UKaaZ

Annals of Phytomedicine 5(2): 87-90, 2016
Journal homepage: www.ukaazpublications.com
Print ISSN : 2278-9839 and Online ISSN : 2393-9885

ANNALS OF PHYTOMEDICINE
An International Journal

Original article

Exploration of various essential oils as fumigant to protect stored grains from insect damage

Vandana Singh, Kuldeep Singh Yadav, Arun Kumar Tripathi*, Sudeep Tandon and Narayan Prasad Yadav**

Botany and Pharmacognosy Department, CSIR-Central Institute of Medicinal and Aromatic Plants, Lucknow-226015, U.P., India
**Plant Pathology Department, CSIR-Central Institute of Medicinal and Aromatic Plants, Lucknow-226015, U.P., India*
***Department of Process Chemistry and Chemical Engineering, CSIR-Central Institute of Medicinal and Aromatic Plants (CIMAP), Lucknow-226015,U.P., India*

Received November 20, 2016: Revised November 29, 2016: Accepted November 30, 2016: Published online December 30, 2016

Abstract

A considerable research effort has been focused on developing effective insecticide from natural origin. The essential oils and its major chemical constituents have great potential to explore as new insecticide due to insect resistance against synthetic insecticide. In this study, the fumigant toxicity assay of selected essential oils was performed against adults of *Callosobruchus maculates* (Chickpea weevil/pulse beetle) and *Tribolium castaneum* (Red flour beetle). Results showed that the *Mentha spicata* var. viridis oil (MVO) was the most potent fumigant against both the pests namely *C. maculates* (100% average mortality at 1% oil concentration) and *T. castaneum* (100% average mortality at 4% oil concentration) among selected essential oils, while *Cymbopogon citratus*, *Cymbopogon nardus* and *Eucalyptus hybrid* exhibited 100% average mortality at 6% oil concentration against *C. maculates* only. Conclusively, MVO had been identified as active lead from the present study and will be further evaluated for its active phytoconstituent(s).

Key words: Essential oil, fumigant toxicity, *Callosobruchus maculates, Tribolium castaneum*

1. Introduction

Food grains are one of the most essential and vital components of Human survival. It has been estimated that approximately 1/4 to 1/3 of the world's grain crop is lost each year during storage and much of this is due to insect attack. Although stored grains can be destroyed by insects, fungi, vertebrate pests, *etc*. Among the most serious economic pests of grains, *C. maculates* (Coleoptera: Chrysomelidae: Bruchidae) and *T. castaneum* (Coleoptera: Tenebrionidae) are important because both larvae and adult attack the whole grains and flour and cause extensive damage. Direct-feeding damage by insects reduces grain weight, nutritional value, and promote germination of stored grain. Infestations also cause contamination, odor, mold growth that reduce the quality of the grain and may make it unfit for processing into food. Unfortunately, to protect the stored grains from insect damage, very limited options of insecticide like malathion and pesticides, *viz*., aluminum phosphide, ethylene-di-bromide and magnesium phosphide are existing in the Indian market. However, significant increasing trend of resistance in stored grain pests like pulse beetle, red flour beetle has been recorded against the above mentioned pesticides (Zettler and Cuperus, 1990; Weaver *et al*., 1991).

The protection of stored agricultural products using plant materials is an age-old practice in India. Keeping in view the hazardous nature of synthetic pesticides, plants and plant materials are being popular for management of stored grains as they are biodegradable and have least mammalian toxicity (Keita *et al*., 2001; Srivastava *et al*., 2001; Tapondjou *et al*., 2005). The essential oils as pest control agents have two positive things in their favor: the first, is that their natural origin makes them safer for people and the environment, and the second, is that they are considered low risk for resistance development by stored product insects. It is believed that it is difficult for pests to develop resistance to a mixture of components with apparently, different mechanisms of pesticidal activity (Derbalah and Ahmed, 2011). The essential oils and their components have been shown to possess potential to be developed as new fumigants and they may have the advantage over conventional fumigants in terms of rapid degradation and local availability (Isman, 2000). Fumigation continues to play a valuable role in many pest control operations. However, both the concepts and the procedures for controlling insects and other organisms are changing like contact toxicity, ovicidal and repellency but fumigants have unique properties and capabilities that permit use in numerous situations where other forms of control are not feasible.

Now-a-days, management of stored product pests, using substances of natural origin is current topic of exploration (Derbalah *et al*., 2011). The use of essential oils may lead to the identification of new bio-insecticides for the benefit of tropical agriculture. Therefore, the present study was designed to test the fumigant toxicity assay of some selected essential oils against *C. maculates* and *T. castaneum*.

2. Materials and Methods

2.1 Essential oils

The essential oils listed in Table 1 were collected from Department of Process Chemistry and Chemical Engineering, CSIR-CIMAP, Lucknow and used as such.

Author for correspondence: Dr. Narayan Prasad Yadav
Scientist, Botany and Pharmacognosy Department, CSIR-Central Institute of Medicinal and Aromatic Plants (CIMAP), Lucknow-226015, Uttar Pradesh, India
E-mail: npyadav@gmail.com; np.yadav@cimap.res.in
Tel.: +91-945124418; Fax No.:+91-522-2716141

DOI: 10.21276/ap.2016.5.2.10

Table 1: Essential oils tested for fumigant toxicity against two stored grain pests, *C. maculatus* and *T. castaneum*

Botanical source (common name)	Family	Major chemical constituents	References
Apium graveolens (Celery)	Apiaceae	4-chloro-4,4-dimethyl-3- (1-imidazolyl) -valerophenone, 1-dodecanol, 9-octadecen -12-ynoic acid	Nagella *et al*., 2012
Cymbopogon citratus (Lemon grass)	Poaceae	Geranial, Neral, Geraniol, Limonene	Mirghani *et al*., 2012; Jayaratne *et al*., 2001
Cymbopogon nardus (Citronella grass)	Poaceae	Citral, Geraniol	Ganjewala, 2009; Maia and Moore, 2011
Eucalyptus hybrid (Mysore gum tree)	Myrtaceae	1,8-cineole,α-pinene.	Theagarajan and Rao, 1970
Geranium columbinum (Geranium)	Geriniaceae	Hexanal, 3-Ethyl-4-methyl-1-pentanol	Radulovic *et al*., 2011
Mentha spicata var. viridis (Ganga oil)	Lamiaceae	Piperitinone oxide, Carvone, Limonine	Khanuja *et al*., 2006
Pogostemon cablin (Patchouli)	Lamiacea	α-patchoulene, α-guaiene	Gokulakrishnan *et al*., 2013
Tagetes minuta (Southern marigold)	Asteraceae	Cis-ocimene, Trans-ocimene, Dihydrotagetone	Weaver *et al*., 1994

2.2 Insect rearing

Parent adults of chickpea beetle (*C. maculates*) and red flour beetle (*T. castaneum*) were obtained from laboratory stock cultures. The chickpea beetles were reared in 5 liters glass jars on chickpea and red flour beetles were reared in 5 liters glass jarson whole wheat flour in controlled climatic chambers at 25 ± 2°C, 50 ± 10% RH and equal light and dark cycle.

2.3 Fumigant toxicity assay

For essential oil treatment, isolated adults of *T. castaneum* and *C. maculatus* were divided into different groups, separately. Each group consisted of 3 replicates, 5 adults per replicate. One group was used as a control; other groups were used for treatment with different concentrations, *viz*., 1%, 2%, 4%, 6% and 8% of essential oils.

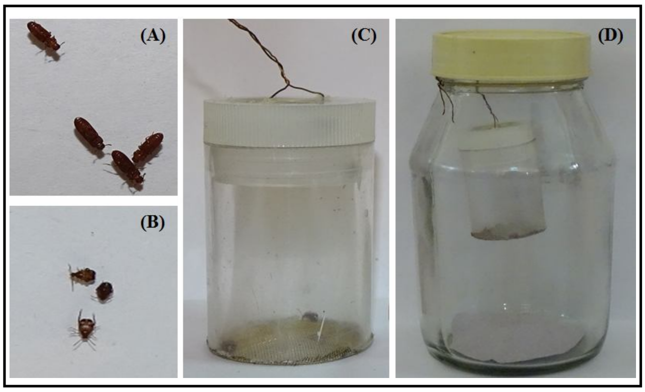

Figure 1: Experimental set up for evaluation of fumigant toxicity assay for stored grain pests. Photographs of (A) adults of *T. castaneum*; (B) adults of *C. maculatus*; (C) plastic vial to hold pests; and (D) assembled apparatus

The five adults were placed in a small plastic vial fitted with 40 mm copper wire net at both ends and suspended in center of an exposure chamber. The air tight Mason jar of 300cc with plastic screw cap was used as exposure chamber (Figure 1). A piece (diameter: 7 cm) of Whatman No. 1 filter paper was exposed with essential oil appropriately diluted in acetone using Eppendorf pipette tip and after 2 min of air drying, it was kept at bottom of exposure chamber to serve as an oil diffuser. The lid of Mason jar was sealed with parafilm. The doses were calculated based on nominal concentrations and assumed 100% volatilization of the oils in the exposure chamber. The whole assembly was held at 50 ± 10% RH with an equal photoperiod of 12:12 h (Light:Dark). Filter paper strips treated with acetone only were treated as control. Mortality was observed after 24 h and graphs were drawn using Graph Pad PRISM® software. Mortality was confirmed by observing any sign of insect movement in response to poking with a needle (Abbasipour *et al*., 2011).

3. Results and Discussion

The essential oil contains several constituents out of which few major are responsible for toxic action against stored grain insects (Agarwal *et al*., 2001). In recent years, a number of plant products have been documented for various bioactivities against stored grain insects. More current research showed that essential oils and their constituents have potential as alternative against currently used fumigants. Major constituents from aromatic plants, mainly monoterpenes, are of special interest because of their potent biological activities in addition to their toxicity to insects (Isman, 2000; Lee *et al*., 2001).

The results of fumigant toxicity assay against *C. maculates* are presented in Figure 2. Local controls were conducted for all the treatments, separately. At 1% oil concentration, *A. graveolens* showed 13.33% average mortality; *C. citratus*, 66.67%; *C. nardus*, 73.33%; *E. hybrid*, 66.67%; *G. columbinum*, 53.33%; MVO, 100.00%; *P. cablin*, 6.67% and *T. minuta*, 53.33% average mortality, respectively. At 2% oil concentration, average mortality was noticed as *A. graveolens*, 20.00%; *C. citratus*, 80.00%; *C. nardus*, 73.33%; *E. hybrid*, 73.33%; *G. columbinum*, 60.00%; MVO, 100.00%; *P. cablin*, 6.67% and *T. minuta*, 66.67%, respectively. At 4% dose, average mortality was recorded for *A. graveolens*, 13.33%; *C. citratus*, 80.00%; *C. nardus*, 80.00%; *E. hybrid*, 93.33%; *G. columbinum*, 66.67%; MVO, 100.00%; *P. cablin*, 6.67% and *T. minuta*, 73.33%, respectively. At 6% and 8% dose, average mortality was found to be 20.00%, *A. graveolens*; 100.00%, *C. citratus, C. nardus E. hybrid*and MVO; 66.67%, *G. columbinum*; 20.00%, *P. cablin* and 86.67%, *T. minuta*, respectively. After summarizing the results, it was observed that there is no change in average mortality even on higher dose beyond 6%. Furthermore, *Mentha spicata* var. viridis oil was found to be most effective against *C. maculates* in fumigant toxicity assay.

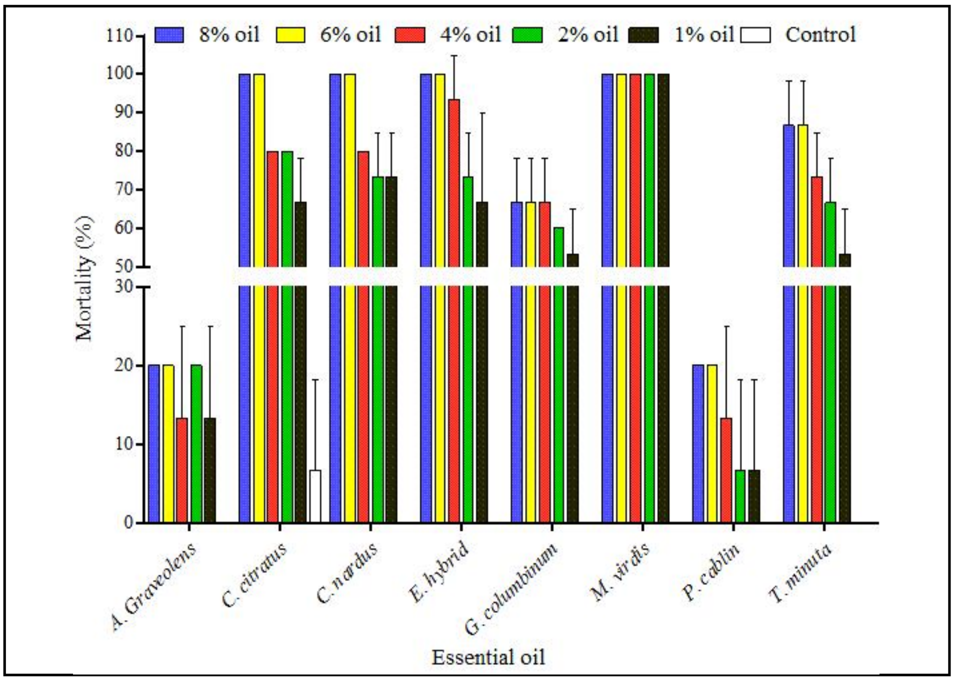

Figure 2: Per cent mortality in adults of *C. maculatus* exposed to selected essential oils at different concentrations.

The percent mortality of *T. castaneum* against selected essential oils is shown in Figure 3. At 1% oil concentration, the average mortality perceived 13.33% and 26.67% after exposure with *C. citratus and* MVO, respectively. *A. graveolens, C. nardus, E. hybrid, G. columbinum, P. cablin* and *T. minuta* found to be ineffective against *T. castaneum* at this concentration. At 2% oil concentration, *A. graveolens*, 6.67%; *C. citratus*, 20.00% and MVO, 86.67% were found to be active against particular pest. All other tested essential oils were found ineffective at particular dose. After increasing the exposure dose of selected essential oils, *i.e.*, up to 4%, the average mortality was increased *A. graveolens*, 13.33%; *C. citratus*, 26.67% and MVO, 100.00%, respectively, but other essential oils remained unproductive at this concentration also. Moreover, the gradual increment in dose up to 8%, the percent mortality of *T. castaneum* adults was unresponsive proportionately as on 6% oil concentration *A. graveolens*, 26.67%; *C. citratus*, 26.67%; *C. nardus*, 20.00% and MVO, 100.00%, respectively and on 8% oil concentration *A. graveolens*, 20.00%; *C. citratus*, 26.67%; *C. nardus*, 20.00%; *E. hybrid*, 6.67%; *G. columbinum*, 13.33%; MVO, 100%; *P. cablin*, 20.00% and *T. minuta*, 6.67%, respectively. The inference of this study is that the essential oil of *Mentha spicata* var. viridis is most effective against *T. castaneum* among selected essential oils. The less per cent mortality of *T. castaneum* after exposure to selected essential oils may be attributed to hard and chitinous body surface compared to *C. maculates.* Considerable differences in *T. castaneum* and *C. maculates* adult's mortality due to essential oil fumigation were observed using different concentrations.Varying activity of different essential oils indicated that the pest controlling and regulating factors were not uniformly present in all the selected aromatic plants.

Since, the *Mentha* spp. has historical significance as a medicinal and insecticidal plant in the traditional knowledge system. In the last few decades, many studies have been reported on the insecticidal activity of several *Mentha* spp. (Kumar *et al*., 2011). MVO can be further explored for its insecticidal activity against different insect species. Therefore, MVO may be recommended at farmer level as it is eco-friendly with hardly any mammalian toxicity and also it may be a good alternative to synthetic pesticide.

4. Conclusion

This research work showed that essential oils have potential as alternative compounds against currently used fumigants. It might further limit the use of synthetic pesticides. *Mentha spicata* var. viridis oil can be further explored for various toxicity assays as it has potential to control *T. castaneum* and *C. maculates*in stored grain. Besides this, other oils which have been found active against *C. maculates* can also be used as pesticide for long storage of grains.

Acknowledgments

The authors are thankful to the Director, CSIR-Central Institute of Medicinal and Aromatic Plants (CIMAP), Lucknow for providing infrastructure facilities to carry out this research work. One of the authors, Dr. Vandana Singh is also thankful to Department of Science and Technology (DST), New Delhi for extending financial support under the Women Scientist Scheme (WoS-A) Grant No. SR/WOS-A/LS-483/2013 for this work. CIMAP Communication No. CIMAP/PUB/2016/88.

Conflict of interest

We declare that we have no conflict of interest.

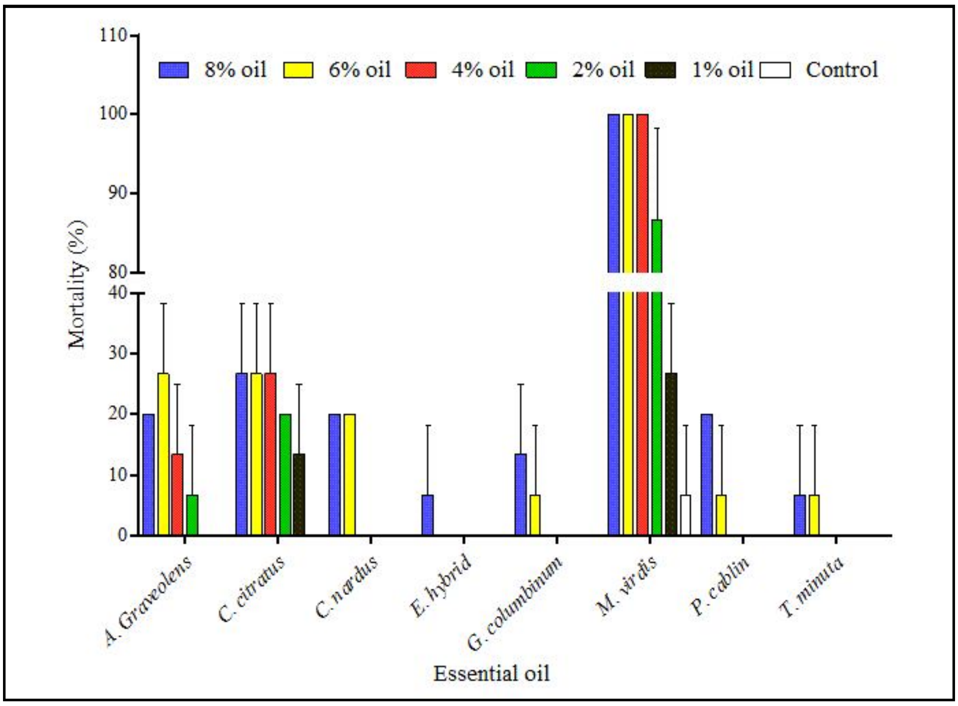

Figure 3: Per cent mortality in adults of *T. castaneum* exposed to selected essential oils at different concentrations.

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