BIOACTIVITY OF CINEOLE AGAINST RICE WEEVIL (*SITOPHILUS ORYZAE* L.) ON STORED WHEAT

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Abstract: These investigations aimed to determine bioactivity of cineole essential oil against rice weevil (*Sitophilus oryzae* L.) on stored wheat. Therefore, three different experiments with cineole were carried out: first, to test efficacy of cineole against different developmental stages of rice weevil in wheat grain; second, to test effective concentration of cineole against rice weevil in space 50% filled up with wheat applying cineole in the concentration range of 50, 100, 150, 200, 250 g m^{-3}; and third, to test effective concentration of cineole in spaces differently filled up with wheat (empty space, 50% and 95% filled up). Though cineole exhibited certain fumigant characteristics it would be needed to apply high concentrations (doses) relative to phosphine or methyl bromide to gain satisfactory results. If the problem of cost-effective commercial production can be solved, cineole could find a place in IPM strategies, especially where the emphasis is on environmental and food safety and on replacing the more dangerous and toxic fumigants and insecticides.

Keywords: essential oils, cineole, fumigation, *Sitophilus oryzae*, cost-effectiveness

Introduction

Many conventional pesticides can adversely affect the environment; requirements for safer means of pest management have become crucial. Chemical inputs are seen to be a primary cause of food contamination and environmental pollution arising from agriculture (Jolánkai et al., 2006). Thus, the application of synthetic fumigants in storage facilities to control stored pests has been a standard practice during the past few decades. Their non-selective and uncritical application brings up some serious issues, such as toxic effects to the wheat that is used in diet for people and livestock (Fishwick, 1988), and contamination of the environment (WMO, 1995). During storage, the primary goal is to preserve quality of wheat and to prevent qualitative and quantitative loss (Győrine Mile and Gyori, 2006). Therefore, the use of safe, low toxicity botanical pesticides is now emerging as one of the prime means to protect crops like wheat, their products and the environment from pesticide pollution, as a global problem (Prakash and Rao, 1997). Generally, botanicals should cause less damage to human and environmental health than conventionals. 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 (Singh and Upadhyay, 1993; Lee et al., 2003; Lee et al., 2001; Park et al., 2003; Rozman et al., 2006; 2007; 2007a). This paper describes laboratory research on cineole essential oil as fumigant against rice 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.

Materials and methods

Investigations were carried out in the laboratory of Diatom Research and Consulting Inc. in Canada. 99% cineole essential oil (produced by „Sigma“, Germany) was applied under controlled laboratory conditions: temperature 30±1°C, 70±5% r. h. in darkness. Culture of rice weevil – *Sitophilus oryzae* L. were reared in the Diatom Research in same conditions. All experiments were of Canadian Western Hard red wheat, with 14% m.c. Three laboratory experiments with 56 samples were set up:

1. experiment –test of fumigant efficacy of cineole against different developmental stages (eggs, larvae, pupae and adults) of rice weevil. By sieving infested wheat all developmental stages of the pest outside the grain were removed. Infested wheat was divided in 200 g samples placed into glass jars of 450 ml capacity (50% filled up). Infested wheat samples of 200g in 450 ml^{-1} volume glass jars were treated with cineole by dribbling onto filter paper (50 g m^{-3}) that was placed on the surface of the grain. The jars were then 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 4 repetitions, and results of alive developed adults were measured two times after the following 13 and 21 days.

2. experiment –test of effective concentration of cineole against rice weevil adults. Each sample contained 200g of wheat placed in glass jars of 450 ml^{-1} volume with 100 adults 2-4 weeks old. 5 concentrations of cineole (50, 100, 150, 200, 250 g m^{-3}) that was dribbled onto filter paper, and non-treated control were tested. The experiment was set up in 4 repetitions, and number of dead adults was measured after 48 hours.

3. experiment - test of efficacy of cineole (50 g m^{-3}) in empty space, in space 50%, and 95% filled up with wheat against rice weevil adults 2-4 weeks old. Test samples for fumigation of empty space were 450 ml volume glass jars containing 0,5g flour, 10 wheat grains, and 100 adults. Test samples for fumigation of space 50% filled up contained 200 g wheat in 450 ml^{-1} volume glass jars with 100 adults, while test samples for fumigation of space 95% filled up contained 360 g wheat in 450 ml^{-1} volume glass jars, and 100 adults. Filter paper that was dribbled with cineole (50 g m^{-3}) 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 4 repetitions, and the number of dead adults was measured after 48 hours.

Statistical analysis - data were subjected to one-way analysis of variance (ANOVA) according to the GLM (general linear model) and LSD test entered in the table. Data processing was conducted by the SAS System for Windows 98. The figures that represent mean values were made by Microsoft Excel 2003.

Results and discussion

The results of efficacy of cineole against different developmental stages (Table 1) showed that number of rice weevil emerged adults being fumigated with 50 g m^{-3} 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. oryzae*. By testing 5 concentrations of cineole (50, 100, 150, 200, 250 g m^{-3}) during 48 hours in space 50% filled up with wheat, 100% mortality of rice weevil was obtained with highest concentration of 250 g m^{-3} (Figure 1). Concentration of 50 g m^{-3} cineole in empty space induced nearly 100% mortality. However, fumigation in space 50% filled up with wheat showed only 50% to 60% efficacy against rice weevil. In space 95% filled up with wheat mortality was 34% (Figure 2).

Table 1. S. oryzae - alive emerged adults after 7 days exposure to cineole (50g m^{-3}) on infested wheat

Treatment	Number of live emerged *S. oryzae** adults after days					
	7 days		13 days		21 days	
	Mean	SD	Mean	SD	Mean	SD
Control (non-treated)	0,00[a]	0,00	41,25[a]	8,22	1421,50[a]	166,39
Cineole 50 g m^{-3}	0,00[a]	0,00	23,35[b]	4,92	1069,75[a]	89,19

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

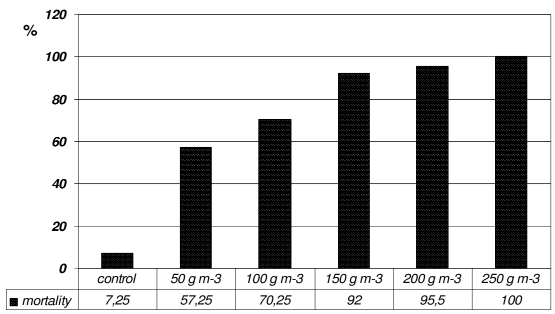

Figure 1. S. oryzae – adults mortality (%) after 48 hours and application of the range of cineole concentrations

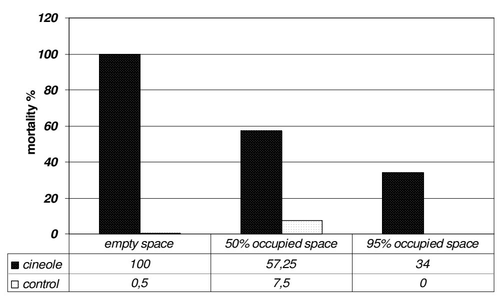

Figure 2. S. oryzae – adults mortality (%) after 48 hours induced by application of 50 g m^{-3} cineole

In general, cineole had certain fumigant effect to *S. oryzae*. 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 cineole in wheat grains, poor permeability of cineole 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 g m^{-3}. According to Champ and Dyte (1976), phosphine dose of 0.03 g m^{-3} and methyl-bromide dose of 1 g m^{-3}, if applied in airtight space, were found to be enough to gain LD_{50} test insects, while Lee et al. (2004) reported required cineole dose of 42 g m^{-3} to gain LD_{50} for *S. oryzae,* roughly equal to our doses. Moreover, one (1) kg of phosphine pellets costs about 28.4 EUR, whilst 1 kg of cineole in packages of 100g reaches 162 EUR.

Conclusions

Though cineole exhibited certain fumigant characteristics against *S. oryzae*, it would be needed to apply high concentrations (doses) relative to phosphine or methyl bromide to gain satisfactory results. We find such a high price for cineole, and for other essential oils, considering the remaining characteristics (scent, sorption, aeration etc.), as serious limiting factor for application of natural essential oils in practice.

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