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Potential Activity of Mineral Oil (KZ), Biocide (Ivomic) and Organophosphorus (Pirimiphos-Methyl) and Their Joint Action Against *Tyrophagus putrescentiae* (Schrank)

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Abstract: Mites are considered one of the most abundant groups of pest arthropods. In tropical and subtropical area, *Tyrophagus putrescentiae* Schrank is a most common mite with high reproductive potential and short life cycle in stored products. It not only damage the stored product causing economic losses, reducing nutrient content but can also cause allergic reactions in humans. An experiment was carried out at laboratory of Stored Product Pests department, Plant Protection Research Institute at Sakha Agricultural Research Station during 2021 season to study the effect of pirimiphos-methyl, ivomic and mineral oil KZ against both adults and larvae of *T. putrescentiae* through toxicity and biological aspects as well as joint action of the tested compounds. Results showed that pirimiphos-methyl was the most effective compound on the tested species followed by ivomic and KZ. Based on LC_{50} values there was no significant difference between the efficacy of the ivomic and pirimiphos-methyl against *T. putrescentiae*. All tested compounds had significant effect on the aspects of biology compared to control. In addition to the combination of pirimiphos-methyl with ivomic and KZ showed an additive or synergistic effect. While, combination of ivomic and mineral oil showed additive effect. According the current findings the ivomic can replace the pirimiphos-methyl to reduce environmental pollution and to ensure also showed the effectiveness of KZ and ivomic in activating the pirimiphos-methyl and can minimize the development of resistance in storage mites to organophosphorus insecticide. Furthermore, ivomic and KZ can utilize as sterilizers to test mite.

Key words: Mites · *Tyrophagus putrescentiae* · Pirimiphos-methyl · Ivomic · Mineral oil KZ · Joint action

INTRODUCTION

In Egypt, one of the goals of Ministry of Agriculture is to increase and keep the yield of cereal crops to reduce gap between consumption and production. Wheat is the main diet for Egypt population. Wheat (*Triticum aestivum* L.) is one of most cereal crops in many countries of the world. The stored product are liable to be attacked not only by insects, but also by mites, which are either free living, granivorous, fungivorous, saprophagous or parasitic or as predator on other mites and insects [1-6].

In Egypt in recent years, mites received an increasing attention from both entomologists and inflict on stored products. Mites occur in grains, seeds and other stored products are causing serious losses every year, specially in areas with high temperature and relative humidity as happens in Egypt [7-14].

Mites not only eat cheese and decrease its quality, but they also often cause allergies and or stomach disorders to persons through handling or eating the infested food [15-17]. *Tyrophagus putrescentiae* is an important pest of stored products having a high fat or protein content. The susceptibility of the food grains to mite attack depends on the high humidity, so fitness and high nutritive value of the food grains at optimum temperature [5, 18, 19]. It can cause problems for foodstuffs ranging from weight reduction and degradation of stored food to accumulation of harmful residues (fungi, dead mites, feces, eggs and bits of foods) through

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their activities [20-23]. This makes the infested grain storage unhygienic. So stored products pests management is an important economic problem and strategies to control these pest by chemical, physical and biological techniques. The most common method for controlling stored products mites is the use of pesticides [24, 25]. The only contact pesticides approved by the Pesticides and Safety Directorate for the treatment of stored grain and oilseed are organophosphorus (OP)compounds pirimiphos-methyl (Actellic), etrimfos (Satisfar) and chloropyrifos-methyl (Reldan) [26, 27]. Results from recent surveys have found widespread resistance in populations of *Acarus siro* to one or more of these compounds. Now it has become necessary to search for methods for pest control with lower mammalian toxicity. There are a number of alternative compounds used effectively against acarine pests in field agriculture, veterinary and public health control programs, which may also prove effective against storage mites. These include insect growth regulators, insect dusts, botanicals, novel compounds and biological control agents [28-34]. The use of different oils as protectants including mineral oils gave promising results. Consequently, the current study was designed to evaluate one organophosphorus (Actellic), one mineral oil (KZ) and one biocide (ivomic) against *T. putrescentiae* through determination, toxicity, biological and joint action activities.

MATERIALS AND METHODS

Pest Culture Technique [*Tyrophagus putrescentiae* (Schrank)]: Adults of *T. putrescentiae* obtained from a culture originally got from Romano cheese infested by the mentioned mite, was established under constant temperature of 29 ± 1°C and 70 ± 5 R.H and feed on Baker's yeastat laboratory of Stored Product Pests department, Plant Protection Research Institute at Sakha Agricultural Research Station during 2021 season. Individuals of mite were cultured in small plastic units of 3 cm in diameter and 4 cm deep, where each was filled up to 0.7 cm by a binary mixture of plaster and charcoal (9:1) and tightly covered by a piece of glass slide, using a rubber band to tighten the cover with the used vial.

Chemical Used

Organophosphorus Compound:

- Common name: Pirimiphos-methyl.
- Trade name: Actellic.
- Chemical name: O-2 diethyl amino-6-methyl-pyrimidin-4-yL O, O-dimethyl Phosphorthioate.

Biocide (Ivomic):

- Ivomic 1 % w/v ivermectin and 10 % w/v clorsulon in a sterile solution.

Mineral Oil (KZ):

- Emulsifiable mineral oil.
- Essential mineral oil (95 %) (w/v).
- Emulsified material (5 %).
- Produced by Kafr El-Ziat Company (Egypt).

Bioassay of the Tested Compounds

Baker's Yeast Method: According to [35], mite individuals [adults (males and females) or larvae] of *T. putrescentiae* were confined in small glass tube and exposed to treated (series of concentrations) or untreated (treated with distilled water) small pieces of Baker's yeast (0.2 g each), placed on filter paper (2 × 2 cm). The adults were divided into groups each of which containing 3 replicates. The last groups were treated by distilled 0.2 ml water. All treatments and control examined at two different intervals (24 and 48 hr) and kept at the same mentioned temperature degree before counts. Mortality was recorded after 24 and 48 hours. All results were corrected according to [36] formula as follows:

$$\text{Mortality \%} = \frac{\text{Mortality \% of treatment-Mortality \% of control}}{\text{100-Mortality \% of control}} \times 100$$

Data were plotted on log dosage-probit papers and statistically analyzed by [37].

Toxicity index of tested compounds were determined according to [38] as follows:

$$\text{Toxicity index} = \frac{\text{LC50 of the most effective compound}}{\text{LC50 of a tested compound}} \times 100$$

Biological Effect of the Tested Compounds (*T. putrescentiae*): Baker's yeast method of [35] was carried out. In this procedure mite individuals were confined in small glass tube and exposed to LC_{50} treated or untreated (treated with distilled water) small pieces of Baker's yeast (0.2 g each), placed on filter paper (2 × 2 cm). The adults were divided into groups each of which containing 3 replicates. The last groups were treated by distilled 0.2 ml water and kept at the same pre-mentioned temperature. Mortality counts were recorded after 24 and 48 hours. All results were corrected according to Abbott's [36]. Survival animals were discarded after 3 days. The number of eggs, larvae, nymph and adults was recorded. Percent of reduction of all stages, sterility and hatchability compared with untreated check was calculated according to the following equation:

$$\frac{C-T}{C}\times 100$$

where:

C = Number of control stage.
T = Number of treated stage.

$$\%\text{Hatchability}=\frac{\text{Number of Larvaes}}{\text{Number of eggs}}\times 100$$

$$\%\ \text{Sterility}=100-[\frac{a\times b}{A\times B}\times 100]$$

where:
a = Number of eggs laid/female in treatment.
b = % of hatchability of treatment.
A = Number of eggs laid/female in untreated control.
B = % of hatchability in untreated control.

Statistical Analysis: The studied treatments were arranged in Completely Randomized Block Design (CRBD) design in three replicates. Analysis of variance was carried out using MSTAT-C Statistical Software Package [39]. The comparison of means was investigated using Duncan's multiple range test [40] test at 0.05% probability.

Joint Action of Tested Compounds Against *T. putrescentiae*: Mixing with medium method was used to estimate the joint action of the binary mixture of pirimiphos-methyl + KZ, pirimiphos-methyl + ivomic and KZ + Ivomic against *T. putrescentiae* (2 – 3 days old) as described by [41]. The components of the mixtures either separately or in combination were applied at the LC_{50} level. The LC_{50} (expected) was concluded from logarithmic dosage and probability line of each test toxicant. The expected LC_{50} of each toxicant was separately dissolved alone or in binary mixture in acetone. One ml of each toxicant alone or in combination was placed at the bottom of petri dish (9 cm in diameter) and left to dry. After complete dryness of the film, ten adults of *T. putrescentiae* (2 -3 days old) were separately transferred into petri dish. The mortality percentage was recorded 2 days after treatment and corrected by Abbott's [36] formula. Control was prepared with acetone only. Each treatment and control was repeated three times. The expected mortality of the mixture was the sum of actual moralities of the dosages used in the combination. The observed mortality of the mixture was the actual mortality obtained from treatment of combined LC_{50}.

The joint action was evaluated by using the following equation:

$$\text{Co-toxicity factor}=\frac{\text{Observed \% mortality-Expected \% mortality}}{\text{Expected \% mortality}}\times 100$$

RESULTS AND DISCUSSION

Toxic Activity of the Tested Compounds: Toxicity of three compounds: one organophosphorus; pirimiphos-methyl, one mineral oil; (KZ) and one biocide; ivomic were studied against *T. putrescentiae* by exposure to treated medium method.

Exposure to Treated Medium for *T. putrescentiae*: The toxic activity of the tested compounds was evaluated against *T. putrescentiae* laboratory strains. Adults of species were exposed to small pieces of Baker's yeast for *T. putrescentiae* were admixed with the desirable concentrations of the tested compounds. Ld-P lines for the tested insecticides are drawn, LC_{50} values with their confidence limits and slope values are tabulated in Table (1). Data indicated that pirimiphos-methyl was the most toxic compound with LC_{50} of 0.6 mg/g for *T. putrescentiae* at 24 hours post treatment. The least toxic compound was the mineral oil with LC_{50} value of 210 mg/g for *T. putrescentiae*. Based on the LC50 values there was no significant difference between the efficacy of the biocide ivomic and pirimiphos-methyl against *T. putrescentiae*. As a conclusion pirimiphos-methyl was the most effective compound on the tested species followed by the biocide; ivomic and KZ oil. The results in Table (2) revealed a toxicity was achieved for all tested toxicants against the larvae of *T. putrescentiae* where the potential rank was as followed, pirimiphos-methyl (1.1 mg/g), ivomic (1.9 mg/g) and KZ (200 mg/g) at 24 hours post treatment. Concerning the tested OP compound. These findings are in good agreement with [33, 42-47] who found that pirimiphos-methyl was the most toxic to both the susceptible and malathion resistant strain of *T. castaneum* and more toxic than malathion.

Effect of Tested Compounds on *T. putrescentiae* Progeny: Results in Table (3) showed that the mean number of eggs laid, hatched eggs, progeny and the percentage of sterility highly affected by the different tested toxicants. There were significant difference between all treatments and control. In addition that data obtained in Table (4) showed that tested materials have sterilizing properties to

Table 1: Toxicity of the tested toxicants to adults of *T. putrescentiae* by exposure to treated medium (after 24 hours)

Toxicant	LC_{50} mg/g	Confidence limits		Slope	Toxicity index
		Lower	Upper		
Organophosphorus (pirimiphos-methyl)	0.60	0.37	0.96	1.4	100.00
Biocide (Ivomic)	0.66	0.55	0.79	5.0	90.91
Mineral oil (KZ)	210.00	150.00	294.00	2.0	0.28

Table 2: Toxicity of the tested toxicants to larvae of *T. putrescentiae* by exposure to treated medium (after 24 hours)

Toxicant	LC_{50} mg/g	Confidence limits		Slope	Toxicity index
		Lower	Upper		
Organophosphorus (pirimiphos-methyl)	1.1	0.8	1.4	2.50	100.00
Biocide (Ivomic)	1.9	1.0	3.6	1.04	57.90
Mineral oil (KZ)	200.0	153.8	260.0	2.50	0.55

Table 3: Analysis of variance for the number of eggs laid, hatched eggs, percentages of hatching, number of offspring and sterility of *T. putrescentiae*

SOV	DF	MS						
		Total eggs	Egg/female	No. of hatching	Hatchability	Adults	% progeny	% sterility
REP (R)	2	12.121	0.041	3.069	1.616	3.478	0.001	8.415
Treatment (T)	3	488.079**	2.269**	509.329**	6829.071**	533.802**	7412.328**	3597.013**
Error	6	1.983	0.047	3.223	1.679	4.593	2.287	2.518

Table 4: Means of hatched eggs, hatchability, progeny and sterility of *T. putrescentiae*

Treatment	Means of						
	Total eggs	Egg/female	No. of hatching	Hatchability	Adults	% progeny	% sterility
Control	40.00[c]	2.70[c]	39.00[c]	100.00[c]	40.00[c]	100.00 [b]	0.00[a]
Pirimiphos-methyl	0.01[a]	0.01[a]	0.01[a]	0.01[a]	0.01[a]	0.01[a]	100.00[c]
Ivomic	0.02[a]	0.02[a]	0.05[a]	0.02[a]	0.02[a]	0.02[a]	100.00[c]
KZ	14.00[b]	0.90[b]	3.67[b]	78.60[b]	4.67[b]	100.00[b]	73.80[b]

Table 5: Toxicity of binary mixtures of pirimiphos-methyl with ivomic, pirimiphos-methyl with mineral oil (KZ) and ivomic with mineral oil (KZ) against *T. putrescentiae* using mixing with feeding medium

Treatment	% mortality after 24 hours	Co-toxicity factor	Combined effect
Pirimiphos-methyl	50		
Ivomic	50.0		
KZ	50.0		
Pirimiphos-methyl + Ivomic	100.0	0.0	Additive or synergism effect*
Pirimiphos-methyl + KZ	100.0	0.0	Additive or synergism effect*
Ivomic + KZ	83.30	-16.70	Additive effect

* The mixtures in pairs which exhibited observed mortality of 100 % may be evaluated as an Additive or synergism effect

T. putrescentiae. Pirimiphos-methyl and the antibiotic ivomic caused 100 % sterility while mineral oil, KZ achieved 73.80 % sterility compared to control. Consequently the biocide ivomic is considered promising sterilizer agent against the tested mites followed by KZ. Chemosterilants are chemicals that arrest or adversely affect reproductive capacity and are therefore obvious candidates for use in pest control programs. Chemosterilants can may act of three principle ways. They may lead to failure to produce ova or sperm, death of sperm or they may produce multiple dominant lethal maturation or severely injure the genetic material in the sperm and ova [48]. Potassium iodide (KI), boric acid at concentration higher than 0.5 %, folic acid prevented egg laying by *Acarus siro* (Astigmata), killed *T. putrescentiae* (Astigmata) when applied to the diet [49-51]. The results in Table (4) had the same trend. According to mean of progeny pirimiphos-methyl and ivomic were the most effective compounds followed by mineral oil (KZ). Percentage of sterility indicated the same trend. In conclusion, all tested compound had significant effect on all tested parameters compared with control. These findings are in good agreement with [52-57].

Effect of Binary Mixtures of the Test Compounds Against *T. putrescentiae*: The joint action of binary mixtures of tested toxicants against *T. putrescentiae* adults was estimated after determination of the expected LC_{50} values of each toxicant (after 24 hours exposure), thus 100 % mortality was expected as a result. The joint action was determined according [41] by estimating the co-toxicity factor (Co-F). This factor was used to distinguish the result into three groups. A positive factor of (20 or more) means synergism, a negative factor of (- 20 or more) means antagonism and intermediate value was considered additive effect. The effect of the tested compounds in binary combinations against *T. putrescentiae* was studied and the results are recorded in Table (5).

The combination of pirimiphos-methyl with ivomic and KZ results showed an additive or synergistic effect. While, combination of ivomic and mineral oil showed additive effect (Table 5).

Ismail [58] reported that, based on laboratory tests, synergistic action was observed in the whitefly by a combination of imidaclopride with jojoba oil or KZ oil approximately 12 and 40 times more respectively than the imidclopride alone. Similar trend was also observed for these mixtures at sublethal dose against biological aspect of whitefly.

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