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FUMIGANT TOXICITY OF *CARUM COPTICUM* ESSENTIAL OIL AGAINST *SITOPHILUS GRANARIUS*

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ABSTRACT

Laboratory bioassays were conducted to assess fumigant toxicity of *Carum copticum* (L.) essential oil against *Sitophilus granarius* (L.). Essential oil was extracted from *C. copticum* seeds by hydrodistillation using a modified Clevenger type apparatus. The concentrations of 6, 9, 12, 15 and 18 µL L^{-1} air were applied for the bioassays. Adults were exposed to the concentrations impregnated to the filter papers, placed into the under surface of the screw lids of glass vials. The vials were kept in incubator set up at 27±1°C and 55±5% r.h. in continuous darkness. The mortality was counted after 3, 6, 10, 24, and 48 h of exposure. The mode of action of essential oil was characterized by insect's knockdown, hyperactivity, convulsion, paralysis; and ultimately insect death. Mortality increased with increasing concentration levels and time exposing to the oil. However the insect mortality did not exceed 89% even after 48 h exposure to the highest concentration (18 µL L^{-1} air). The present study suggests that *C. copticum* essential oil may have the potential to apply as grain protectants against stored-products insect pests. However, further investigations are necessary to confirm these findings.

Key words: Essential oil, fumigant toxicity, *Sitophilus granarius*, *Carum copticum*, stored-products protection

INTRODUCTION

The granary weevil, *Sitophilus granarius* (L.), is a serious insect pest of various food grains under storage. This species has global distribution; usually found in grain storage facilities and damage harvested grains that are being stored. The larvae feed inside the grain until pupation, after which they bore a hole out of the grain and emerge (Rees, 1996). With regards to the importance of stored products, protection of them from insect-pests infestations should be considered. Natural pesticides based on plant-essential oils may be one of the most promising alternatives to synthetic insecticides and fumigants; because they are of natural origin and they don't have hazards to human and environment (Isman, 2000).

Carum copticum (L.) (Apiaceae) is a medicinal plant grown in Iran, Pakistan and Egypt. The plant bears white flowers and small brownish fruits. It is used as household medicine, and the oil is used as pharmaceutical applications (Sahaf et al., 2007). The insecticidal efficacy of *C. copticum* essential oil has been investigated against *Callosobruchus maculatus* (F.), *Sitophilus oryzae* (L.), *Tribolium castaneum* (Herbst) and *Plodia interpunctella* (Hubner) (Sahaf and Moharramipour, 2008; Sahaf et al., 2007; Shojaaddini et al., 2008). However, fumigant toxicity of *C. copticum* oil has not been assessed against *S. granarius*.

The aim of the present study was to investigate fumigant toxicity of *C. copticum* essential oil against adults of *S. granarius*.

MATERIALS AND METHODS

Rearing of tested insects

Sitophilus granarius was obtained from the cultures maintained in the Entomology Laboratory-Tarbiat Modares University for at least 3 years with no history of exposure to insecticides. Adults were reared on wheat (variety Pishtaz Madary, m.c. ≈ 12%) at 27±1°C and 65±5% r.h. in continuous darkness.

Collection and preparation of essential oil

Carum copticum seeds were purchased from a research farm in Ferdowsi University-Mashhad-Iran. Seeds were packed in bags and kept in the refrigerator at 4°C. Oil extraction was performed using a Clevenger type apparatus. About 40 g of seeds were ground and put into the Round-bottom flask over water at a temperature around 100°C. As the water was heated the steam passed through the plant material, vaporizing the volatile compounds. Volatile oil assembled in the reservoir was collected after the 4 h distillation process. Subsequently, anhydrous sodium sulphate was applied to remove water. Essential oil was stored in self standing screw cap microtubes covered with foil at 4°C until beginning of the experiment.

Bioassay test for fumigant toxicity

Fumigant toxicity of essential oil was assessed against adults of *S. granarius*. Glass vials with the volume of 280 ml were used for the experiments and 25 adults were introduced into each vial. Filter papers (Whatman No. 1) were placed into the under surface of the screw lids of glass vials and impregnated with different concentrations of essential oil. The concentrations of 6, 9, 12, 15 and 18 μL L^{-1} air were applied for the experiment. Each concentration and control was replicated four times. All vials were placed in incubators set at 27±1°C and 55±5% r.h. in continuous darkness. The adult mortality was counted after 3, 6, 10, 24, and 48 h of exposure.

Statistical analysis

There were no mortality in control groups; so, there was no need to correct the mortality data. Mortality percentages were transformed to square root of arcsine to normalize the data. The data were subjected to one-way analysis of variances to determine significant differences between exposure time and concentration levels (SPSS, 2007). Data was subjected to Probit analysis (Finney, 1971) to estimate Lethal $Concentration_{50}$ (LC_{50}) and Lethal $Time_{50}$ (LT_{50})values and their 95% confidence limits using SAS 6.12 software (SAS Institute, 1997).

RESULTS

Percentage mortalities of *S. granarius* adults exposed to different concentrations of *C. copticum* essential oil are presented in Fig.1.

No mortality was observed 3 h after exposing to different concentrations of the oil. However, the mortality increased with increasing the exposure time. Based on LC_{50} values, 46.37 and 10.85 μL L^{-1} air of *C. copticum* essential oil was required to obtain 50% mortality of *S. granarius* adults after 24 and 48 h of exposure, respectively (Table 1).

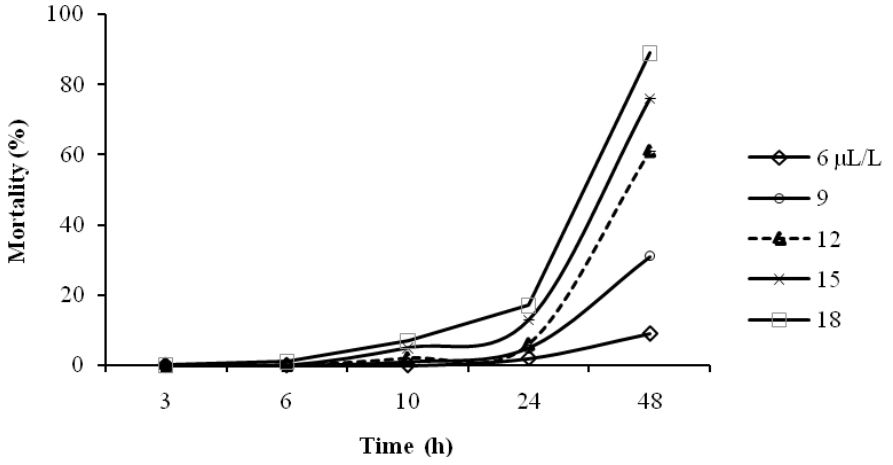

Fig. 1- Percentage mortality (%) ± SE of *Sitophilus granarius* adults exposed to different concentrations of *Carum copticum* essential oil for 3, 6, 10, 24, and 48 h.

Table 1. Lethal Concentration$_{50}$ (LC_{50}) values for *Carum copticum* essential oil against *Sitophilus granarius* adults resulting from 24 and 48 h laboratory fumigation

Time (h)	LC_{50} (µL/L air)	95% Cl (µL L^{-1}air)		Slope	d.f.	χ^2	*P* value
		Lower	Upper				
24	46.37	30.06	156.9	2.37	3	0.790	0.852
48	10.85	10.26	11.45	5.38	3	0.899	0.586

CI: Confidence intervals

LT_{50} estimates indicated no significant differences (no overlap in 95% confidence limits) between concentrations of 15 and 18 µL L^{-1} air (Table 2).

Table 2. Lethal time$_{50}$ (LT_{50}) values of *Sitophilus granarius* adults exposed to 15 and 18 (µL L^{-1}) concentration of *Carum copticum* essential oil

Concentration (µL L^{-1})	LT_{50} (h)	95% Cl (h)		Slope	d.f.	χ^2	*P* value
		Lower	Upper				
15	35.74	21.84	111.6	3.96	3	18.6	0.01
18	30.01	14.60	144.6	3.99	3	28.3	0.01

CI: Confidence intervals

DISCUSSION

Observation of *S. granarius* exposed to *C. copticum* essential oil indicated their knockdown, hyperactivity, convulsion, paralysis; and finally insect death. Isman (2006) stated that rapid effect of essential oils is because of their neurotoxic mode of action.

According to the results obtained from present study insects mortality increased with increasing concentration levels and exposure time. This is in agreement with Sahaf and Moharramipour (2008). However, in most cases the efficacy of essential oil decreased over time, which can be attributed to their high volatility and low stability (Mikhaiel, 2011; Ogendo et al., 2008; Rozman et al., 2007; Sahaf et al., 2007). In our study, if the experiment was continued up to 48 h, may be adults mortality declined due to the reduction of essential oil efficacy.

Insect's susceptibility to the same essential oil differs from species to species. According to the Sahaf and Moharramipour (2008), LC_{50} values of *C. copticum* essential oil against adults of *C. maculatus* after 24 h exposure time was 0.90 µL L^{-1} air. In the other study Sahaf et al., (2007) stated that 0.91 µL L^{-1} air of *C. copticum* oil was required to obtain 50% mortality of *S. oryzae*, and 33.14 µL L^{-1} air in the case of *T. castaneum*. Shojaaddini et al., (2008) also estimated 257.83 and 91.36 µL L^{-1} air of *C. copticum* oil to control 50% of *P. interpunctella* adults and larvae, respectively. Based on LC_{50} values, *S. granarius* seems to be more tolerant to *C. copticum* oil than other Coleopteran species.

Most of the plants and herbs are locally available and can be applied for pest control programs in small scales. However, there is need to conduct further studies to assess their insecticidal efficacy against stored-product insect pests.

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