Fumigant toxicity of three plant essential oils against adults of *Ephestia kuehniella* Zeller (Lep.: Pyralidae)

Habib Abbasipour*, Alireza Seyedi, Mohammad Mahmoudvand

Department of Plant Protection, College of Agricultural Sciences, Shahed University, Tehran, Iran

Abstract: Recently, there has been a growing interest in research concerning the possible use of plant extracts as alternatives to synthetic insecticides. Essential oils are among the best-known substances tested against insects. These compounds may act as fumigants, contact insecticides, repellents and antifeedants. The objective of the present study was to test the possible properties of medicinal plants, Ferula gummosa Boiss (Apiaceae), Rosmarinus officinalis L. (Lamiaceae) and Mentha piperita (Lamiaceae) essential oil vapors against Ephestia kuehniella to elucidate their fumigant toxicity. The experiments were conducted at 27 ± 1 °C, $60\pm5\%$ R.H. and in dark condition. The essential oils were obtained from resin of F. gummosa and dried leaves of R. officinalis and M. piperita, and subjected to hydrodistillation using a modified Clevenger-type apparatus. The mortality of adults was tested at different concentrations and different exposure times (1–24h). The results showed that the mortality increased with increases in concentration and exposure time. After 12h, high increases in mortality were seen. Data probit analysis demonstrated that lethal concentration to kill 50% of the population (LC₅₀) for F. gummosa, R. officinalis and M. piperita was estimated as 44.26, 2.15 and 0.97µl/l air, respectively. Between these essential oils, M. piperita was almost more toxic than R. officinalis and F. gummosa. The present study suggests that essential oils from these medicinal plants may be potential grain protectants as botanical alternative fumigants.

Key words: Ferula gummosa, Rosmarinus officinalis, Mentha piperita, Ephestia kuehniella, fumigant toxicity

Introduction

Insect damage in stored food grains may amount to 10-40% in countries where modern storage technologies have not been introduced (Shaaya et al., 1997). Among the stored product pests, Mediterranean flour moth or mill moth (*Ephestia kuehniella*) is a worldwide pest. These caterpillars are serious pests, particularly of stored grain and flour products. They are often found feeding on flour, cereals and dry grain products in food storage areas. They can cause significant problems in machines with their silk webbing. Their close association with human foods makes them prime target for control methods other than chemical pesticides (Lynn and Ferkovich, 2004). Essential oils are among the best known substances tested against insects.

Distillation of aromatic plants yields essential oil, long used in food industries, and more recently for aromatherapy and as herbal medicine (Coppen, 1995; Buckle, 2003). It is well known that a considerable number of plant species, besides their popular use as medicine in many countries, possess insecticidal activities. These plants have been suggested as alternative source of materials for insects control because they contain a range of bioactive chemicals. The practice of using plant derivatives or botanical insecticides as we now know them, in agriculture dates back at least two millennia in ancient China, Egypt, Greece, and India (Thacker, 2002).

^{*} Corresponding author e-mail: habbasipour@yahoo.com

In recent years, essential oils have received a great deal of attention as pest control agents. They are volatile and can function as fumigants, and may also be applicable to the protection of stored products. In many storage systems, fumigants are the most economical and convenient tools for managing stored-grain insect pests, not only because of their ability to kill a broad spectrum of pests but also because of their easy penetration into the commodity while leaving minimal residues (Mueller, 1990).

Rosemary (*Rosmarinus officinalis* L.) is a very important medicinal and aromatic plant, which belongs to the Lamiaceae family and has been cultivated for a long time (Stefanovits-Bányai et al., 2003). Rosemary oil is relatively effective against insect and mite pests. It has been shown that the aromatic vapour of rosemary oil has ovicidal and larvicidal effects on several stored product pests (Tunc et al., 2000; Papachristos and Stampoulos, 2004) as a fumigant. The oil can have sublethal effects as well, for example acting as a repellent to onion thrips, *Thrips tabaci* Lind. (Koschier and Sedy, 2003).

Peppermint (*M. piperita*) oil is one of the most popular and widely used essential oils, mostly because of its main components menthol and menthone (Gul, 1994). Amer and Mehlhorn (2006) studied larvicidal effect of peppermint against *Aedes*, *Anopheles*, and *Culex* larvae (Diptera, Culicidae).

Ferula gummosa Boiss (Apiaceae) is a perennial plant native to Iran and Afghanistan. Like many plants, this species contains active biological compounds including monoterpenes, alfa pinene and alkaloids which have been demonstrated in phytochemical analysis of different extract of this species.

The aim of the present work was the evaluation of these three plant essential oil on flour moth, *Ephestia kuehniella* as alternatives to synthetic insecticides.

Material and methods

Insect rearing

Ephestia kuehniella reared in plastic containers (20cm length \times 14cm width \times 8cm height) containing wheat flour mixed with yeast (10:1, w/w). The culture was maintained in darkness in a growth chamber set at 27 \pm 1°C and 60 \pm 5% RH. All experimental procedures were conducted under conditions identical to those of the culture.

Plant materials

Resins of *F. gummosa* were collected in September 2008 from Kordestan, Iran, hydrodistilled to extract their essential oils. Leaves of *Rosmarinus officinalis, Mentha piperita* were collected on October 2008 from Tehran, Iran. The leaves of *Rosmarinus officinalis, Mentha piperita* were dried naturally on laboratory benches at room temperature (23-27°C) for 6 days until it was crisp. The leaves were hydrodistilled to extract their essential oils.

Extraction of essential oils

Essential oils were extracted from the plant samples by hydrodistillation using a modified Clevenger type-apparatus. The dry plant material including 20g of resins of *F. gummosa* or 50g of leaves *R. officinalis* and *M. piperita* was put in a round bottom flask and 600ml distilled water was added to cover the material. The flask was heated in a heater mantel at a temperature around 100°C so that vapors came up that condensed in the cooling water. Volatile oil assembled in the reservoir was collected after the 4h distillation process.

Fumigant toxicity

To determine the fumigant toxicity of three essential oils on adult insects, filter papers (Whatman No. 1, cut into 2cm diameter pieces) were impregnated with oil at doses calculated to give equivalent fumigant concentrations. Then the impregnated filter paper was attached to the under-surface of the screw cap of a glass vial. The caps were screwed tightly on the vial containing ten adults (1-7 days old). Each concentration and control was replicated five times. Mortality was determined after 3, 6, 9, 12 and 24h from commencement of exposure. Percentage insect mortality was calculated using the Abbott correction formula for natural mortality in untreated controls (Abbott, 1925).

To assess 50 and 95% lethal concentrations of adult insects, different dilutions were prepared to evaluate mortality of insects after a preliminary dose setting experiment. Control insects were kept under the same conditions without any essential oil. Each dose was replicated five times. The number of dead and live insects in each bottle was counted 24h after initial exposure to the essential oil. The dead insects were monitored for at least 48h after recording the data and no affected insects recovered.

Results and discussion

These experiments were conducted in order to determine whether the insecticidal activity of these three essential oil were attributable to fumigant activity. In all cases, considerable differences in insect mortality were noted with different concentrations and exposure times. An increase in susceptibility of the moths was observed as concentrations of the oils were increased.

The highest concentration of the *M. piperita* (3.32µl/l air) and *R. officinalis* (5.81µl/l air) oil proved able to induce more than 50% mortality after 9h, but *F. gummosa* (62.5µl/l air) achieved a level of 50% at 12h after treatment. At the highest concentration of all these essential oil, nearly the complete mortality of moths was achieved after 24h of exposure. In this experiment the slope of the mortality curve was very steep from 9 h to 24h and before this time, the slope leveled off (Fig. 1). Data probit analysis demonstrated that lethal concentration to kill 50% of the population (LC₅₀) for *F. gummosa*, *R. officinalis* and *M. piperita* was estimated as 44.26, 2.15 and 0.97µl/l air, respectively (Table 1). Between these essential oils, *M. piperita* was almost more toxic than *R. officinalis* and *F. gummosa*. Findings of this study showed that essential oils of *F. gummosa*, *R. officinalis* and *M. piperita* have potent fumigant toxicity against adults of *E. kuehniella*. From the three essential oils screened, *M. piperita* showed the highest activity against the adults of *E. kuehniella*.

Results of this study and earlier studies indicate that some plant extracts and essential oils might be useful for managing insects in enclosed spaces because of their fumigant action. Commercial success with these products based on well-known chemistry will likely provide an impetus for the development and commercialization of future pesticides based on essential oils with even greater potency (Isman, 2000). Furthermore, studies have shown that essential oils are readily biodegradable and less detrimental to non-target organisms than conventional chemical pesticides (Tunc et al., 2000). However, the possibility of employing this natural fumigant in the management of Mediterranean meal moth is plausible, but is worthy of further investigation. Future research should focus on residues on target commodity and the influence of any residues on product acceptability.

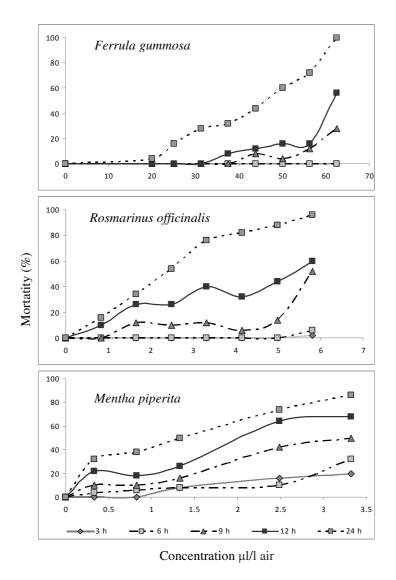


Figure 1. Mortality (%) of *E. kuehniella* exposed to essential oils from *F. gummosa*, *R. officinalis* and *M. piperita* for various concentrations.

Table 1. LC_{50} and LC_{95} values of F. gummosa, R. officinalis and M. piperita essential oils on adults of E. kuehniella.

Plant Species	$LC_{50}^{a,b}$	LC ₉₅ a,b	Slope±SE	Chi	df	P-value
				square		
				(x2)		
Ferula gummosa	44.26(39.98-50.57)	99.01(77.63-155.21)	1.70±0.75	1.39	7	0.98
Rosmarinus officinalis	2.15(1.89-2.39)	6.56(5.52-8.32)	3.39±0.32	1.72	5	0.88
Mentha piperita	0.97(0.72-1.23)	12.51(7.07-35.75)	1.48±0.23	5.87	4	0.20

^a Units LC₅₀ and LC₉₅= μ l/l air, applied for 24 h at 27°C.

^b 95% lower and upper fiducial limits are shown in parenthesis.

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