

PRELIMINARY STUDIES ON EFFICACY OF VAPORIZED ETHYL FORMATE AND CARBON DIOXIDE FORMULATIONS AGAINST STORED-GRAIN INSECTS

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Abstract: Ethyl formate (EF) is a fumigant currently used abroad for stored grain protection against pests, being a potential alternative to methyl bromide. This paper presents the results of the efficacy of fumigant mixture of ethyl formate and carbon dioxide trials on *Tribolium castaneum* (red flour beetle) adults and larvae. The good efficacy of fumigant mixture of ethyl formate and carbon dioxide was demonstrated with ethyl formate 50 mg/l and carbon dioxide 1%; for the adults of *T. castaneum* one hour exposure was enough for the 95% mortality; for the larval stage, 98,5% mortality was obtained after 3hrs of exposure.

Further studies are required to ascertain the true applicability of this fumigant mixture in order to introduce it in stored products protection.

Key words: ethyl formate, carbon dioxide, fumigation, stored grain insects

INTRODUCTION

Ethyl formate (EF) is a fumigant currently used for stored grain protection against pests (Muthu et al., 1984 cited by Damcevski and Annis, 2001; Navarro, 2006; YongLin and Mahon, 2006; Darby et al., 2009) as it is a potential alternative to methyl bromide. Ethyl formate has the advantages of very rapid action and loss from the gaseous phase and breakdown by the grain; this property could make it useful in rapid disinfestation. Because some undesirable properties of EF, such as poor penetration through grain, significant losses to grain sorption, high concentrations of fumigant required to control insects, and flammability risks, it was developed the combination with carbon dioxide that significantly enhanced efficacy of the fumigant against stored grain insects (Haritos et al., 2006; Damcevski et al., 2009; Dojchinov et al., 2010).

The aim of the current work was to investigate the response of *Tribolium castaneum* (Herbst) adults and larvae to exposure of fumigant mixture of ethyl formate and carbon dioxide in order to introduce it in stored products protection.

Materials and Methods

The insect test *Tribolium castaneum* (flour beetle) was reared on the artificial diet in a temperature controlled room at 25±1°C and 75±5% relative humidity. Adults of 14-15 days post-emergent and last stage larvae were used in the experiments. 50 individuals of each stage of the insect were put into a glass bottle (8/5 cm) without growth substrate and 4 bottles (replicates) were exposed to different treatment variants in desiccators with 2.5 l capacity containing a hole with rubber stoppers. After the exposure the dead and living insects were removed from the desiccators and were counted. Each variant was exposed to the mixture of ethyl formate and carbon dioxide, at 25±1°C, for 1 and 3 hours (because of relevant results, no more time exposure were used) to a series of proposed doses as:

- (i) Variant 1 – ethyl formate 50 mg/l (0.135 ml) and carbon dioxide 25 ml (1%);

- (ii) Variant 2 – ethyl formate 75 mg/l (0.203 ml) and carbon dioxide 50 ml (2%);
- (iii) Variant 3 – ethyl formate 100 mg/l (0.270 ml) and carbon dioxide 75 ml (3%);
- (iv) Variant 4 – ethyl formate 125 mg/l (0.337 ml) and carbon dioxide 100 ml (4%);

V3 and V4 were not tested because V2 has 100% mortality.

Dosage calculation was obtained by evacuating the desiccators to the desired volume of air and supplying with the same volume of CO₂ through the rubber stoppers of the hole of desiccators using a syringe. The liquid ethyl formate was introduced by syringe into desiccators in a desired quantity too. A control desiccator with the same stage of the insect was exposed to the normal atmosphere for the same time period.

The effects of the EF/CO₂ formulations were immediately recorded after exposure time and after 24, 48 and 72 hours of removing the insects from the desiccators.

RESULTS AND DISCUSSIONS

Table 1 shows the effect of the mixture of ethyl formate and carbon dioxide on the adults of *T. castaneum* and table 2 shows the mortality of the larvae under EF/CO₂ formulations. However, only V1 and V2 doses were used because even at V2 concentrations, all life stages of *T. castaneum* were fully controlled. It can be observe from the table that the larvae are more resistant to the fumigant mixture; 61 out of 80 larvae were found dead immediately after 1 hour exposure, but after 24 hours some of them were alive (only 52 remain dead); at 72hrs of exposure period, 49 dead larva was obtained which is the final results of mortality.

Table 3 shows the percentage of mortality and can be observe that after 72 hour from the exposure time of 1 hour, 95% of adults and 61,25% of larvae were dead; at 3 hours exposure, 100% of adults and 98,75% of larvae were dead.

The subsequent trials will do to find the proper concentrations, exposure time and appropriate insect stage to control stored-grain insects.

CONCLUSIONS

- The good efficacy of fumigant mixture of ethyl formate and carbon dioxide was demonstrated even at lower tested concentration (ethyl formate 50 mg/l and carbon dioxide 1%);
- For the adults of *Tribolium castaneum* one hour exposure was enough for the 95% mortality; for the larval stage after 3 hour exposure was obtained 98.5 % mortality;
- Further laboratory and field studies are required to ascertain the true applicability of this fumigant mixture.

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Table 1Effect of EF/CO₂ formulations on adults of *T. castaneum*

<i>1 hour exposure</i>				
Variant	Number of dead individuals at.....hours after the exposure			
	immediately	24 hours	48 hours	72 hours
V1	200	193	190	190
V2	200	200	200	200
Control	0	0	0	0
<i>3 hours exposure</i>				
V1	200	200	200	200
Control	0	0	0	0

Table 2Effect of EF/CO₂ formulations on larvae of *T. castaneum*

<i>1 hour exposure</i>				
Variant	Number of dead individuals at.....hours after the exposure			
	immediately	24 hours	48 hours	72 hours
V1	153	130	130	123
V2	200	200	200	200
Control	0	0	0	1
<i>3 hours exposure</i>				
V1	200	200	200	197
Control	0	0	0	2

Table 3Mortality (%) at 72 hours of the *T. castaneum* under EF/CO₂ formulations

Variant	Adults		Larvae	
	1 hour exposure	3 hours exposure	1 hour exposure	3 hours exposure
V1	95	100	61,5	98, 5
V2	100	100	100	100
Control	0	0	0.01	0.02