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Bio-insecticide effect of black cumin against Granary weevil, *Sitophilus granarius* L. (Coleoptera: Curculionidae) on stored wheat

[1] M Karakas, [2] E Bolukbasi

[1] Department of Biology, Science Faculty, Ankara University Ankara, Turkey
[2] Department of Biology, Science and Arts Faculty, Amasya University, Amasya, Turkey

Abstract
These investigations aimed to determine bio-insecticide effect of black cumin essential oil against granary weevil, *Sitophilus granarius* L. on stored wheat. Therefore three different experiments with black cumin were carried out: I. To test efficacy of black cumin against different developmental stages of granary weevil in soft wheat grain; II. To test effective concentration of black cumin against granary weevil in space 50% filled up with wheat applying black cumin in the concentration range of 25, 50, 100, 175 and 275 mg/ml; III. To test effective concentration of black cumin in spaces differently filled up with wheat (empty space, 50% and 95% filled up). Though black cumin exhibited certain fumigant characteristics it would be needed to apply high concentrations relative to phosphine or methyl bromide to gain satisfactory results. If the problem of cost-effective commercial production can be solved, black cumin could find a place in IPM strategies, especially where the emphasis is on environmental, food and human safety and on replacing the more dangerous and toxic fumigants and insecticides.

Keywords: *Sitophilus granarius*, granary weevil, bio-insecticides, plant extract, essential oil

1. Introduction

The wheat granary weevil, *Sitophilus granarius* Linnaeus, 1758 (Coleoptera: Curculionidaed) is one of the most serious pests of stored grain, especially in temperate regions (Niewiada *et al*., 2005) [10]. This pest is internal feeder and cause considerable loss to cereals affecting the quantity as well as quality of the grains (Mile and Gyori, 2006; Ebadollahi, 2011) [9, 3].

Synthetic insecticides have been considered the most effective and accessible means to control insect pests of stored products (Huang and Subramanyam, 2005) [6]. These chemicals are associated with undesirable effects on the environment due to their slow biodegradation in the environment and some toxic residues in the products for vertebrates especially for mammalian health (Jolankai *et al*., 2006; Halder *et al*., 2010) [7, 4].

The adverse effects of synthetic pesticides have amplified the need for effective and biodegradable pesticides. Natural products are an excellent alternative to synthetic pesticides as a means to reduce negative impacts to human health and the environment (Thomas *et al*., 2002) [14]. The popularity of botanical pesticides is once again increasing and some plant products are being used globally as green pesticides (Hamza *et al*., 2016) [5]. Among various kinds of natural substances that have received particular attention as natural agents for insect management are essential oils and extracts from aromatic plants. So, the focus over the last few years has been investigations of essential oils bioactivity in order to find out the most fumigant active chemical components (Park *et al*., 2003; Rozman *et al*., 2006; 2007) [11, 12, 13].

This paper describes laboratory research on black cumin essential oil as fumigant against granary weevil to find out whether essential oils can act as potential source of new fumigants, as well as whether they can be produced and used in fumigation in economically justified manner.

2. Materials and Methods

2.1 Insect Culture

The wheat granary weevil, *Sitophilus granarius* L. was reared in a 1 L wide-mounted glass jars containing soft wheat grains. Mouth of the jars covered with a fine mesh cloth for ventilation and to prevent escape of the weevils. Cultures were maintained in an incubator at 27 ± 1 °C and 60 ± 5 % relative humidity (Davis and Bry, 1985) [2]. Insects used in all experiments were 1 to 7 day old adults. All experimental procedures were carried out under the same environmental conditions as the cultures. The wheat granary weevil adults were obtained from the stock culture of the laboratory of the Plant Protection Department, Faculty of Agriculture, Ankara. The life cycle can be completed in as little 30 to 40 days during the culture conditions but takes considerable longer in cooler conditions. Adult granary weevils can live up to eight months and can produce up to four generations per year.

2.2 First Test

Test of fumigant efficacy of black cumin against different developmental stages (eggs, larvae, pupae and adults) of granary weevil.

By sieving infested wheat all developmental stages of the pest outside the grain were removed. Infested wheat was divided into 440g samples placed into glass jars of 1000 ml capacity (50% filled up). Infested wheat samples of 440g in 1000 ml volume glass jars were treated with black cumin by dribbling (25 mg/ml) onto filter paper that was placed on the surface of the grain. The jars were than tightly sealed. Equal number of samples was set up as non-treated control. After 7 days the samples were opened for aeration and assessment of the results. The experiment was set up in three repetitions, and results of alive developed adults were measured three times after the following 7, 14 and 21 days.

2.3 Second test

Test of effective concentration of black cumin against granary weevil adults.

Each sample contained 440g of wheat placed in glass jars of 1000 ml volume with 200 adults 2-4 weeks old. Five concentrations of black cumin (25, 50, 100, 175 and 275 mg/ml) that was dribbled onto filter paper, and non-treated control were tested. The experiment was set up in three repetitions, and number of dead adults was measured after 48 hours.

2.4 Third test

Test of efficacy of black cumin (25 mg/ ml) in empty space, in space 50% and 95% filled up with wheat against granary weevil adults 2-4 weeks old.

Test samples for fumigation of empty space were 1000 ml volume glass jars containing 1g flour, 20 wheat grains and 200 adults. Test samples for fumigation of space 50% filled up containing 450g wheat in 1000 ml volume glass jars with 200 adults, while test samples of space 95% filled up containing 900g wheat in 1000 ml volume glass jars, and 200 adults. Filter paper that was dribbled with black cumin was placed at the bottom of the each sample (empty space) and on the surface of the wheat (space 50% and 95% filled up). Non-treated control was set up in the same manner. The experiment was set up in three repetitions, and the number of dead adults was measured after 48 hours.

Data was subjected to one-way analysis of variance (ANOVA) according to the general linear model and LSD test entered in the table.

3. Results and Discussion

The results of efficacy of black cumin against different developmental stages (Table 1) showed that number of granary weevil emerged adults being fumigated with 25 mg/ml with exposure of 48 hours was slightly lower than their number in the control, apparently applied dose was not sufficient for effective control of younger developmental stages of *S. granarius*. By testing five concentrations of black cumin (25, 50, 100, 175 and 275 mg/ml) during 48 hours in space 50% filled up with wheat, 100% mortality of granary weevil was obtained with highest concentration of 275 mg/ml (Figure 1). Concentration of 25 mg/ml black cumin in empty space induced nearly 100% mortality. However, fumigation in space 50% filled up with wheat showed only 50% to 60% efficacy against granary weevil. In space 95% filled up with wheat mortality was 34% (Figure 2).

In general, black cumin had certain fumigant effect to *S. granarius*. Fumigation of empty space showed favourable results, while fumigation of space filled up with wheat 50% and 95% proved to be ineffective and unacceptable. The cause could be high sorption of black cumin in wheat grains poor permeability of black cumin vapour into seed interspace and into grains, which considerably reduced fumigation effect. In other words, to gain as similar results as obtained with phosphine and methyl-bromide cineole concentrations should range from 200-250 mg/ml. According to Champ and Dyte (1976) [1], phosphine dose of 0.03 mg/ml methyl-bromide dose of 1 mg/ml, if applied in airtight space, were found to be enough to gain LD_{50} test insects, while Lee *et al*., (2004) [8] reported required cinole dose of 42 mg/ml to gain LD_{50} for *Sitophylus oryzae*, roughly equal to my doses.

Table 1: *S. granarius* alive emerged adults after 7 days exposure to black cumin (25 mg/ml) on infested wheat

Treatment	Number of live emerged S. granarius* adults after days					
	7 days		14 days		21 days	
	Mean	SD	Mean	SD	Mean	SD
Control	0.00^a	0.00	56.16^a	10.10	1631.50^a	169.46
Black cumin 8.8 ml	0.00^a	0.00	29.39^b	5.60	1121.10^a	96.11

*Means in the same column followed by the same letters are not significantly (P>0.05) different as determined by the LSD test.

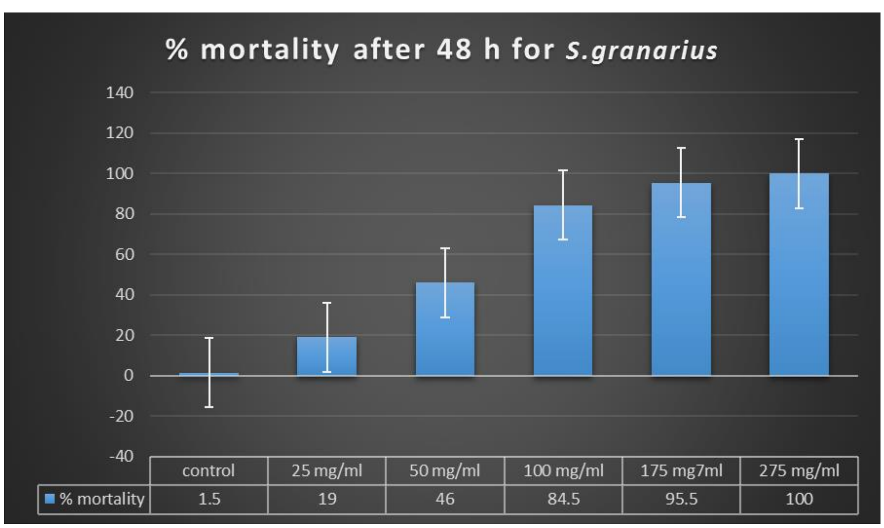

Fig 1: *S. granarius* - adults mortality (%) after 48 hours and application of the range of black cumin concentrations.

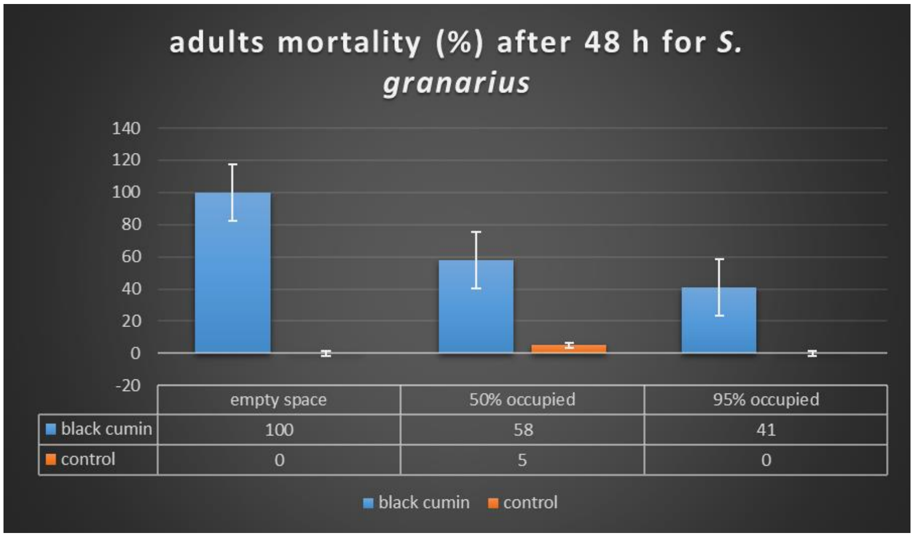

Fig 2: *S. granarius* – adults mortality (%) after 48 hours induced by application 0f 25 mg/ml black cumin

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