positive allelopathic effects on onions by improving the chlorophyll, phenol, and protein contents in onion plants, but Aerosol 200° application was shown to decrease the total protein content.

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Authors' contributions

AS and SQ did the conceptualization. AS, SQ, NA, KA, and NA contributed in the formal analysis. AS, SQ, NA, KA, and NA took part in the investigation. AS and NA wrote the original draft. AS and SQ did the writing–review and approved the final manuscript. All authors read and approved the final manuscript.

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Competing interests

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