

INSECTICIDAL ACTIVITIES OF ESSENTIAL OILS FROM SOME CULTIVATED AROMATIC PLANTS AGAINST *SPODOPTERA LITTORALIS* (BOISD)

Salaheddine Souguir*, Ikbal Chaieb, Zohra Ben Cheikh, Asma Laarif

Entomological Laboratory, Regional Research Center in Horticulture and Organic Agriculture, Chott Mariem 4042, Sousse, Tunisia

Received: February 1, 2013

Accepted: October 16, 2013

Abstract: Medicinal plant species were tested for their fumigant activity against *Spodoptera littoralis* third instar larvae. Responses varied according to plant species and parts used. For the present investigation, volatile oils were obtained from: *Foeniculum vulgare* (flowers and seeds), *Coriandrum sativum* (seeds), *Daucus carota* (flowers), *Pelargonium graveolens* (leaves and flowers), *Origanum majorana* (leaves and flowers), and *Salvia officinalis* (leaves). Fumigant activity was observed after 24 hours of exposure. All essential oils were proved to be toxic to the third instar larvae. However, the highest mortality was observed in the essential oil of *S. officinalis* leaves, *C. sativum* seeds, *F. vulgare* seeds, *D. carota* flowers, and *O. majorana* leaves with $LC_{50} = 23.050 \mu\text{l/l}$ air, $68.925 \mu\text{l/l}$ air, $95.075 \mu\text{l/l}$ air, $99.300 \mu\text{l/l}$ air, and $100.925 \mu\text{l/l}$ air, respectively. Other oils showed an LC_{50} between 101 and $183 \mu\text{l/l}$ air.

Key words: *Coriandrum sativum*, *Daucus carota*, essential oils, *Foeniculum vulgare*, Fumigant activity, LC_{50} , *Origanum majorana*, *Pelargonium graveolens*, *Salvia officinalis*, *Spodoptera littoralis*

INTRODUCTION

Insect pests are a major constraint on crop production, especially in developing countries. Due to the growing concerns over health hazards, environmental pollution, and negative effects on non-target organisms, essential oils have begun to play an increasingly prominent role as alternatives to synthetic pesticides (Sharma *et al.* 2006). Many floral volatiles have anti-microbial or anti-herbivore activity (De Moraes *et al.* 2001; Friedman *et al.* 2002; Hammer *et al.* 2003). Such activity acts to protect valuable reproductive parts of plants from damage (Dudareva *et al.* 2004).

However, intensive screening is necessary to select compounds with pesticidal properties, which are harmless to the environment and ecosystem. Research is urgently needed on potential botanical extracts which are safe and which leave little or no residues, and which are naturally derived with minimal technology. There are more than 2400 plant species belonging to 189 plant families which are said to be rich sources of bioactive organic compounds (Rao *et al.* 2005).

The *Spodoptera littoralis* (Boisd) adult lays between 300 and 500 eggs. The larva attacks the foliage of plants. This is the most serious pest of various crops such as cotton, chilli, and tobacco. Those crops are economically important. This pest has already developed resistance to many chemical pesticides (Niranjankumar and Regupathy 2001).

The aim of our study was to assess the insecticide activity of the essential oils obtained by six Mediterranean plants and their different parts, against the third instar larvae of *S. littoralis*, using the fumigant bioassay.

MATERIALS AND METHODS

Plant material and essential oils extraction

Origanum majorana L., *Foeniculum vulgare* L., *Coriandrum sativum* L., *Daucus carota* L., *Pelargonium graveolens* L'Hér., and *Salvia officinalis* L. were cultivated on the biological fields of the Regional Center for Research on Horticulture and Organic Agriculture (RCRHOA).

Essential oils were extracted from the used plant parts using a Clevenger-type water steam distillation apparatus (Papachristos and Stamopoulos 2004). The distilled essential oils were stored in the refrigerator at 4°C until being used in the bioassay.

Insect rearing

Spodoptera littoralis (Lepidoptera: Noctuidae) were reared on an artificial diet in the laboratory. Rearing conditions were: a 12 h photo regime at $28\pm 2^{\circ}\text{C}$ and $75\pm 5\%$ relative humidity (RH). Insect cultures were continuously refreshed with wild moths captures from the RCRHOA organic farm (El-Minshawy *et al.* 2009).

*Corresponding address:
souguir.salaheddine@hotmail.fr

Table 1. Investigated plant material

Botanical name	Family	Part used	Yield [%]
<i>F. vulgare</i>	Umbelliferae	seeds	2.78 ^d ±0.308
		flowers	1.02 ^c ±0.267
<i>O. majorana</i>	Lamiaceae	leaves	1.22 ^c ±0.300
		flowers	0.93 ^c ±0.081
<i>P. graveolens</i>	Geraniaceae	leaves	0.18 ^a ±0.080
		flowers	0.24 ^{ab} ±0.100
<i>C. sativum</i>	Umbelliferae	seeds	0.54 ^b ±0.118
<i>D. carota</i>	Umbelliferae	flowers	0.15 ^a ±0.025
<i>S. officinalis</i>	Lamiaceae	leaves	0.43 ^{ab} ±0.050

Small letters – comparison between yields of essential oil in the same plant part (Duncan's test $p < 0.005$)

Insecticidal activity: Fumigant Test

The insecticidal activity of the essential oils against the third instar larvae of *S. littoralis*, was determined by fumigant bioassay using the closed container method. A group of 10 larvae were put into the bottom of a 40 ml the plastic container. Paper discs were treated with different concentrations of essential oils: 0, 25, 50, 100, and 200 µl/l air. The discs were then attached to the inside top of the container and the container was then closed.

Mortality was determined after 24 h of the treatments. All bioassays were performed in 5 repetitions. All of the treated larvae were held in the same rearing conditions. An insects was considered dead if it did not move when observed outside of the container.

Statistical analysis

The data were corrected using Abbott's formula (Abbott 1925) for the mortalities then subjected to probit analyses using SPSS (v. 2011) to estimate LC₅₀ and LC₉₀ the values of each of the essential oils against the third instar larvae of *S. littoralis* (Finney 1971). Means were separated at the 5% significance level by Duncan's test.

RESULTS

Rates of oil production

The oil production yields of the different parts of the tested plants are given in table 1. The results showed that seeds and flowers of *F. vulgare*, and the leaves and flow-

ers of *O. majorana* produced the highest rate of oil, with a yield > 1%.

Insecticidal activity of essential oils against *S. littoralis* larvae (Third instar)

All the essential oils tested were active towards third instar larvae of *S. littoralis*. All of them caused mortality: a dose of 200 µl/l air was required to obtain over 80% of mortalities for essential oils of *O. majorana* (flowers), and *F. vulgare* (flowers and seeds).

The essential oils of *C. sativum* (seeds) and *D. carota* (flowers) at the lowest tested dose of 100 µl/l air, caused 72% and 70% mortality, respectively. While a dose of 50 µl/l air was required to obtain over 92.5% mortality for essential oils of *S. officinalis* (Table 2).

Among the nine tested essential oils, the most potent insecticidal oil was extracted from *S. officinalis* leaves LC₅₀ = 23.050 µl/l air (LC₉₀ = 41.625 µl/l air). The next most potent insecticidal oil was extracted from *C. sativum* seeds LC₅₀ = 68.925 µl/l air (LC₉₀ = 125.475 µl/l air), followed by *D. carota* essential oils from the flowers (EO) by LC₅₀ = 99.300 µl/l air (LC₉₀ = 170.550 µl/l air), leaves of *O. majorana* EO by LC₅₀ = 100.925 µl/l air (LC₉₀ = 250.425 µl/l air), and *F. vulgare* seeds EO by LC₅₀ = 101.050 µl/l air (LC₉₀ = 199.825 µl/l air). The flowers of *O. majorana* essential oils had an LC₅₀ = 104.725 µl/l air (LC₉₀ = 216.350 µl/l air). The least potent essential oils were those of the flowers and leaves of *P. graveolens* by LC₅₀ = 179.300 µl/l air (LC₉₀ = 299.750 µl/l air) and LC₅₀ = 182.350 µl/l air (LC₉₀ = 287.975 µl/l air), respectively (Table 3).

Table 2. Mortality in percent of *S. littoralis* larvae, after 24 h of exposure to different concentrations of essential oils

Parts of plant	Concentration of EO's [µl/l air]				
	0	25	50	100	200
<i>F. vulgare</i> flowers	0 ^a .A±0.00	12.50 ^a .AB±5.00	47.50 ^b .B±25.00	45 ^b .ABC±33.17	87.50 ^c .CD±12.58
<i>F. vulgare</i> seeds	0 ^a .A±0.00	15 ^b .AB±5.77	32.50 ^c .BC±12.58	65 ^d .BC±12.91	87.50 ^c .CD±5.00
<i>O. majorana</i> leaves	0 ^a .A±0.00	37.50 ^b .C±17.08	40 ^b .BC±21.60	60 ^b .BC±35.59	72.50 ^b .BC±26.30
<i>O. majorana</i> flowers	0 ^a .A±0.00	25 ^b .BC±10.00	27.50 ^b .BC±5.00	60 ^b .BC±14.14	80 ^d .CD±21.60
<i>C. sativum</i> seeds	0 ^a .A±0.00	24 ^b .BC±13.42	36 ^b .BC±15.17	72 ^c .CD±21.68	100 ^d .D±0.00
<i>D. carota</i> flowers	0 ^a .A±0.00	4 ^a .A±5.48	20 ^b .AB±12.25	70 ^c .C±18.71	90 ^d .CD±12.25
<i>P. graveolens</i> leaves	0 ^a .A±0.00	4 ^a .A±5.48	6 ^{ab} .A±5.48	16 ^b .A±8.94	58 ^c .AB±13.04
<i>P. graveolens</i> flowers	0 ^a .A±0.00	4 ^a .A±5.48	6 ^a .A±5.48	34 ^b .AB±16.73	52 ^c .A±8.37
<i>S. officinalis</i> leaves	0 ^a .A±0.00	70 ^c .D±14.14	92.50 ^d .D±9.57	100 ^d .D±0.00	100 ^d .D±0.00

Small letters – comparison between doses of same essential oil (Duncan's test $p < 0.005$)

Capital letters – comparison between essential oils in the same concentrations (Duncan's test $p < 0.005$)

Table 3. Calculated lethal concentrations (LC₅₀ and LC₉₀) of third instar larvae of *S. littoralis* exposed to different essential oils

Parts of plants	Lethal concentrations [µl/l air]	
	LC ₅₀	LC ₉₀
<i>O. majorana</i> leaves	100.925 (30–510.75)	250.425 (230.001–865.258)
<i>O. majorana</i> flowers	104.725 (47.225–248.875)	216.350 (143.425–745.550)
<i>F. vulgare</i> flowers	101.050 (27–502.65)	199.825 (125.650–1905.250)
<i>F. vulgare</i> seeds	95.075 (50.200–177.800)	184.150 (128.200–439.050)
<i>C. sativum</i> seeds	68.925 (47.600–105.050)	125.475 (94.175–226.825)
<i>D. carota</i> flowers	99.300 (48.725–220.950)	170.550 (116.325–533.250)
<i>P. graveolens</i> leaves	182.350 (166.825–202.125)	287.975 (259.525–327.825)
<i>P. graveolens</i> flowers	179.300 (123.800–421.425)	299.750 (207.875–904.625)
<i>S. officinalis</i> leaves	23.050 (17.050–31.475)	41.625 (32.800–61.150)

DISCUSSION

Essential oils (EO) from plants may be an alternative source of *S. littoralis* third instar larvae control. The reason for this is because EO are a source of bioactive compounds that are safe for human health and the environment.

The first reason we chose these particular plant parts was because they were available during the season of the year when we decided to do our research. The second reason was for the yields of essential oils extracted from these parts.

Many researches have reported on the effectiveness of plant essential oils against insects, especially stored-products insects. In our study, we are concentrated on *S. littoralis*. Pavela (2004) reported that some medicinal plants essential oils are larvicidal to the third instar larvae of *S. littoralis*. Krishnappa *et al.* (2010) tested oils obtained from *Thymus persicus* L. also on *S. littoralis* larvae.

Results showed that 100 µl/l of air, toxic enough to obtain 100% mortality of *S. littoralis* third instar larvae, using the *S. officinalis* essential oils. We needed 200 µl/l air from *C. sativum* (seeds), and *D. carota* (flowers) essential oils to obtain 100% mortality. But to realize 100% mortality for the others essential oils used in this study, we needed a little bit more than 200 µl/l air.

Compared with other researches, de Sousa *et al.* (2005) reported that *C. sativum* essential oils kill 53.99% of *Callosobruchus maculatus* (Fabricius) at a concentration of 2.5% (w/w). Hazrat and Soaib (2012) also showed that those essential oils from *C. sativum* are effective against Mosquito larvae.

Rana *et al.* (2012) found that EO from *F. vulgare* kills 100% of *Culex quinquefasciatus* (Linnaeus) larvae at 250 ppm after 40 min. In addition, Pavela (2004) tested oils from *O. majorana* and *S. officinalis* against *S. littoralis*

larvae. He showed that when using 10% (w/v) from each oil, he was able to obtain a 77.8% mortality with the use of *O. majorana*, and 97.7% with the use of *S. officinalis*.

Based on the Probit analysis, EO from the leaves of *S. officinalis* revealed that LC₅₀ = 23.050 µl/l air. Oils obtained from *F. vulgare* seeds, *C. sativum* seeds, and *D. carota* flowers, showed an LC₅₀ < 100 µl/l air. But with other oils extracted from *O. majorana* (leaves and flowers), *F. vulgare* flowers, and *P. graveolens* (leaves and flowers), we obtained an LC₅₀ > 100 µl/l air.

Likewise, a lethal dose (LC₅₀) has been determined by many authors. Rana and Rana (2012) tested *F. vulgare* EO against *C. quinquefasciatus* and found an LC₅₀ = 24.69 ppm. Ebadollahi (2011) evaluated oils from *F. vulgare* against *Sitophilus granarius* (Linnaeus) (LC₅₀ = 27.30 µl/l air) and against *S. oryzae* (Linnaeus) (LC₅₀ = 44.16 µl/l air). Sedaghat *et al.* (2011) studied the effect of *C. sativum* essential oils against *Anopheles stephensi*, and determined an LC₅₀ = 120.95 ppm. Pavela and Chermenskaya (2004) tested oils of *S. officinalis* against the third instar larvae of *S. littoralis*, LC₅₀ = 4.7 µg/ml.

These results demonstrate that the essential oils of *S. officinalis* leaves, *C. sativum* seeds, *D. carota* flowers, and *F. vulgare* seeds, may be serving as a lepidopteran agricultural pest control of *S. littoralis*.

The possible sites of action of essential oil toxicity are acetylcholinesterase and the octopamingeric system in insects (Kostyukovsky *et al.* 2002; Evans 1981).

CONCLUSION

The increasing number of investigations on plant-insect chemical interactions revealed that plant secondary metabolites, like essential oils, can be used as a pest control agent. This study could also contribute toward

preserving the agroecological systems that are in danger because of excessive use of synthetic insecticides.

Essential oils can be mixed with some water. It is important to be careful with the concentrations to be used on those hothouse plants that need to be protected from attacks of *S. littoralis* larvae. There is also a need to develop extraction methods which are easy, understandable for users, and can be used at the farm level.

ACKNOWLEDGMENTS

The authors thank Ms Kahouli Souad and Chhaibi Kawther, for their help in insect rearing and bioassay realization.

REFERENCES

- Abbott W. 1925. A method for computing the effectiveness of an insecticide. *J. Econ. Entomol.* 18: 265–267.
- De Moraes C.M., Mescheer M.C., Tumlinson J.H. 2001. Caterpillar induced nocturnal plant volatiles repel nonspecific females. *Nature* 410: 577–580.
- De Sousa A.H., Maracajá P.B., da Silva R.M., de Moura A.M., de Andrade W.G. 2005. Bioactivity of vegetal powders against *Callosobruchus maculatus* (Coleoptera: Bruchidae) in caupi bean and seed physiological analysis. *Rev. Biol. Ciênc. Terra.* 5 (2): 96–103.
- Dudareva N., Pichersky E., Gershenzon J. 2004. Biochemistry of Plant Volatiles. *Plant Physiol.* 135 (4): 1893–1902.
- Ebadollahi A. 2011. Susceptibility of Two *Sitophilus* species (Coleoptera: Curculionidae) to Essential Oils from *Foeniculum vulgare* and *Satureja hortensis*. *Ecologia Balkanica* 3 (2): 1–8.
- El-Minshawy A.M., Zeid M. 2009. Rearing the Larvae of the Cotton Leaf Worm *Spodoptera littoralis* (Boisd.) on Semi-Artificial Diet. *Zeitschrift für Angewandte Entomologie* 70 (1–4): 101–104.
- Evans P. 1981. Multiple receptor types for octopamine in the locust. *J. Physiol.* 318: 99–122.
- Finney D.L. 1971. *Probit Analysis*, 3rd Ed. Cambridge University Press, Cambridge, UK, 333 pp.
- Friedman M., Henika P.R., Mandrell R.E. 2002. Bactericidal activities of plant essential oils and some of their isolated constituents against *Campylobacter jejuni*, *Escherichia coli*, *Listeria monocytogenes* and *Salmonella enteric*. *J. Food Prot.* 65 (10): 1545–1560.
- Hammer K.A., Carson C.F., Riley T.V. 2003. Antifungal activity of the components of *Melaleuca alternifolia* (tea tree) oil. *J. Appl. Microbiol.* 95 (4): 853–860.
- Hazrat B., Soaib A.H. 2012. Plants secondary metabolites for mosquito control. *Asian Pac. J. Trop. Dis.* 12: 166–168.
- Kostyukovsky M., Rafaeli A., Gileadi C., Demchenko N., Shaaya E. 2002. Activation of octopaminergic receptors by essential oil constituents isolated from aromatic plants: possible mode of activity against insect pests. *Pest Management Sci.* 58 (11): 1101–1106.
- Krishnappa K., Elumalai K., Anandan A., Govindarajan M., Mathivanan T. 2010. Insecticidal properties of *Thymus persicus* essential oil and their chemical composition against armyworm, *Spodoptera litura* (Fab.) (Lepidoptera: Noctuidae). *International J. Rec. Sci. Res.* 8: 170–176.
- Niranjankumar B.V., Regupathy A. 2001. Status of insecticide resistance in tobacco caterpillar *Spodoptera litura* (Fabricius) in Tamil Nadu. *Pestic. Res. J.* 13 (1): 86–89.
- Pavela R. 2004. Insecticidal activity of certain medicinal plants. *Fitoterapia* 75 (7–8): 745–749.
- Pavela R., Chermenskaya T. 2004. Potential insecticidal activity of extracts from 18 species of medicinal plants on larvae of *Spodoptera littoralis*. *J. Plant Protect. Sci.* 40 (4): 145–150.
- Papachristos D.P., Stamopoulos D.C. 2004. Fumigant toxicity of three essential oils on the eggs of *Acanthoscelides obtectus* (Say) (Coleoptera: Bruchidae). *J. Stored Products Res.* 40 (5): 517–525.
- Rana I.S., Rana A.S. 2012. Efficacy of essential oils of aromatic plants as larvicide for the management of filarial vector *Culex quinquefasciatus* Say (Diptera: Culicidae) with special reference to *Foeniculum vulgare*. *Asian Pac. J. Trop. Dis.* 2 (3): 184–189.
- Rao N.V., Maheswari T.U., Manjula K. 2005. Review on Botanical Pesticides as Tools of Pest Management. Narosa Publishing House Pvt., Ltd., New Delhi, India, p. 1–16.
- Sedaghat M.M., Dehkord A.S., Abai M.R., Khanavi M., Mohtarami F., Salim Y. 2011. Larvicidal Activity of Essential Oils of Apiaceae Plants against Malaria Vector, *Anopheles stephensi*. *Iran J. Arthropod-Borne Dis.* 5 (2): 51–59.
- Sharma A., Kaushal P., Sharma K.C., Kumar R. 2006. Bioefficacy of some plant products against Diamondback moth *Plutella xylostella* L. (Lepidoptera: Yponomeutidae). *J. Entomol. Res. Soc.* 30: 213–217.
- SPSS. SPSS Version 20. SPSS Inc, 233 S. Wacker Drive; Chicago, Illinois: 2011.