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Comparison between the insecticidal efficacy of a novel pellet formulation and an obsolete one, Phostoxin

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Abstract

Due to insect resistance to common fumigants such as Phostoxin, and some arguments about the genotoxicity of this pellet, production of alternative pellets with same application seems necessary. The aim of this work was comparison between insecticidal efficacy of Phostoxin and novel pellets based on Eucalyptol, botanical constituent, and Poly (vinyl alcohol) against *Tribolium castaneum*, *Callosobruchus maculatus* and *Rhyzopertha dominica* in 28±2 °C, 24 and 48 h exposure time. Mortality percent in 24 h ± S.E. of 0.25 and 0.3 g Phostoxin pieces in 159 liter barrels against more tolerant (*R. dominica*) and more susceptible (*T. castaneum*) species were 33.3±4.4, 90±4.4, 45±2.8 and 100, respectively. Mortality percent in 24 h ± S.E. of 3 and 4 g novel pellets (with 3:5 dose) in 20 liter casks against more tolerant (*T. castaneum*) and more susceptible (*C. maculatus*) species were 0, 8.33±4.4, 35±2.8 and 48.33±4.4, respectively. In general, Phostoxin pellets were more effective than novel pellets, but results showed that Eucalyptol based pellets have potential toxicity to surrogate instead of obsolete Phostoxin pellets.

Keywords: Eucalyptol, Fumigant toxicity, Phostoxin, PVA, stored product pests

1. Introduction

Chemical control of stored products pests using obsolete chemicals pesticides such as Phostoxin, methyl bromide and Sulphuryl fluoride may cause special problems on stored products and the environment [1, 2]. Insect resistance to common fumigants such as phosphine etc. is a global issue now and control failures have been reported in field situations in some countries [1, 2, 3, 4]. In addition, there have been some arguments about the genotoxicity of phosphine [5].

It is believed that essential oils have the advantage over conventional fumigants in terms of low mammalian toxicity, rapid degradation and local availability. They do not leave residues toxic to the environment and have medicinal properties for humans [6]. Genus *Eucalyptus* (family: Myrtaceae) has various species that contains large amounts of essential oil. Their essential oil showed insecticidal efficacy in many documents [7, 8, 9]. 1,8-Cineole was most important compound of the genus *Eucalyptus*, and largely responsible for its insecticidal properties in majority of researches [10, 11].

The vapor pressure of plant essential oils including 1,8-Cineole is low. Evidently, compounds of plant origin can be used only for small-scale applications or for space treatment [12]. However their use in some formulations such as pellets, granules etc. as controlled release system could improve their efficiency as applied insecticides.

Controlled release technology has emerged as an alternative approach with the promise to solve the problems accompanying with the use of some agrochemicals, while avoiding possible side effects with other methods. In this technology, selection of carrier substrate (polymers in most cases) is more important step. PVA (poly (vinyl) alcohol) is a famous carrier polymer in control release purposes in agriculture that has good gas barrier properties and good printability. Its water solubility, reactivity, and biodegradability make it a potentially useful material in biomedical, agricultural, and water treatment areas [13].

The aim of this work was comparison between insecticidal efficacy of Phostoxin and novel pellet based on Eucalyptol, botanical constituent, and Poly (vinyl alcohol) against *Tribolium castaneum* (Herbst), *Callosobruchus maculatus* F. and *Rhyzopertha dominica*.

2. Material and Methods

2.1. Chemicals

1,8-Cineole with 154.25 g/mol molar mass and Assay (GC, area%) $\geq 98\%$ was purchased from Merck (Darmstadt, Germany). Poly (vinyl alcohol) (Molar Weight: 72000 g/mol; hydrolysis mole: 98) was purchased from Merck (Darmstadt, Germany). All experiments were done in 2011 (June-September).

2.2. Pellets preparation

PVA and 1,8-Cineole mixture was prepared by Dry Mixing Method (DMM) in ice temperature on stirrer with 500 rpm for 4 h. Preparation of mixture was done in tight 100 ml balloons and mixing power provided with magnetic stirrer. Then prepared mixture was transferred to FTIR pellet maker apparatus (Thermo Nicolet part No. 0016-035, USA) and pellets were produced by hydraulic pressure under 100 Kg. Production of a pellet from prepared mixture was done in about 1 min. Average of ten 1 g pellets diameter and thickness were measured by calliper that were 1.3 ± 0.1 and 0.93 ± 0.01 cm, respectively [14].

2.3. Insects

The insects were obtained from laboratory cultures from Islamic Azad University, Izeh branch, Iran. Rust red flour beetle *Tribolium castaneum* (Herbst), Cowpea beetle *Callosobruchus maculatus* (F.) and lesser grain borer *Rhyzopertha dominica* (F.), were reared on flour (mixed with yeast), cowpea and wheat (12% R.H.), respectively. Flour, wheat and cowpea were purchased from a local market in Izeh, Khuzestan province, Iran. Purchased materials were kept in oven (120 °C) for 2 h in order to kill all probable organisms, before use as culturing media. Experimental procedures were carried out at $28 \pm 2^\circ\text{C}$ in a dark room, and $65 \pm 5\%$ R.H. was provided only for culturing media.

2.4. Bioassays

Adults (1-3 days) were used as test insects. Experiments were carried out at $28 \pm 2^\circ\text{C}$ in a dark room, and $65 \pm 5\%$ R.H. Mortality was assessed 24 and 48 h after treatment. One gram novel pellets were used as experimental traits and phostoxin pieces on this study were 0.25 and 0.3 g in weight. Barrels with 159 L volume and 20 L casks were used as fumigation chambers for Phostoxin and pellets, respectively. Fumigation chambers were sealed by parafilm and adhesive tape (5 cm diameter). Insects were located in small vials dangled from barrel and cask caps.

2.5. Data analysis

Mortality percentages were calculated by the Abbott correction formula for natural mortality in the untreated control (Abbott, 1925). Mortality data were altered in $\arcsin \sqrt{x}$ then were analyzed using the SPSS 18.0 software. Mean comparisons were performed using one way analysis of variance (ANOVA). Tukey test at $\alpha = 0.01$ was used to determine the difference between the mean of mortalities.

3. Results

Mortality percent of 0.25 g Phostoxin piece in 24 h at 159 liter barrels against more tolerant (*R. dominica*) and more susceptible (*T. castaneum*) species were 33.3 ± 4.4 , 90 ± 4.4 respectively (Fig 1). Also mortality percent of 0.3 g Phostoxin piece in 24 h at 159 liter barrels against more tolerant (*R. dominica*) and more susceptible (*T. castaneum*) species were 45 ± 2.8 and 100, respectively. While, percentage of mortality

of 0.25 g pieces of Phostoxin in 48 h against *R. dominica*, *C. maculatus* and *T. castaneum* were 74 ± 4.4 , 100 and 100, respectively. Also, percentage of mortality of 0.3 g pieces of Phostoxin in 48 h against *R. dominica*, *C. maculatus* and *T. castaneum* were 94 ± 2.8 , 100 and 100, respectively (Table 1).

Table 1: Mortality percent \pm S.E. of different stored product beetles exposed to phostoxin 0.25 and 0.3 g pieces in 24 and 48 h

Species	24 h		48 h	
	0.25 g	0.3 g	0.25 g	0.3 g
<i>T. castaneum</i>	90 ± 4.4	100	100	100
<i>C. maculatus</i>	63 ± 2.8	88 ± 2.8	100	100
<i>R. dominica</i>	33.3 ± 4.4	45 ± 2.8	74 ± 4.4	94 ± 2.8

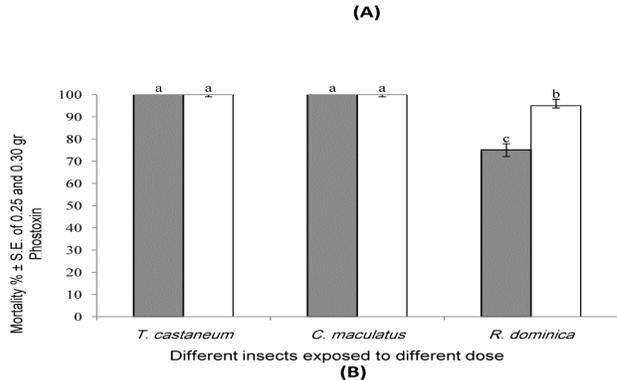
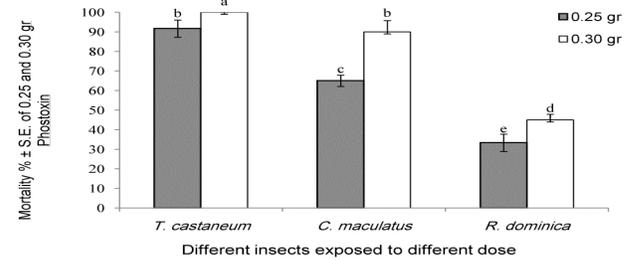


Fig 1: Mortality percent of 0.25 and 0.30 g Phostoxin pellets in 24 h (A) and 48 h (B) (Different letters over columns indicate significant differences according to Tukey test at $\alpha = 0.01$. Columns with the same letter are not significantly different. Vertical bars indicate standard error (\pm); very small values are not represented).

Mortality percent of 3 g botanical pellets in 24 h at 20 liter casks against more tolerant (*T. castaneum*) and more susceptible (*C. maculatus*) species were 0 and 33.8 ± 4.4 respectively (Fig 2). Also mortality percent of 4 g botanical pellets in 24 h at 20 liter casks against more tolerant (*T. castaneum*) and more susceptible (*C. maculatus*) species were 35 ± 2.8 and 48.33 ± 4.4 , respectively. While, percentage of mortality of 3 g botanical pellets in 48 h against *R. dominica*, *C. maculatus* and *T. castaneum* were 57 ± 2.8 , 100 and 22 ± 4.4 , respectively. Also, percentage of mortality of 4 g of botanical pellets in 48 h against *R. dominica*, *C. maculatus* and *T. castaneum* were 86 ± 4.4 , 100 and 47 ± 6.4 , respectively (Table 2).

Table 2: Mortality percent \pm S.E. of different stored product beetles exposed to botanical pellet 3 and 4 g in 24 and 48 h

Species	24 h		48 h	
	3 g	4 g	3 g	4 g
<i>T. castaneum</i>	0	35 ± 2.8	22 ± 4.4	47 ± 6.4
<i>C. maculatus</i>	33.8 ± 4.4	48.33 ± 4.4	100	100
<i>R. dominica</i>	28.1 ± 4.4	40 ± 4.4	57 ± 2.8	86 ± 4.4

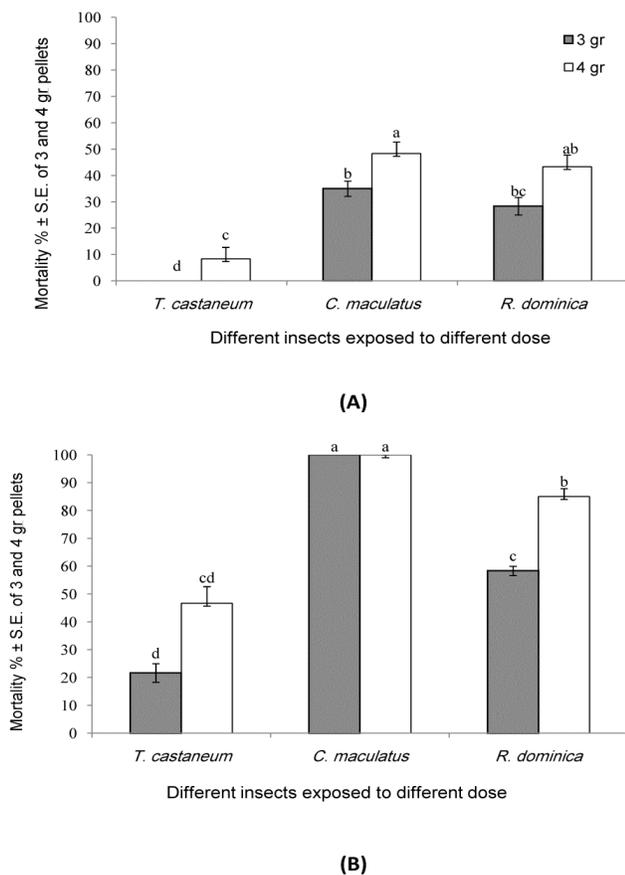


Fig 2: Mortality percent of novel 3 and 4 g pellets in 24 h (A) and 48 h (B) (Different letters over columns indicate significant differences according to Tukey test at $\alpha=0.01$. Columns with the same letter are not significantly different. Vertical bars indicate standard error (\pm); very small values are not represented).

As figures 1 and 2 shown, significant difference was observed between mortality of different insect species and different exposure times and also different pesticide weights in majority of experiments.

4. Discussion

Chemical insecticides such as phostoxin and methyl bromide etc. have high efficiency to control insect pests in storages [3]. These compounds have some disadvantages like genotoxicity, environmental pollution, resistance occurring in insect populations along with the risk of being misused too [1, 3, 4]. According to Montreal clean air act protocol, using mentioned chemical insecticides in storages is banned since 2005 in developed countries and since 2015 in developing countries like Iran [15]. In the current study, phostoxin pieces were compared with natural origin pellets to show the novel pellets potential as insecticide in storage.

Natural pesticides have some benefits in compare with chemicals such as their low toxicity to human and other non target organisms, having no harmful residue in environment and reducing the chance of misuse etc. [6]. Plant essential oils are relatively new and promising alternatives for chemical insecticides now [6, 12, 14]. Their use as fumigant agent in large scales encountered some problems such as low vapor pressure that could be solved using some formulations such as control released formulations [12]. The vapor pressure of 1,8-Cineole (Eucalyptol) is very low (< 1 mm Hg at 20 °C) while Phosphine (31,920 mm Hg at 23 °C), Methyl bromide (1250 mm Hg at 20 °C) and Sulphuryl fluoride (12,087 mm Hg at 20

°C) have high vapor pressure. Evidently, compounds of plant origin can be used only for small-scale applications or for space treatment [12].

Controlled release formulations were applied in agriculture and studied using different polymers as pesticide delivery agents by many researchers [14, 16, 17, 18, 19, 20]. Polymers with biodegradable, non-pollutant residues and nontoxic characteristics could be beneficial in pesticide delivery systems in agriculture [18]. However, application of natural polymers is more recommended [16, 17, 19, 20]. Some researchers used naturally origin polymers as barrier matrix [16, 17, 19, 20]. While other researchers used petrochemical polymers as barrier in their formulations [18]. Using natural polymers could reduce the risk of pollution and health problems. despite of, chemical origin polymers have better binding potential and release efficiency [12, 13, 16].

In current study we used pellet formulation with fumigant active ingredient for the first time. So, the pellet making chamber was completely sealed. Obviously, size and shape of formulation is related with the efficiency of release. Similar studies used different size of formulations from spheres with 8 cm [16] to beads with 1.07-1.34 mm [19, 20] diameter. Smaller diameter with higher surface/volume relation could reduce the release time.

Our results showed that control of stored product beetles using Phostoxin, was more effective than natural compounds. Despite of, residue and side effects of botanical materials and their derivatives are less than Phostoxin. Total benefits of natural compounds in addition to their potential insecticidal efficiency make their use justifiable. Overall, we are hopeful that similar studies will be done on new alternatives for chemical pesticides.

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