Ovicidal activity of essential oils from five plants against two stored-product insects

İ. Tunc a,*, B.M. Berger b, F. Erler a, F. Daglı a

a Akdeniz University, Faculty of Agriculture, Plant Protection Department, PK 510, Antalya, Turkey
b Georg-August University, Institute for Plant Pathology and Plant Protection, Göttingen, Germany

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Abstract

The fumigant activity of essential oil vapours distilled from anise Pimpinella anisum, cumin Cuminum cyminum, eucalyptus Eucalyptus camaldulensis, oregano Origanum syriacum var. bevanii and rosemary Rosmarinus officinalis were tested against eggs of two stored-product insects, the confused flour beetle, Tribolium confusum, and the Mediterranean flour moth, Ephestia kuehniella. The exposure to vapours of essential oils from anise and cumin resulted in 100% mortality of the eggs. Oregano achieved mortalities as high as 77 and 89% in T. confusum and E. kuehniella, respectively. The highest mortalities caused by essential oils of eucalyptus and rosemary were 45 and 65%, respectively. At a concentration of 98.5 μl anise essential oil/l air, the LT99 values were 60.9 and 253.0 h for E. kuehniella and T. confusum, respectively. For the same concentration of the essential oil of cumin, the LT99 value for E. kuehniella was 127.0 h. As the essential oils from other plants investigated were less active their estimated LT99 values were too far beyond the tested exposure range to be reliable. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Stored-product insects; Essential oils; Ovicidal activity; Tribolium; Ephestia; Anise; Cumin

1. Introduction

In the search for alternatives to conventional fumigants, essential oils extracted from aromatic plants have been widely investigated. Their toxicity to stored-product insects has been of special interest during the last decade. Most of these studies assessed fumigant activity of

* Corresponding author. Fax: +90-242-227-45-64.
these compounds on adults and to a lesser extent larvae. Very little consideration has been
given to the egg stage and only recently have authors described contact and/or fumigant
toxicity against eggs of stored-product insects of essential oils or their major components
(Shaaya et al., 1993; Ho et al., 1997; Huang et al., 1997; Obeng-Ofori et al., 1997; Obeng-Ofori
and Reichmuth, 1997). As the essential oils are intended to be used like fumigants to disinfest
commodities, they should have the ability to kill all stages of insects. In this context, eggs and
pupae are of particular concern, because they may exhibit a higher tolerance to chemical
agents (e.g. phosphine) (Bell, 1978) than active stages.

Therefore, in order to better understand the response of the egg stage to essential oils — and
especially their vapours — or their constituents, there is a need for systematic investigations of
the effect of different essential oil concentrations and exposure periods on stored-product
insects.

The present study was undertaken to investigate the effect of vapours of essential oils of five
aromatic plant species that are grown in Turkey against the eggs of two important stored-
product insects, the confused flour beetle, *Tribolium confusum* du Val and the Mediterranean
flour moth, *Ephestia kuehniella* Zeller.

### 2. Materials and methods

*T. confusum* was reared on a mixture of wheat flour, bran and yeast, and *E. kuehniella* on
wheat flour, bran, yeast and glycerol. Eggs, 0–24 h old, of both species were used in the tests.
Insect rearing and all experimental procedures were carried out at 26 ± 1°C and 65 ± 5% r.h.

Seeds of anise *Pimpinella anisum* L. and cumin *Cuminum cyminum* L., dried fruits of
eucalyptus *Eucalyptus camaldulensis* Dehn, and dried foliage of oregano *Origanum syriacum* L.
var. *bevanii* (Holmes) Letswaart and rosemary *Rosmarinus officinalis* L., were used for
extraction of essential oils by water steam distillation as described by Sarac and Tunç (1995)
and Tunç and Şahinkaya (1998). For details of the operating conditions of the gas
chromatograph to identify and quantify the constituents of rosemary, refer to Müller-Riebau et
al. (1995).

The eggs were exposed to essential oils on cloning plates (Nunc, Denmark) modified for this
purpose. A set of cloning plates consisted of a bottom plate (5.6 × 8.2 cm) with 60 microwells
of 4 mm dia. and 2 mm depth and a cover plate (5.9 × 8.4 cm). Holes of 3 mm dia. were
drilled over each microwell in the cover plate. A piece of serigraphic cloth was placed between
the two plates to avoid escape of hatched larvae while allowing for air circulation. The
exposure device was described in more detail by Tunç et al. (1997). One egg was
accommodated in each microwell and thus sixty eggs on each plate. Each plate was divided
into three sections each accommodating 20 eggs and making one replicate. One plate was used
for each concentration and exposure time combination. Experiments were repeated twice so
that each concentration × exposure time was represented by 6 units of 20 eggs. The plates were
placed in 650 ml glass jars with screwed lids. Essential oils were applied on blotting paper
(2 × 8 cm) which was attached to the lower side of the lids. Essential oil concentrations of 24.6,
49.2, 98.5 and 196.9 μl/l air, and exposure periods of 24, 48, 72 and 96 h were investigated.
After exposure, plates were taken out of the jars and final mortality counts were made after 11
days for *T. confusum* eggs and after 9 days for *E. kuehniella* eggs with a stereomicroscope. Unhatched eggs were counted as dead. Mortality data were corrected for natural mortality in controls and were subjected to probit analysis to estimate LT$_{50}$ and LT$_{99}$ values (Sokal and Rohlf, 1973).

3. Results

Vapours showed variable toxicity to eggs of *T. confusum* and *E. kuehniella* (Figs. 1 and 2). However, only the essential oil of anise achieved a mortality of 100% against the eggs of both species. The essential oil from cumin had such activity only against the eggs of *E. kuehniella*. The essential oil of oregano achieved 77% and 89% mortalities against *T. confusum* and *E. kuehniella*, respectively, at the highest concentration and exposure period investigated (196.9 µl/l air, 96 h). Rosemary was less active and achieved a mortality of only up to 65% in *T. confusum* and 24% in *E. kuehniella*. The highest mortality caused by eucalyptus oil was 18 and 45% for *T. confusum* and *E. kuehniella*, respectively.

The time required for a 99% kill (LT$_{99}$) was 60.9 and 253.0 h for *E. kuehniella* and *T. confusum*, respectively, at a concentration of 98.5 µl anise/l air (Table 1). A LT$_{99}$ of 127.0 h was estimated for *E. kuehniella* at the same concentration of cumin. However the estimated LT$_{99}$ values of cumin for *T. confusum* and for the other essential oils for both species were too far beyond the tested ranges to be reliable, giving results in thousands of hours. The only exception was oregano which had a LT$_{99}$ value of 327.0 h for *E. kuehniella* at 196.9 µl/l air. The toxicity of essential oils to the eggs in descending order was anise, cumin, oregano, rosemary and eucalyptus.

The main ingredients of the essential oil of rosemary, as quantified with gas

<table>
<thead>
<tr>
<th>Doses (µl/l air)</th>
<th>Anise</th>
<th>Cumin</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>LT$_{50}$</td>
<td>LT$_{99}$</td>
</tr>
<tr>
<td><em>T. confusum</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td>24.6</td>
<td>59.8</td>
<td>424.0</td>
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<tr>
<td>49.2</td>
<td>38.0</td>
<td>630.2</td>
</tr>
<tr>
<td>98.5</td>
<td>8.3</td>
<td>253.0</td>
</tr>
<tr>
<td>196.9</td>
<td>**</td>
<td>**</td>
</tr>
<tr>
<td><em>E. kuehniella</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td>24.6</td>
<td>60.1</td>
<td>429.1</td>
</tr>
<tr>
<td>49.2</td>
<td>34.2</td>
<td>191.9</td>
</tr>
<tr>
<td>98.5</td>
<td>12.1</td>
<td>60.9</td>
</tr>
<tr>
<td>196.9</td>
<td>8.0</td>
<td>43.1</td>
</tr>
</tbody>
</table>

* Estimated LT$_{50}$ or LT$_{99}$ values were too far beyond the tested exposure range to be reliable.

** It was not possible to estimate LT$_{50}$ or LT$_{99}$ values due to 100% mortality at all exposure periods used.
Fig. 1. Percent mortality in the eggs of *T. confusum* exposed to five essential oils at different concentrations and exposure times.
Fig. 2. Percent mortality in the eggs of *E. kuehniella* exposed to five essential oils at different concentrations and exposure times.
chamomile, were camphor (41.9 mg/ml), borneol (28.6 mg/ml), 1,8 cineole (26.1 mg/ml), linalool (10.8 mg/ml), menthol (3.9 mg/ml) and pulegone (2.7 mg/ml). A further compound, however, could not be identified with the available standards and analytical procedure. The main ingredients of the essential oils from the other plants have been described earlier (Saraç and Tunc, 1995; Tunc and Şahinkaya, 1998).

4. Discussion

The essential oils investigated in this study are used as pharmaceuticals and in flavouring and are therefore considered less harmful to humans than most conventional insecticides. Furthermore, studies have shown that they are readily biodegradable (Baysal, 1997) and less detrimental to non-target organisms than pesticides (Yeğen et al., 1998). Despite these advantages, the potential for pest control or crop protection with these essential oils has not been investigated until recently. However, the first successful trials on the control of insects (Saraç and Tunc, 1995) and fungal diseases (Yeğen et al., 1992; 1998) with volatile essential oils have been published and encourage studies on their practical use against stored-product insects.

Comparison of the results presented here with earlier investigations (Saraç and Tunc, 1995) demonstrates that responses of the egg stage and active stages of stored-product insects to essential oils are different. Eggs of T. confusum were more tolerant towards anise oil than the adults. However, the eggs of E. kuehniella were less tolerant towards the essential oil than the last larval instars at comparable concentrations and exposure periods (Saraç and Tunc 1995).

Compared with the investigation of Shaaya et al. (1993), our results demonstrate a much lower toxicity of the essential oil of oregano against eggs of T. confusum and E. kuehniella. The species of oregano tested here may have differed from that of Shaaya et al., or seasonal changes in the chemical composition of the essential oil may have been encountered, as reported by Müller-Riebau et al. (1996, 1997).

Information on the distribution characteristics of the essential oils after application to a commodity, e.g. absorption and desorption, is scarce, but there are indications from the studies of Shaaya et al. (1997) that some absorption by the commodity investigated (wheat) took place. Higher concentrations and longer exposure periods were needed to achieve a similar level of mortality for a given species than required when the oils were applied in a space fumigation.

Aromatic plants contain, in general, essential oils at concentrations of 1–3% (Çakır, 1992). Therefore, large quantities of plant material have to be processed in order to obtain essential oils in quantities sufficient for commercial scale tests. However some constituents may exhibit a much higher activity than the whole essential oil. This should encourage the breeding of plant varieties that produce such compounds in elevated amounts. Synthetic production of these compounds may also be an option in order to gain enough material for practical use as plant protection products.

Our results and those reported earlier clearly indicate variations in the activity of essential oils regarding the stage of the insect, the species of the insect and the plant origin of the essential oil. Not all the essential oils tested showed satisfactory activity, but the essential oils
of anise, and also cumin, proved to be promising as control agents against stored-product insects, especially *E. kuehniella*.

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**References**


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