Annals of Agricultural Science (2013) **58**(1), 97–103

Faculty of Agriculture, Ain Shams University

Annals of Agricultural Science

www.elsevier.com/locate/aoas

ORIGINAL ARTICLE

Essential oils from some Egyptian aromatic plants as an antimicrobial agent and for prevention of potato virus Y transmission by aphids

Mona M. Oraby, Ali M. El-Borollosy *

Department of Agricultural Microbiology, Faculty of Agriculture, Ain Shams University, P.O. Box 68, Hadayek Shubra 11241, Cairo, Egypt

Received 1 January 2013; accepted 12 January 2013
Available online 27 February 2013

KEYWORDS

Aromatic plants;
Essential oil;
Linalool;
Antimicrobial;
PC/IC-RT-PCR;
PVY^{O};
Aphid transmission

Abstract Essential oils from different Egyptian aromatic plants (*Mentha piperita*, *Ocimum basilicum*, and *Thymus vulgaris*) were tested for their inhibitory effect on some selected harmful bacteria and yeast (*Escerichia coli*, *Psedumonas aeruginosa*, *Staphylococcus aureus*, *Streptococcus pyogenes*, and *Candida albicans*). Aerial parts from plants were directed to steam distillation for essential oil extraction. Oil yields were expressed in relation to dry weight of plant material, which found to be 0.39%, 0.20% and 0.55% w/w for *M. piperita*, *O. basilicum* and *T. vulgaris*, respectively. The types and percentage of essential oil constituents were determined using gas chromatography (GC). GC data revealed that the main compounds from *M. piperita* were menthol (35.44%) and menthone (20.11%), *O. basilicum* main component was linalool (45.11%), while *T. vulgaris* oil main component was thymol (75.76%). Preparations studied for their inhibitory effect were raw oils for sensitivity test and in the form of emulsions for spraying application. Emulsions were prepared depending on commercial liquid dish wash soap (Peril®) and Tween 80 as emulsifying agents. Results of sensitivity tests indicated that the most effective oil against bacteria and yeast was that of *O. basilicum* followed by that of *T. vulgaris*. *O. basilicum* oil was highly effective on *S. pyogenes* giving a zone of 19 mm more than that produced by Ampicillin, which was of 15 mm in diameter. Oil of *O. basilicum* was slightly more effective on *C. albicans* when compared with clotrimazole as an antifungal agent. Spraying potato tubers with *O. basilicum* oil emulsified with soap prior to brown rot bacteria (*Ralstonia solanacearum*) infection and preservation at 4 °C gave the best results followed by *T. vulgaris* oil with soap, as only 2 and 3 tubers out of 10 used shows rot symptoms, for

* Corresponding author.
E-mail address: ali.elborollosy@yahoo.com (A.M. El-Borollosy).

Peer review under responsibility of Faculty of Agriculture, Ain Shams University.

Production and hosting by Elsevier

 http://dx.doi.org/10.1016/j.aoas.2013.01.013

O. basilicum and *T. vulgaris* oils, respectively. After feeding *Myzus persicae* aphids on *potato virus y potyvirus* (common strain) (PVY^O) infected tobacco plants for 1 h, insects were print-captured individually and virus was successfully detected by immunocapture reverse transcriptase polymerase chain reaction PC/IC-RT-PCR, as the 801 bp coat protein gene (*cp*) bands were detected within agarose gel. Spraying tobacco plants with *O. basilicum* or *T. vulgaris* oils both emulsified with soap gave excellent results, as 8 and 6 plants out of 10 treated confirmed to be PVY^O-free by giving negative I-ELISA results, respectively. It was also observed that adding soap as an emulsifier has a killing effect on aphids.

Introduction

Plants produce a high diversity of secondary metabolites with a prominent function of protecting plants against predators and microbial pathogens due to their biocidal properties against different plant pathogens. Some metabolites are also involved in defense mechanisms against abiotic stress and are important in the interaction of plants with other harmful organisms (Schafer and Wink, 2009; Abdel-Reheem et al., 2012).

There are three major groups of secondary metabolites, including terpenes, phenylpropenoids, and N- and S-containing compounds (Wink, 1999). Among these secondary metabolites, it is estimated that over 3000 essential oils are known, of which about 300 are commercially important and used for their flavor and fragrance or as an antimicrobial agent and for many other medical purposes (van de Braak and Leijten, 1999).

Most of the antimicrobial activity in essential oils is found in the oxygenated terpenoids (alcohols and phenolic terpenes), while some hydrocarbons also exhibit antimicrobial effects (Burt, 2004). Interactions between these components may lead to antagonistic, additive, or synergistic effects. Some studies have demonstrated that whole essential oils usually have higher antibacterial activity than the mixtures of their major components, suggesting that the minor components are critical to the synergistic activity, though antagonistic and additive effects have also been observed (Mourey and Canillac, 2002; Bassolé and Juliani, 2012).

Phytoalexins were found to be produced from some plants as a response to microbial infection; these inducible defense substances comprise a large number of compounds with very different structures, that is, isoprenoids, flavonoids, and stilbenes (Bell and Charlwood, 1980). Isoprenoids meanly induced as a result of fungal or bacterial infection and act as antibiotics against a broad spectrum of these pathogenic microorganisms. Phytoalexins' (including isoprenoids) synthesis is induced by excreted proteins (elicitors) produced by the plant pathogen, that is, cell-degrading enzymes. These excreted elicitors bind to specific receptors on the outer surface of plasma membrane of the plant cells (Ames and Gold, 1989; Scheel and Parker, 1990). Binding of such elicitors results in the formation of signal substances as a part of signal transduction chain leading to induce the transcription of genes encode the enzymes of phytoalexin synthesis. Elicitors also cause the death of the infected cell in addition to its surrounding cells by what so called "cell death program" (Scheel and Parker, 1990).

Compounds of the essential oil of *Croton cajucara* were separated by thin layer chromatography and exposed to *Candida albicans*, *Lactobacillus casei*, *Staphylococcus aureus*, *Streptococcus sobrinus*, and *Porphyromonas gingivalis* showed that the proliferation of bacterial cells was inhibited by still uncharacterized molecules, but monoterpenes were confirmed as the antifungal components of the essential oil. The effects of linalool on the cell biology of *C. albicans* were evaluated by electron microscopy, which showed that linalool induced a reduction in cell size and abnormal germination (Alviano et al., 2005).

Controlling stylet-borne plant viruses transmitted by aphids is one of the greatest challenges facing vegetable growers today. Attempts to control these diseases by killing winged aphids with potent chemical aphicides have proven futile and human health risky. Natural oils have an advantage of less phytotoxic effect when applied to seedling in their early growth stages, compared with chemical insecticides (Iovieno et al., 2002). Mineral oil is known from a long time such as an effective mean to control aphids and to reduce non-persistent viruses spread (Brachet et al., 2001).

Powell (1992) stated that mineral oil sprayed onto potato Y *potyvirus* (PVY) infected tobacco plants reduced acquisition of this virus by *Myzus persicae* aphids. Although the pre-penetration activities of aphids were longer on oil treated leaves, the inhibitory effect of the oil could not be attributed to differences in the duration of stylet penetration. Both acquisition and inoculation of the virus were reduced by the presence of oil on the plant surface. Transmission of PVY to sweet pepper by the green peach aphid, *M. persicae*, was inhibited by foliar applications of 1.0% or 2.0% neem seed oil to infected source plants or to uninfected recipient plants. Neem seed oil interfered with virus acquisition and inoculation in a manner comparable to that of commercial horticultural oil (Lowery et al., 1997).

Therefore, the aim of this investigation is to study *Mentha piperita*, *Ocimum basilicum*, *and Thymus vulgaris* essential oils and its action as a natural, safe, and reliable bio-controlling agent against some selected human microbial pathogens, also for avoiding plant virus transmission by vector insect which can further replace the risky and expensive chemical insecticides.

Materials and methods

Essential oil extraction

The oil extracts were obtained from 50 g from *M. piperita*, *O. basilicum*, and *T. vulgaris* fresh plants aerial parts by steam distillation according to Barazandeh (2002). This was performed by distillation for 40 min in an all-glass apparatus. The aqueous phase was extracted with dichloromethane (3×50 ml). The organic phase was dried with sodium sulfate

and filtered, and the solvent was evaporated using rotary evaporator until dryness. The oils were solubilized in ethyl acetate for gas chromatography and mass spectrometry analysis.

Gas chromatography/mass spectrometry (GC/MS) analysis

Analysis of the essential oils was carried out using an HP5890 Series II Gas Chromatograph, HP5972 mass selective detector and Agilent 6890 Series Autosampler (Agilent Technologies, USA). A Supelco MDN-5S 30 m × 0.25 mm capillary column (Sigma–Aldrich Co., USA) with a 0.5 μm film thickness was used with helium as the carrier gas at a flow rate of 1.0 ml/min. The GC oven temperature was programed at an initial temperature of 40 °C for 5 min, then heated up to 140 °C at 5 °C/min and held at 140 °C for 5 min and then heated to 280 °C at 9 °C/min and held for five additional minutes. Injector and detector temperatures were set at 250 °C. Mass spectrometry was run in the electron impact mode (EI) at 70 eV. The identification of the chemical constituents of the oil was determined by their retention indices and interpretation of their mass spectra and confirmed by mass spectral library search using the National Institute of Standards and Technology (NIST) database. The retention indices (RI) were calculated for all of the volatile constituents using a homologous series of C_8–C_{20} *n*-alkanes according to Van den Dool and Kratz (1963).

Preparations used

Raw oils were used for sensitivity test and in the form of emulsions for spraying application. Emulsions were prepared depending on commercial liquid dish wash soap (Peril®) and Tween 80 as emulsifying agents. Oil dilution was prepared by adding 5 ml of oil to 100 ml of distilled water with 3 ml from either soap or Tween 80. Controls were emulsion of cooking corn oil, water with emulsifying agents only, and water without emulsifiers or oil.

Source of selected pathogens

Oils' antimicrobial activity tests were carried out against *Escerichia coli*, *Psedumonas aeruginosa*, *S. aureus*, *Streptococcus pyogenes*, and *C. albicans* (kindly obtained from Dept. of Dermatology, Fac. of Medicine, Ain Shams Univ.), and *Rhizoctonia solani* was obtained from Dept. of Plant Pathology, Fac. of Agric., Ain Shams Univ.). *Ralstonia* (*Pseudomonas*) *solanacearum* was isolated from market potato tubers showing brown rot symptoms depending on Kelman's tetrazolium chloride (TZC) medium (Sigma–Aldrich, USA) (Kelman, 1954). *Potato virus y potyvirus* (common strain) (PVY^O) was isolated depending on indirect enzyme linked immunosorbant assay (I-ELISA) according to Koenig (1981) using specific polyclonal antibodies (Agdia Inc., USA) and *Chenopodium quinoa* as local lesion hosts and maintained on *Nicotiana tabacum* cv. White Burley plants.

Sensitivity test

To determine the antimicrobial activities of each tested essential oil, the disk diffusion method was utilized (Sisti et al., 2008). *E. coli*, *P. aeruginosa*, *S. aureus*, and *St. pyogenes* were cultured on nutrient agar media, while *C. albicans* was cultured on malt and yeast extract media. Cell suspensions (10^4 CFU ml^{-1}) were prepared from each microorganism in broth media, nutrient broth inoculated with each bacterial species was incubated for 24 h at 37 °C, and malt and yeast extract broth inoculated with yeast was incubated for 48 h at 30 °C. Sterile 6.3 mm filter paper disks were saturated with each tested essential oil preparation. The disks were allowed to dry at room temperature in a sterile airflow laminar chamber for 1 h and then they were placed in the center of fresh nutrient agar plates or malt and yeast extract agar plates previously seeded with 100 μl of cell suspension of each bacterial and fungal species, respectively. The cultures were incubated at 37 and 30 °C for 24 and 48 h for bacteria and yeast, respectively. Each experiment was replicated three times. Antibiotics were used as positive control; ampicillin was used as antibacterial standard, while miconazole nitrate was used as antifungal standard. The antimicrobial activities were evaluated by measuring the inhibition zone diameters (millimeters).

Inhibition of potato brown rot during preservation

Clean and surface sterilized (using 4% sodium hypochlorite for 10 min) market potato tubers (size of about 5 × 3 cm) were scratched using sterilized scalpel. Five tubers were used for each preparation and controls as follows: (a) Potatoes were sprayed with the preparation and then swapped with *R. solanacearum* using sterilized cotton swap dipped in the bacterial suspension (10^8 cells/ml). (b) Tubers were swapped with bacteria, then sprayed directly with each preparation, and also sprayed daily for 5 days. Each tuber was swapped with constant bacterial inoculum of 1.5 ml; treatments were kept on plastic tray at 4 °C and at room temperature (about 30 °C) as a control and observed daily for rot development till 20 days.

Effect of lavender oil on PVY^O transmission by M. persicae

Preparation of vector insects. M. persicae colonies were started from a non-viruliferous single virginiparous female (obtained from Plant Protech. Res. Instit., ARC, Dokki, Egypt) and reared on turnip plants in an environmental growth chamber under controlled conditions [23:16 °C (day/night) and a photoperiod of 16:8 h (light/dark)].

Groups of 30–40 individuals of *M. persicae* were collected, starved for 1 h and placed, for 1 h acquisition period, on PVY^O infected *N. tabacum* cv. White Burley plants, which had been inoculated 3 weeks previously. Negative controls were managed similarly using healthy plants.

Print-capture detection of PVY^O within M. persicae using immunocapture reverse transcriptase polymerase chain reaction. The PC/IC-RT-PCR was used to detect PVY^O within individual aphid. A sample of 20 aphids were squashed individually on Whatman 3MM paper using the round bottom of an Eppendorf tube. Insect was extracted from paper with 100 μl of 0.5% Triton X-100, the extract was then added to a 2 μg/ml of anti-PVY IgG precoated PCR tubes, and PCR protocol was performed according to Olmos et al. (1997) to detect the virus coat protein gene (*cp*). The following used primers (Invitrogen Corp., USA) were designed using Primer Premier software (PREMIER Biosoft International, USA) depending on the nucleotide sequence of PVY^O mentioned by Ghosh et al. (2002):

5′CAAATGACACAATTGATGCA3′ (Sense).
5′CACTTGTTCTTGACTCCAAGTAG3′ (Antisense).

PCR products (10 µl) were analyzed by 1.5% agarose gel electrophoresis. Bands size was determined from the gel photograph using Gel-Pro Analyzer software (Media Cybernetics, USA).

Insect transmission. Healthy tobacco plants (carrying five leaves) were treated by spraying with different preparations mentioned before (using 10 plants for each treatment). Aphids which were fed on infected plants transferred to feed on treated healthy plants (20 aphids per plant) for 1 h inoculation feeding period, then sprayed with insecticide, and kept in an insect-proof cage. After 15 days, plants were assayed for PVY^{O} infection using I-ELISA.

Results

Oil yield and chemical constituents

Oil yields were expressed in relation to dry weight of plant material, which found to be 0.39%, 0.20% and 0.55% w/w for *M. piperita*, *O. basilicum* and *T. vulgaris*, respectively.

The chemical composition of the essential oils obtained was analyzed by GC/MS analysis as shown in Table 1. The main compounds from *M. piperita* were menthol (35.44%) and menthone (20.11%), *O. basilicum* main component was linalool (45.11%), while *T. vulgaris* oil main component was thymol (75.76%).

Sensitivity test

Sensitivity test results for the bacteria and yeast used against essential oils obtained from *M. piperita*, *O. basilicum*, and *T. vulgaris* are shown in Table 2. Results indicated that the most effective oil against bacteria and yeast was that of *O. basilicum* followed by that of *T. vulgaris*. *O. basilicum* oil was highly effective on *S. pyogenes* giving a zone of 19 mm more than that produced by Ampicillin which was of 15 mm in diameter. Oil of *O. basilicum* was slightly more effective on *C. albicans* when compared with clotrimazole as an antifungal agent.

Table 1 Main constituents of the tested essential oils.

Compound	RI[a]	*Mentha piperita*	*Ocimum basilicum*	*Thymus vulgaris*
p-Cymene	1023			5.5
1,8-cineole	1029	5.9	6.4	
γ-Terpinene	1055			4.5
Linalool	1097		45.11	2.3
Menthone	1151	20.11		
Menthofuran	1162	11.81		
Menthol	1170	35.44		
Thymol	1288			75.76
Carvacrol	1295			4.63
Eugenol	1353		10.5	

[a] Retention index calculated on the basis of retention time of a mixture of *n*-alkanes (C_8–C_{30}).

Table 2 Sensitivity test results for the bacteria and yeast against some essential oils.

Oil source plant	Inhibition zone diameter (mm)				
	Bacteria				Yeast
	Gram negative		Gram positive		
	Escerichia coli	*Psedumonas aeruginosa*	*Staphylococcus aureus*	*Streptococcus pyogenes*	*Candida albicans*
Mentha piperita	9	12	11	12	8
Ocimum basilicum	16	14	17	19	15
Thymus vulgaris	11	13	14	16	13
Ampicillin	22	19	13	15	–
Clotrimazole	–	–	–	–	14

Table 3 Potato brown rot inhibition by some essential oils.

Condition		Treatment										
		Number of tubers showing rot symptoms[a]										
		Tween			Soap			Corn oil and tween	Corn oil and soap	Water and tween	Water and soap	Water
		Mp	*Ob*	*Tv*	*Mp*	*Ob*	*Tv*					
Preserved at 4 °C	A	7	3	4	5	2	3	4	3	9	7	10
	B	8	5	6	6	4	3	4	3	10	7	10
Preserved at RT	A	8	5	7	6	4	5	5	3	10	8	10
	B	8	7	8	7	5	5	5	3	10	8	10

Mp = *M. piperita*, *Ob* = *O. basilicum* and *Tv* = *T. vulgaris* oils emulsified with either tween or soap.
[a] Ten potato tubers were reserved at 4 °C or at room temperature (RT).
A: Spraying prior to infection, B: Infection before spraying.

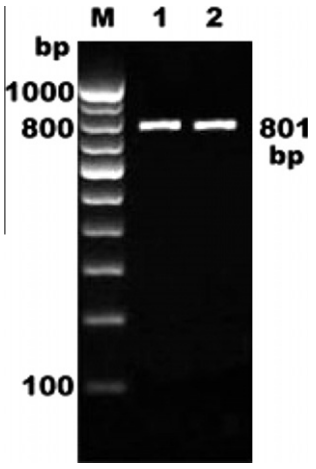

Fig. 1 PC/IC-RT-PCR for detection of PVY^O *cp* gene within individual aphid. M: 1000 bp DNA marker (Invitrogen, USA).

Inhibition of potato brown rot during preservation

Results in Table 3 revealed that spraying potato tubers with *O. basilicum* oil emulsified with soap prior to *R. solanacearum* infection and preservation at 4 °C gave the best results followed by *T. vulgaris* oil with soap, as only 2 and 3 tubers out of 10 used shows rot symptoms for *O. basilicum* and *T. vulgaris* oils, respectively. While high number of rotting tubers was obtained with *M. piperita* and tween treatments.

Effect of lavender oil on PVY^O transmission by M. persicae

Detection of PVY^O within M. persicae using PC/IC-RT-PCR

Print-capture PCR was used successfully for detecting *potato virus y potyvirus* (common strain) (PVY^O) within individual aphid. All of the 20 aphids studied gave positive PCR results in the form of *cp* gene bands with the expected size of 801 bp (Fig. 1). This result assumed that approximately all the insects which will be used to study virus transmission were carrying virus particles.

Insect transmission

Using *O. basilicum* oil emulsified with soap for spraying tobacco plants revealed excellent results Table 4, as eight plants out of 10 treated confirmed to be PVY^O-free by giving negative I-ELISA results. Followed by *T. vulgaris* oil with soap which gave six negative plants, also *O. basilicum* oil with tween gave good results for six plants which were found to be virus-free. It was observed that adding soap as an emulsifier has a killing effect on aphids as controls gave 4 and 3 negative plants for corn oil and water both with soap, respectively.

Table 4 Effect of oils treatments on PVY^O transmission by *M. persicae*.

Plant no.	Treatment										
	I-ELISA values and results[a]										
	Tween			Soap			Corn oil and tween	Corn oil and soap	Water and tween	Water and soap	Water
	Mp	*Ob*	*Tv*	*Mp*	*Ob*	*Tv*					
1	0.215 −	0.091 −	0.097 −	0.215 −	0.099 −	1.055 +	0.091 −	1.003 +	0.851 +	0.202 −	1.086 +
2	0.211 −	1.058 +	0.801 +	0.095 −	0.204 −	0.911 +	0.212 −	0.871 +	0.715 +	0.905 +	1.099 +
3	0.699 +	0.085 −	0.191 −	0.981 +	0.644 +	0.052 −	0.894 +	0.094 −	1.025 +	1.085 +	0.967 +
4	0.891 +	0.233 −	0.900 +	0.202 −	0.055 −	0.885 +	0.699 +	0.799 +	1.200 +	1.001 +	0.854 +
5	1.058 +	0.915 +	1.003 +	0.771 +	0.199 −	0.200 −	0.999 +	0.202 −	0.941 +	0.947 +	0.911 +
6	1.098 +	0.205 −	0.912 +	0.211 −	0.177 −	0.812 +	1.094 +	0.914 +	0.711 +	0.885 +	1.022 +
7	1.122 +	0.998 +	0.201 −	0.811 +	0.097 −	0.241 −	0.851 +	0.899 +	0.992 +	1.100 +	0.941 +
8	0.811 +	0.097 −	0.094 −	0.051 −	0.200 −	0.041 −	1.044 +	0.928 +	1.099 +	0.911 +	0.881 +
9	0.081 −	0.215 −	0.819 +	0.915 +	0.098 −	0.094 −	0.912 +	0.200 −	1.033 +	0.091 −	1.056 +
10	0.209 −	0.892 +	0.785 +	1.002 +	0.785 +	0.243 −	1.109 +	0.089 −	0.851 +	0.211 −	0.923 +
Healthy[a]	0.210 −	0.197 −	0.231 −								
Infected[b]	1.087 +	0.977 +	0.952 +								

Mp = *M. piperita*, *Ob* = *O. basilicum* and *Tv* = *T. vulgaris* oils emulsified with either tween or soap.

[a] Each I-ELISA value (at 405 nm) was the average of three readings, Results: + positive, − negative.

[b] Controls I-ELISA values for healthy and infected (15 days postinoculation) tobacco plants, without any treatment.

Discussion

Food processors, food safety researchers, and regulatory agencies have been increasingly concerned with the growing number of food-borne illness outbreaks caused by some bacteria (Friedman et al., 2002). Infections due to bacterial species also remain a serious therapeutic problem as emerging resistance of these species is seriously decreasing the number of effective antimicrobials. The food industry has tended to reduce the use of chemical preservatives in their products due to increasing pressure of consumers or legal authorities, to either completely remove or to adopt more natural alternatives for the maintenance or extension of product shelf life (Nychas, 1995).

Plants and their essential oils are potentially useful sources of antimicrobial compounds. Numerous studies have been published on the antimicrobial activities of plant compounds against many different types of microbes, including food-borne pathogens (Tassou et al., 2000; Rančić et al., 2005).

It was found that linalool have the highest inhibitory effect over other aromatic plants' essential oils components (Sonbolia et al., 2006; Bassolé and Juliani, 2012). Among the aromatic plant studied, the major constituents found were the monoterpenes, 1,8-cineole and linalool. These compounds are previously known for its antimicrobial activity (Baba-Moussa et al., 1999; Sonbolia et al., 2006). Helander et al. (1998) attributed the 1,8-cineole and linalool antimicrobial action to its alcoholic characteristics which can cause bacterial and fungal membrane-disturbing activities.

Monoterpenes play the most important role in protecting the experimented plants as antimicrobial agents (Wilkinson and Cavanagh, 2005) especially as antivirus. It may dissolve or penetrate the viral capsules and denature its proteins or nucleic acids (DNA or RNA). Also, it may play a role in denaturing elicitor proteins produced by these pathogens and preventing the cell death program induction in the infected plant cells and induces the transcription of genes encode the enzymes of interior phytoalexin synthesis in these infected plants (Scheel and Parker, 1990).

O. basilicum oil was found to be rich with linalool which may explain its good sensitivity test results obtained against some harmful bacteria and yeast during this investigation. The constituent's data were in harmony with Sartoratto et al. (2004) and Pistelli et al. (2012) who found that *O. basilicum* oil is rich in linalool with high antimicrobial effect.

Concerning potato brown rot inhibition experiment results concluded that potato tubers can be kept safe from brown rot at 4 °C by spraying them with *O. basilicum* oil emulsified with soap. Besides the microbial inhibitory effect of *O. basilicum* oil, the presence of soap can help in increasing this effect and gave an efficient method in protecting potatoes from infection.

Print-capture PCR was used successfully for detecting *potato virus y potyvirus* (common strain) (PVY^O) within individual aphid. The obtained results and test efficiency were in harmony with what performed by Varveri (2000), who detected potato, Y *potyvirus* in single *M. persicae*.

The obtained results favored the use of *O. basilicum* oil with soap, for oil can suppress insect infestation, while soap has a killing effect on delicate insects with sucking mouthparts like aphids. Concerning such point of view, in-harmony results were also found by (Lowery et al., 1997; Asjes, 2000) as they studied the efficiency of combining oil with soap for controlling virus vector insects. Plant essential oils emulsified with soap will be harmless to human health when compared with the risk of using chemical insecticide (Iovieno et al., 2002).

Using oil in soap preparations can increase the killing effect on insects with sucking mouthparts by suffocation (Asjes and Blom-Barnhoorn, 2002; Gorski and Tomczak, 2010). This can manage the insects damaging effects on crops and also reducing their ability of pathogens transmission especially viruses, resulting an increase in crops quality and quantity.

References

Abdel-Reheem, M.A.T., Moussa, Zaynab A., Oraby, Mona M., Aboud, Fatma S., Abdou, Dalia A.M., 2012. Antimicrobial activity of lavender oil treated with nicotinamide. Afr. J. Microbiol. Res. 21, 4574–4584.

Alviano, W.S., Mendonca-Filho, R.R., Alviano, D.S., Bizzo, H.R., Souto-Padron, T., Rodrigues, M.L., Bolognese, A.M., Alviano, C.S., Souza, M.M., 2005. Antimicrobial activity of *Croton cajucara* Benth linalool-rich essential oil on artificial biofilms and planktonic microorganisms. Oral Microbiol. Immunol. 20, 101–105.

Ames, B.N., Gold, L.S., 1989. Pesticides, risk and applesauce. Science 244, 754–755.

Asjes, C.J., 2000. Control of aphid-borne Lily symptomless virus and Lily mottle virus in Lilium in the Netherlands. Virus Res. 71, 23–32.

Asjes, C.J., Blom-Barnhoorn, G.J., 2002. Control of aphid vector spread of Lily symptomless virus and Lily mottle virus by mineral oil/insecticide sprays in Lilium. Acta Hort. 570, 277–281.

Baba-Moussa, F., Akpagana, K., Bouchet, P., 1999. Antifungal activities of seven West African Combretaceae used in traditional medicine. J. Ethnopharmacol. 66, 335–338.

Barazandeh, M.M., 2002. Essential oil composition of *Lavandula latifolia* Medik from Iran. J. Essent. Oil Res. 14, 103–104.

Bassolé, I.H.N., Juliani, H.R., 2012. Essential oils in combination and their antimicrobial properties. Molecules 17, 3989–4006.

Bell, E.A., Charlwood, B.V., 1980. Secondary plant products. Encyclopedia of Plant Physiology, vol. 8. Springer Verlag, Berlin.

Brachet, P., Pinasseau, F., Cailleteau, B., Verrier, J.L., 2001. Preventative control of non-persistent aphid transmitted tobacco virus in France. In: CORESTA Congress Proceedings, Cape Town (South Africa), p. 58.

Burt, S., 2004. Essential oils: their antimicrobial properties and potential applications in foods: a review. Int. J. Food Microbiol. 94, 223–253.

Friedman, M., Henika, R.P., Mandrell, E.R., 2002. Bactericidal activities of plant essential oils and some of their isolated constituents against *Campilobacter jejuni*, *Escherichia coli*, *Listeria monocytogenes*, and *Salmonella enterica*. J. Food Protect. 65, 1545–1560.

Ghosh, S.B., Nagi, L.H.S., Ganapathi, T.R., Paul Khurana, S.M., Bapat, V.A., 2002. Cloning and sequencing of Potato virus Y coat protein gene from an Indian isolate and development of transgenic tobacco for PVY resistance. Curr. Sci. 82, 7–10.

Gorski, R., Tomczak, M., 2010. Usefulness of natural essential oils in the control of Foxglove aphid (*Aulacorthum solani* Kalt) occurring on eggplant (*Solanum melongena* L.). Ecol. Chem. Eng. 17, 345–349.

Helander, I.M., Alakomi, H.L., Latva-Kala, K., Mattila-Sandholm, T., Pol, I., Smid, E.J., Gorris, L.G.M., Von Wright, A., 1998. Characterization of the action of selected essential oil components on Gram-negative bacteria. J. Agric. Food Chem. 46, 3590–3595.

Iovieno, P., Lahoz, E., Porrone, F., Biondani, Cristina., Carella, Angela., Contillo, R., 2002. Use of light mineral oil to reduce the severity of PVY and CMV infections on tobacco in Italy. Il Tabacco 10, 57–61.

Koenig, R.C., 1981. Indirect ELISA methods for broad specificity detection of plant viruses. J. Gen. Virol. 55, 53–62.

Lowery, D.T., Eastwell, K.C., Smirle, M.J., 1997. Neem seed oil inhibits aphid transmission of potato virus Y to pepper. Ann. Appl. Biol. 130, 217–225.

Mourey, A., Canillac, N., 2002. Anti-Listeria monocytogenes activity of essential oils components of conifers. Food Control 13, 289–292.

Nychas, G.J.E., 1995. Natural antimicrobials from plants. In: Gould, G.W. (Ed.), New Methods of Food Preservation Academic Professional. Blackie, London, pp. 58–89.

Olmos, A., Cambra, M., Dasi, M.A., Candresse, T., Esteban, O., Gorris, M.T., Asensio, M., 1997. Simultaneous detection and typing of plum pox potyvirus (PPV) isolates by heminested PCR and PCR–ELISA. J. Virol. Methods 68, 127–137.

Pistelli, L., Mancianti, F., Bertoli, A., Cioni, P.L., Leonardi, M., Pisseri, .F., Mugnaini, L., Nardoni, S., 2012. Antimycotic activity of some aromatic plants essential oils against Canine Isolates of *Malassezia pachydermatis*: an in vitro assay. Open Mycol. J. 6, 17–21.

Powell, G., 1992. The effect of mineral oil on stylet activities and potato virus Y transmission by aphids. Entomol. Exp. Appl. 63, 237–242.

Rančić, A., Soković, M., Vukojević, J., Simić, A., Marin, P., Duletić-Laušević, S., Doković, D., 2005. Chemical composition and antimicrobial activities of essential oils of *Myrrhis odorata* (L.) Scop, *Hypericum perforatum* L. and *Helichrysum arenarium* (L.) Moench. J. Essent. Oil Res. 17, 341–345.

Sartoratto, A., Machado, Ana L.M., Delarmelina, Camila, Figueira, G.M., Duarte, Marta Cristina T., Rehder, Vera Lúcia G., 2004. Composition and antimicrobial activity of essential oils from aromatic plants used in Brazil. Braz. J. Microbiol. 35, 275–280.

Schafer, H., Wink, M., 2009. Medicinally important secondary metabolites in recombinant microorganisms or plants: progress in alkaloid biosynthesis. Biotechnol. J. 4, 1684–1703.

Scheel, D., Parker, J.E., 1990. Elicitor recognition and signal transduction in plant defense gene activation. Z. Naturforsch. 45, 569–575.

Sisti, M., DeSanti, M., Fraternale, D., Ninfali, P., Scoccianti, V., Brandi, G., 2008. Antifungal activity of *Rubus ulmifolius* Schott standardized in vitro culture. LWT 41, 946–950.

Sonbolia, A., Babakhanib, B., Mehrabianc, A.R., 2006. Antimicrobial activity of six constituents of essential oil from salvia. Z. Naturforsch. 61, 160–164.

Tassou, C.C., Koustomanis, K., Nychas, J.G., 2000. Inhibition of *Salmonella enteritidis* and *Staphylococcus aureus* in nutrient broth by mint essential oil. Food Res. Int. 33, 273–280.

van de Braak, S.A.A.J., Leijten, G.C.J.J., 1999. Essential Oils and Oleoresins: A Survey in the Netherlands and Other Major Markets in the European Union. CBI, Centre for the Promotion of Imports from Developing Countries, Rotterdam, The Netherlands, p. 116.

Van den Dool, H., Kratz, P.D.A., 1963. Generalization of the retention index system including linear temperature programmed gas–liquid partition chromatography. J. Chrom. 11, 463–471.

Varveri, Christina, 2000. Potato Y potyvirus detection by immunological and molecular techniques in plants and aphids. Phytoparasitica 28, 2.

Wilkinson, M.J., Cavanagh, H.M.A., 2005. Antibacterial activity of essential oils from Australian native plants. Phytother. Res. 19, 643–646.

Wink, M., 1999. Functions of Plant Secondary Metabolites and their Exploitation in Biotechnology. Sheffield Academic Press, Sheffield, UK, pp. 362.