

INVESTIGATION of SEVERAL AEROSOL INSECTICIDE COMPOSITIONS CONTAINING SYNERGISTS

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Abstract Two aerosol insecticide compositions were investigated in laboratory conditions against a complex of synanthropic insects. Both compositions have high acute insecticidal activity and have a long residual effect on glass, but not on plywood. It is shown that the knockdown effect was reversible on plywood on the 3rd and 7th days after treatment for cockroaches and on the 7th day for bugs. Fifteen aerosol compositions containing cypermethrin, imiprothrin alone or mixed with the synergists piperonyl butoxide and/or MGK-264 in different concentrations were prepared specially for laboratory use and were investigated. It is shown that the synergism ratio was higher in houseflies than in cockroaches in compositions containing PBO, but it was higher in cockroaches when it contained an MGK-264-like synergist.

Key Words Cockroach housefly piperonyl butoxide MGK-264 resistance

INTRODUCTION

Detoxication of many xenobiotics, which are complex ethers of organic acids, is catalyzed by various enzymatic systems, specifically monooxygenases (MO) and nonspecific esterases (NE). This is also true for synthetic pyrethroids. Representatives of two orders of the class Insecta: German cockroach and house fly, served as biological models with different levels of MO and NE activity.

Piperonyl butoxide (PBO) and MGK-264 are known as inhibitors of insect detoxication systems. Both synergists are used in various insecticide compositions, mainly in aerosol cans. The best synergism occurred in a mixture of PBO or MGK-264 with pyrethrins. Insecticide resistance can be a serious problem in house fly and cockroach control. When a resistance mechanism is metabolic, it is often possible to negate that mechanism by use of synergists (Cochran, 1987, 1994; Yu and Hsu, 1993; Wu et al., 1998). Several registered insecticide compositions include the synergists PBO and/or MGK-264, which are intended for medical disinfection in public health for control of synanthropic insects in Russia: microencapsulated "Effective Ultra" (Netherland), aerosol cans "Dikhlophos VP", "Dikhlophos Super", "Dikhlophos L", "Knockout LN" (Russia) and others.

MATERIALS and METHODS

The sensitive standard strain of German cockroach *Blattella germanica* (L.), oriental cockroach *Blatta orientalis* L., bed bug *Cimex lectularius* L., rat flea *Xenopsylla cheopis* Roths., and house fly *Musca domestica* L. (strain Cooper) have been used in this study.

The biological efficacy, i.e., direct spray, and the residual efficacy on porous (plywood) and non-porous (glass) surfaces of the aerosol formulations #1 and #2 against the insects mentioned above have been tested under laboratory conditions (temperature at 25±1°C, humidity between 55±10% RH).

Direct spray test: crawling insects are held in 0.5 l glass vessels (3 replicates, 20 insects each), and the aerosol can is held in a clamp and aligned so that spray is directed into the open top

of the vessels from a distance of 45 cm (dose 20 g/m²). After spraying, insects are transferred to clean containers for observation of knockdown and mortality.

Residual test: plywood and glass surfaces (200 cm², 3 replicates each) sprayed from a distance of 45 cm (dose 20 g/m²). After 24 h test insects were placed on surfaces for 15 min. Then they were transferred to clean containers for observation of knockdown and mortality.

Direct spray for flying insects: 300 flies were held in a 2 m³ glass box, and a 1 g/m³ aerosol composition was sprayed into it. Knockdown times were observed at 1% to 99% flies (min), then we calculated C₁₅ (concentration of active ingredient (AI) in the air which caused 99% fly mortality at 15 min, mg/m³), Q₁₅ (quantity of aerosol which caused 99% fly mortality at 15 min, mg/m³), and KT₅₀ min, value has been determined graphically.

The toxicological method implies the topical treatment of the insects, which were first anesthetized with diethyl ether for 20 s. Acetone solutions (1 µl) at various concentrations were applied to the mesonotum of the house flies imago and to the prothorax of cockroaches. The level of mortality was estimated within 24-48 h. The degree of toxicity was estimated through determination of LD₅₀ µg/g (the dose leading to the death of 50% of the experimental individuals). In order to determine LD₅₀, a series of logarithmically decreasing concentrations inducing 20-80% mortality of insects was prepared. Working solutions of the studied compounds (or mixtures) were obtained by successive dilution. Preparation of dilutions of the aerosol mixture from cans was in two steps: the first to remove propellant, the second to prepare a series of acetone concentrations. The data on mortality of insects as a function of the concentration of the studied compounds (or mixtures) were subject to regression analysis, with determination of LD₅₀ and confidence intervals (Popov, 1965a). In studies of the joint action of compounds in mixtures, the synergism ratio (SR) was calculated according to the additive rule (Popov, 1965b). For determination of knockdown time, we fixed the time after treatment with insecticide, when the insect can no longer retain itself on the upright-drilling glass surface.

RESULTS and DISCUSSION

Investigated aerosol cans contained a mix of pyrethroids (#1 permethrin and cypermethrin, #2 imiprothrin and cypermethrin) and the synergist MGK-264. Used as a direct-acting spray, both compositions kill cockroaches and other crawling insects (Table 1). The knockdown effect was rapid in composition #2, perhaps because imiprothrin was available there. The residual effect on the glass surfaces was >21 days for all insects. On the plywood, the residual effect was short for females of the German cockroach, especially in the composition #2. German cockroach males were more sensitive to residuals than were females (Table 2).

The knockdown effect of composition #2 residuals was also investigated for male and female German cockroaches and bugs. Exposure on glass or plywood was 15 minutes, dose 20 g/

Table 1. Direct spray of aerosol compositions

Aerosol composition, #	Test-insect (mixed sex)	% knockdown at time ... (min)					% Killing at 24 hours
		1	5	10	30	60	
#1 permethrin 0.3% cypermethrin 0.2% MGK-264 1.0%	German cockroach	0	100	100	100	100	100
	Oriental cockroach	0	20	50	100	100	100
	Bed bug	0	90	100	100	100	100
	Rat flea	20	100	100	100	100	100
#2 imiprothrin 0.08% cypermethrin 0.1% MGK-264 0.8%	German cockroach	100	100	100	100	100	100
	Oriental cockroach	80	100	100	100	100	100
	Bed bugs	100	100	100	100	100	100
	Rat flea	100	100	100	100	100	100

Table 2. Residual activity of aerosol compositions

Aerosol composition, #	Test-insect	Sex	Surface	% Killing at time ... (days)				
				1	3	7	14	21
#1 permethrin 0.3% cypermethrin 0.2% MGK-264 1.0%	German cockroach	M	Glass	100	100	100	100	100
		F		100	100	100	100	100
		M	Plywood	80	60	50	30	10
		F		30	30	10	0	0
	Bed bug	MF	Glass	100	100	100	100	100
		MF	Plywood	100	100	100	100	100
	Rat flea	MF	Glass	100	100	100	100	100
	#2 imiprothrin 0.08% cypermethrin 0.1% MGK-264 0.8%	German cockroach	M	Glass	100	100	100	100
F				100	100	100	97	87
M			Plywood	30	12	3	10	5
F				23	2	0	10	0
Bed bug		MF	Glass	100	100	100	100	100
		MF	Plywood	90	90	50	60	50
Rat flea		MF	Glass	100	100	100	100	100

Table 3. Knockdown effect of aerosol composition #2, containing cypermethrin, imiprothrin, and MGK-264

Surface/ Insect	Sex	Percentage knockdown in minutes									
		1	5	10	15	20	30	60	120	24 hours	48 hours
1st day after treatment											
Plywood	M	0	0	0	0	7±3	13±5	23±6	50±10	23±7	30±10
German cockroach	F	0	0	0	0	0	17±5	27±5	33±10	10±3	23±10
Glass	M	20±7	80±10	100	100	100	100	100	100	100	100
German cockroach	F	10±3	40±8	90±5	90±5	93±7	100	100	100	100	100
3rd day after treatment											
Plywood	M	-	-	-	3±3	-	8±2	28±5	30±10	12±5	-
German cockroach	F	-	-	-	5±3	-	28±10	27±10	43±15	2±2	-
Plywood Bed bug	MF	-	-	-	30±10	-	-	37±10	90±5	90±5	-
7th day after treatment											
Plywood	M	-	-	-	0	-	0	10±3	10±3	3±3	3±3
German cockroach	F	-	-	-	3±3	-	10±3	23±5	20±5	0	0
Glass	M	-	-	-	100	-	100	100	100	100	100
German cockroach	F	-	-	-	97±2	-	100	100	100	100	100
Plywood Bed bug	MF	-	-	-	0	-	13±10	20±5	70±10	50±15	-

m², 24 h after spray (Table 3). Results show that there were slight, short, and unstable effects on insects even in the 1-7 days interval after spray.

Fifteen aerosol compositions containing cypermethrin, imiprothrin alone or mixed with the synergists piperonyl butoxide (PBO) and/or MGK-264 in different concentrations were prepared specially for laboratory use and were investigated on German cockroach males and house flies (mixed sex) with the topical method. The synergism ratio (SR) was higher in house flies than in cockroaches in compositions containing PBO, but it was higher in cockroaches when there were MGK-264 like synergists (Table 4).

Table 4. Synergism ratio (SR) and toxicity of several insecticide compositions for insects

#	Composition	% a.i. in aerosol can	German cockroaches male		House flies both sexes	
			LD ₅₀ , µg/g	SR	LD ₅₀ , µg/g	SR
1.	Cypermethrin	0.01	0.135±0.020	–	0.27±0.03	–
2.	Cypermethrin MGK-264	0.01 0.8	0.054±0.016	2.5	0.19±0.06	1.4
3.	Cypermethrin PBO	0.01 0.8	0.087±0.023	1.6	0.16±0.07	1.7
4.	Cypermethrin PBO	0.01 0.4	0.096±0.028	1.4	0.13±0.02	2.1
5.	Cypermethrin PBO MGK-264	0.01 0.4 0.4	0.112±0.045	1.2	0.20±0.06	1.4
6.	Imiprothrin	0.008	1.890±0.193	-	0.53±0.08	-
7.	Imiprothrin MGK-264	0.008 0.8	0.790±0.051	2.4	0.58±0.06	0.9
8.	Imiprothrin PBO	0.008 0.8	1.020±0.130	1.9	0.26±0.04	2.0
9.	Imiprothrin PBO	0.008 0.4	1.210±0.210	1.6	0.68±0.07	0.8
10.	Imiprothrin PBO MGK-264	0.008 0.4 0.4	1.890±0.663	1.0	1.05±0.12	0.5
11.	Cypermethrin Imiprothrin	0.01 0.008	0.210±0.041	–	0.65±0.08	–
12.	Cypermethrin Imiprothrin MGK-264	0.01 0.008 0.8	0.035±0.012	6.0	0.55±0.07	1.2
13.	Cypermethrin Imiprothrin PBO	0.01 0.008 0.8	0.210±0.063	1.0	0.33±0.07	2.0
14.	Cypermethrin Imiprothrin PBO	0.01 0.008 0.4	0.350±0.081	0.6	0.48±0.09	1.4
15.	Cypermethrin Imiprothrin PBO MGK-264	0.01 0.008 0.4 0.4	0.470±0.152	0.5	0.33±0.09	2.0

Table 5. Knock-down effect, KT_{50} and toxicity of several insecticide aerosol compositions for *Musca domestica* in glass box (1 g/m³)

#	Composition	% a.i. in aerosol can	Knock-down 1% (min)	Knock-down 99% (min)	KT_{50} (min)	C_{15} mg/m ³	Q_{15} mg/m ³
1.	Cypermethrin	0.01	4'13"	9'06"	6'45"	0.061	613
2.	Cypermethrin MGK-264	0.01 0.8	4'00"	7'33"	5'45"	0.053	533
3.	Cypermethrin PBO	0.01 0.8	3'26"	6'53"	5'15"	0.048	484
4.	Cypermethrin PBO	0.01 0.4	3'23"	8'13"	5'45"	0.057	571
5.	Cypermethrin PBO MGK-264	0.01 0.4 0.4	3'18"	7'42"	5'30"	0.046	460
6.	Imiprothrin	0.008	3'13"	22'31"	13'00"	0.134	1680
7.	Imiprothrin MGK-264	0.008 0.8	3'41"	16'12"	10'00"	0.099	1237
8.	Imiprothrin PBO	0.008 0.8	1'52"	23'40"	13'00"	0.155	1932
9.	Imiprothrin PBO	0.008 0.4	2'14"	22'18"	12'00"	0.119	1487
10.	Imiprothrin PBO MGK-264	0.008 0.4 0.4	1'36"	20'30"	11'15"	0.118	1469
11.	Cypermethrin Imiprothrin	0.01 0.008	2'11"	5'44"	4'00"	0.076	420
12.	Cypermethrin Imiprothrin MGK-264	0.01 0.008 0.8	2'01"	6'27"	4'30"	0.087	483
13.	Cypermethrin Imiprothrin PBO	0.01 0.008 0.8	3'25"	11'30"	7'30"	0.154	855
14.	Cypermethrin Imiprothrin PBO	0.01 0.008 0.4	2'20"	12'23"	7'30"	0.159	885
15.	Cypermethrin Imiprothrin PBO MGK-264	0.01 0.008 0.4 0.4	2'38"	11'41"	7'00"	0.174	967

When mixed with imiprothrin, the presence of MGK-264 results in lesser time for the housefly knock-down effect and greater mortality. In a two-component mixture this effect was lower (Table 5). We know that the most fermentative activity occurred in resistant insect strains such as MO and NE. Our previous investigations show this fact to be true for house flies (Eremina and Bakanova, 1998; Vavilova et al., 1998; Eremina et al., 1999). Application of the synergist PBO with permethrin leads to an increase of susceptibility of houseflies Cooper strain at 10-fold, R-permethrin strain at 100 fold, and VP strain at 60 fold (Eremina and Roslavtseva, 1997). It was to

be expected that in house flies and cockroaches the resistant field strains synergist ratio will be higher than in sensitive laboratory strains.

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