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Growth of selected entomopathogenic fungi species and isolates on media containing insecticides

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Two isolates of *Beauveria bassiana*, *Metarhizium anisopliae*, *Peacilomyces farinosus*, and *Paecilomyces fumosoroseus* were examined on media to which three insecticides at three different doses were added.

Key words: Entomopathogenic fungi.

INTRODUCTION

A number of insecticides were successively investigated to determine the effect of these preparations on the growth of entomopathogenic fungi. The earliest studies mainly concerned arsenate compounds (Vonkassovič, 1925). In the following years other groups of insecticides were tested, such as: organochlorine (Karl, Neubauer, 1953, 1956; Telenga et al. 1959), organophosphates (Sechurina, 1958; Majchrowicz, Miętkiewski, 1976), pyrethroids (Bajan, Fedorko, Kmitowa, 1986; Vänninen, Hokkanen, 1988), and chitin synthesis inhibitors (Keller, 1978; Anderson et al. 1989; Sapieha, Miętkiewski, 1991-92). In most cases the insecticides inhibited the growth of entomophatogenic fungi. However, in some cases the preparations had an opposite effect. It seems that chitin synthesis inhibitors are the least toxic to fungi species.

It was also demonstrated that fungi sometimes racted differently when the same preparation had been applied. A comparative analysis of the effects of chemical preparations on particular strains of selected fungi species was conducted. Similar studies were also conducted by Bajan (1985), Fedorko (1985), Anderson and Roberts (1983), and Olmert and Kenneth (1974).

The objective of this study was to determine the effect of three widely used insecticides on the growth of two isolates of four entomopathogenic hypomycetous species.

MATERIALS AND METHOD

Three types of insecticides were used in the experiment: Bi-58, Fastac and Pirimor (Table 1).

The fungi were grown on Sabouraud medium to which the above preparations were added at 3 dosages:

A – concentration 10 times lower than the recommended dose,

B – concentration identical to the recomended dose,

C – concentration 10 times higher than the recomended dose.

Table 1

List and application doses of insecticides

Trade name of preparation	Name and content active ingredient	Class of toxic	Quantity of preparation per litre of medium			Producer
			Dose A	Dose B	Dose C	
Bi 58 EC	dimeotat 37 %	II	0.1 ml	1 ml	10 ml	Chemie AG Bitterfeld--Wolfen
Fastac 10 EC	alfametryne 10 %	IV	0.025 ml	0.25 ml	2.5 ml	Shell International Petroleum Co. Ltd.
Pirimor 50 DG	pirymikarb 50 %	III	0.06 g	0.6 g	6 g	ICI Agrochemicals

Four species of fungi were investigated: *Baeuveria bassiana* (Balls.) Vuill., *Metarhizium anisopliae* (Metsch.) Sorok., *Paecilomyces farinosus* (Dick ex Fr.) Brown et Smith, *Paecilomyces fumosoroseus* (Wize) Brown et Smith, each of above species occurred in the form of two isolates of different origin. Isolates from group I (n° 1) were isolated from forest soil and those from group II (n° 2) from arable soil.

Insecticides were added to the medium after it had been sterilized and cooled to a temperature of 60°C. The medium containing the highest concentration of pesticides was prepared first (C). The dilution method was used to obtain remaing concentration in media. The medium which did not contain any insecticides was the control. The cultures were set up in triplicate at 22-23°C. The size of the fungi colonies was estimated after 5, 15 and 20 days. The size of the colonies was expressed in percent in relation to the control.

RESULTS

The size of the fungi colonies was dependent upon the concentration of insecticides applied to the medium. The preparations of Bi 58 and Pirimor at concentrations (C) ten times higher that the recommended dose inhibited the growth of all the investigated fungi. In the medium containing Fastac at concentration C, fungi colonies attained 85 % of the size of the control (Table 2). The four fungi species grew

on the medium with insecticides at recommended dosage (concentration B). At this dosage Bi 58 reduced the growth rate of *M. anisopliae* and *P. farinosus* by 20 % as compared to the control.

Table 2

The size of entomopathogenic fungi colonies on media containing insecticides (expressed in % relation to the control)

Species	No of isolate	Diameter of colony after: (days)	Concentration of insecticides								
			Bi-58 37 EC			Fastac 10 EC			Pirimor 50 DG		
			A	B	C	A	B	C	A	B	C
Beauveria bassiana	I	5	81 ± 0.10	73 ± 0.10	bw	80 ± 0.04	45 ± 0.05	52 ± 0.05	83 ± 0.04	81 ± 0.12	bw
		15	96 ± 0.13	95 ± 0.17	bw	101 ± 0.00	89 ± 0.09	89 ± 0.10	102 ± 0.04	102 ± 0.08	bw
		20	96 ± 0.21	100 ± 0.11	bw	101 ± 0.00	99 ± 0.10	95 ± 0.23	99 ± 0.20	107 ± 0.10	bw
	II	5	88 ± 0.04	59 ± 0.08	bw	91 ± 0.05	47 ± 0.10	60 ± 0.07	91 ± 0.05	73 ± 0.10	bw
		15	86 ± 0.08	61 ± 0.12	bw	87 ± 0.05	58 ± 0.22	50 ± 0.08	79 ± 0.19	77 ± 0.05	bw
		20	84 ± 0.36	61 ± 0.20	bw	90 ± 0.09	63 ± 0.19	46 ± 0.17	88 ± 0.14	79 ± 0.05	bw
Metarhizium anisopliae	I	5	79 ± 0.05	56 ± 0.08	bw	90 ± 0.00	50 ± 0.00	40 ± 0.00	65 ± 0.20	58 ± 0.05	bw
		15	99 ± 0.27	64 ± 0.18	bw	90 ± 0.05	70 ± 0.20	58 ± 0.17	98 ± 0.14	96 ± 0.23	bw
		20	97 ± 0.29	59 ± 0.15	bw	93 ± 1.32	73 ± 0.25	62 ± 0.09	101 ± 0.29	88 ± 0.24	bw
	II	5	97 ± 0.09	73 ± 0.00	bw	92 ± 0.12	48 ± 0.10	38 ± 0.05	95 ± 0.11	77 ± 0.05	bw
		15	89 ± 0.05	74 ± 0.10	bw	94 ± 0.26	67 ± 0.15	66 ± 0.15	100 ± 0.18	87 ± 0.09	bw
		20	92 ± 0.05	79 ± 0.16	bw	104 ± 0.36	75 ± 0.40	72 ± 0.10	113 ± 0.10	96 ± 0.10	bw
Paecilomyces farinosus	I	5	100 ± 0.08	64 ± 0.05	bw	98 ± 0.05	27 ± 0.05	57 ± 0.05	104 ± 0.05	63 ± 0.00	bw
		15	95 ± 0.20	72 ± 0.11	bw	95 ± 0.15	47 ± 0.00	54 ± 0.14	100 ± 0.07	73 ± 0.05	bw
		20	92 ± 0.07	75 ± 0.05	bw	92 ± 0.17	49 ± 0.00	55 ± 0.07	99 ± 0.00	77 ± 0.07	bw
	II	5	95 ± 0.09	62 ± 0.00	bw	95 ± 0.05	64 ± 0.10	59 ± 0.19	98 ± 0.09	70 ± 0.05	bw
		15	92 ± 0.15	77 ± 0.05	bw	100 ± 0.05	82 ± 0.18	67 ± 0.14	98 ± 0.20	76 ± 0.09	bw
		20	95 ± 0.23	83 ± 0.00	bw	104 ± 0.21	85 ± 0.20	65 ± 0.14	104 ± 0.20	82 ± 0.15	bw
Paecilomyces fumosoroseus	I	5	102 ± 0.08	84 ± 0.17	bw	94 ± 0.07	57 ± 0.34	73 ± 0.20	107 ± 0.20	88 ± 0.10	bw
		15	100 ± 0.08	99 ± 0.12	bw	101 ± 0.08	80 ± 0.32	81 ± 0.12	103 ± 0.10	97 ± 0.08	bw
		20	96 ±0.08	98 ±0.05	bw	97 ±0.14	86 ±0.20	83 ±0.11	98 ±0.20	94 ±0.31	bw
	II	5	106 ± 0.06	89 ± 0.05	bw	102 ± 0.05	44 ± 0.20	85 ± 0.04	104 ± 0.00	89 ± 0.08	bw
		15	99 ± 0.11	103 ± 0.27	bw	100 ± 0.04	70 ± 0.00	81 ± 0.05	103 ± 0.00	98 ± 0.05	bw
		20	100 ± 0.07	108 ± 0.15	bw	101 ± 0.04	83 ± 0.00	83 ± 0.15	105 ± 0.26	101 ± 0.40	bw

Explanation: concentration of pesticides: A – 10-times lower than the recommended dose; B – identical to the recommended dose; C – 10-times higher than the recommended dose; bw – no growth; ± – standart deviation; I – isolated from forest soil; II – isolated from arable soil

The *B. bassiana* and *P. fumoroseus* colonies were comparable to those of the control. Fastac had a similar effect. In this case, only *B. bassiana* colonies attained 99 % of the size of the control. It was demonstrated that Pirimor was the least toxic. The *B. bassiana* colonies exhibited the largest increase in growth and attained 107 % of the size of the control.

The insecticides which were at concentrations (A) 10 times lower than the recommended dose only slightly affected the growth of fungi. In the medium containing Bi 58 and Fastac, the size of the fungi colonies was comparable with that of the control. However, in the presence of Pirimor the growth of fungi colonies was more enhanced than in the control (*M. anisopliae* colonies attained 113 % of the size of the control).

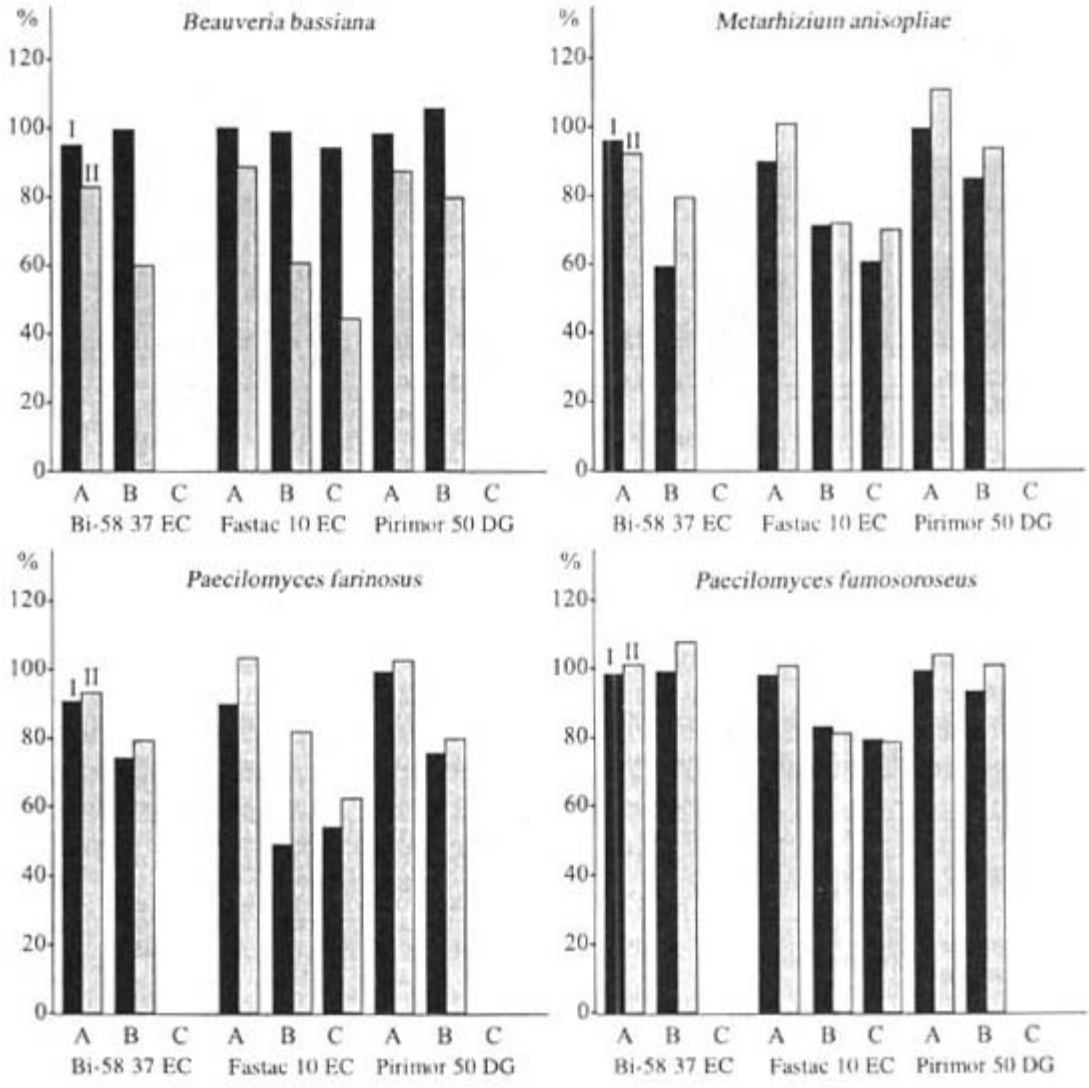

Fig. 1. The size colonies of isolates of selected fungi species on media containing insecticides (expressed in % in relation to the control)
Concentration of insecticides: A – 10-times lower than the recommended dose; B – identical to the recommended dose; C – 10-times higher than the recomended dose; I, II – n° of isolate

The size of the fungi colonies varied depending on the type of isolate and species of fungi used in the experiment (Fig. 1). The most significant differences in the size of fungi colonies were found in the case of *B. bassiana* isolates; isolate n° 1 of this species formed bigger colonies than isolate n° 2. The remaining 3 species showed opposite tendencies. The differences between *B. bassiana* isolates were more significant in the medium containing higher concentrations of insecticides, e.g. in the medium with Fastac at concentration C, the colonies of isolate no1 attained 95 % of the size of the control, whereas those of isolate n° 2 – only 46 %.

The differences in the size of isolates n° 1 and 2 as regards *M. anisopliae*, *P. farinosus* and *P. fumosoroseus* colonies were not so significant as in the case of *B. bassiana* isolates. However, the colonies of isolate n° 2 were bigger than those of isolate n° 1. The differences were marked the most in the case of *P. farinosus*, where the colonies of isolate n° 2 were always bigger than those of isolate n° 1. In the medium containing Fastac at concentration B, the colonies of isolate n° 2 of this fungus attained 85 % of the size of the control, whereas those of isolate n° 1 – only 49 %.

DISCUSSION

The toxic effect of insecticides on the investigated fungi was dependent upon the concentration of those preparations in the medium. It was demonstrated that the preparations of Bi 58 and Pirimor at concentrations (C) ten times higher than the recommended dose were lethal to all the investigated fungi. G a r d n e r, S u t t o n and N o b l e t (1979) indicated that high concentrations of diflubenzuron and methamyl are toxic to fungi. They also found that the growth of *B. bassiana* colonies on the medium with carbaryl at concentration 0.4 % was inhibited to a lower degree than at 0.04 %. In our experiment, in some casses fungi colonies grew better at higher concentrations of pesticides than at lower in the medium. At recomended dosage Bi 58 inhibited the growth of *M. anisopliae* and *P. farinosus* more than that of the remaining species of fungi. Similar results were obtained by V ä n n i n e n and H o k k a n e n (1988) for Oxamyl. At this dosage Fastac only slightly inhibited the growth of fungi. These findings are accordance with those of B a j a n, F e d o r k o, K m i t o w a (1986). V ä n n i n e n and H o k k a n e n (1988) found no changes in the growth of fungi when pyretroid Ripcord was applied.

According to I g n o f f o et al (1975) the insecticides of Azodrin and Fumazone at concentrations ten times lower than the recommended dose do not affect the growth of *Nomuraea rileyi*. In our experiment the investigated fungi species did not react to this dose of preparation as well. In the present experiment, the effect of insecticides on the growth of particular (specific) isolates of fungi was also determined. The most significant differences in the size (growth rate) of colonies were found in the case of *B. bassiana* isolates; the colonies of isolate n° 1 of this species derived from the forest soil were always bigger than those of isolate n° 2 obtained from the arable soil.

Olmert and Kenneth (1974) indicated that Itopaz and Ravion were very toxic to two of the five investigated *B. bassiana* isolates. In the studies conducted by Anderson and Roberts (1983) the growth of one of the six *B. bassiana* isolates was inducted by Carbaryl 50 WP and Oxamyl 2 EC, whereas the growth of the remaining isolates was inhibited. These authors compared the isolates of fungi collected from different hosts. In the present experiment, the isolates were derived from various habitats.

On the medium with insecticides, the colonies of *M. anisopliae*, *P. farinosus* and *P. fumosoroseus* isolates were bigger than those of isolates derived from forest soil although the differences were not pronounced as in the case of *B. bassiana*. Bajan (1985) and Bajan, Fedorko, Kmitowa (1986) found the strains of *P. farinosus* reacting differently in the presence of Fastac in the medium.

From the results obtained in the present study and fragmentary data from other authors, it may be inferred that the growth rate of isolates of entomopathogenic fungi depends not only on the origin of isolates but is also associated with the host and type of habitat.

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