PHOSPHINE RESISTANCE IN STORED PRODUCT INSECTS IN THE UNITED STATES

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ABSTRACT

Feral strains of stored product insect pests collected from various commodity groups stored throughout the United States were tested for resistance to phosphine. Discriminating dosage tests showed that phosphine resistance was found in all species tested. Frequencies of resistance among individual strains varied greatly; however, strains of Rhyzopertha dominica (F.) from wheat showed the greatest variations and strains of Cadra cautella (Walker) from peanuts and Tribolium confusum Jacquelin duVal from flour showed the smallest variations. Overall, the moths were the most resistant, followed by R. dominica. Resistance does not appear to be sufficiently high to cause control failures and proper fumigation of commodities infested with these resistant strains should provide complete control.

INTRODUCTION

Prior to the FAO survey of pesticide susceptibility of stored grain pests (Champ and Dyte, 1976), phosphine resistance in stored food insects was unknown except from several artificially selected laboratory strains. From that survey, phosphine resistance in the U.S. was measured in 1 strain of Tribolium castaneum (Herbst), 1 strain of Oryzaephilus surinamensis (L.), and 2 strains of Tribolium confusum Jacquelin duVal. Although stored product Lepidopterans were not tested in this survey, Zettler (1983) first reported that phosphine resistance was evident in strains of Plodia interpunctella (Hubner) and Cadra cautella (Walker) collected from peanut warehouses in the Southeastern United States. Control failures attributed to phosphine resistance began to occur about this time in other parts of the world (Borah and Chahal, 1979; Tyler et al., 1983).

Most surveys of pesticide resistance in the U.S. have been concerned with residual insecticides and not fumigants (Bansode and Campbell, 1979; Beeman et al., 1982; Zettler 1982; Haliscak and Beeman, 1983; Horton, 1984; Halliday et al., 1988; Arthur et al., 1988). However, with the cancellation of liquid fumigants for grain fumigation in 1984-1985, phosphine use has increased in the United States. Consequently, one would expect that the phosphine resistance spectrum had changed significantly in recent years. Thus, several studies were initiated to monitor phosphine resistance in stored product pests in the U.S. This paper presents a partial summary of these studies.

MATERIALS AND METHODS

Insects

The insects used in these tests were field strains of the major pest species collected from storage facilities containing various commodities. Indianmeal moth, P. interpunctella (Hubner), almond moth, C. cautella

(Walker), and red flour beetle, T. castaneum Herbst were collected from storage facilities in Georgia, Florida and Alabama; lesser grain borer, Rhyzopertha dominica (F.) and red flour beetle were collected from farm storage bins containing wheat in Oklahoma; red flour beetle and confused flour beetle, T. confusum Jacquelin duVal, were collected from flour mills throughout the entire country; and the cigarette beetle, Lasioderma serricorne (F.), was collected from tobacco warehouses in the Southeastern U.S. These feral strains were brought to the Stored Product Insects Research and Development Laboratory and reared on the appropriate diet (Boles and Marzke, 1966) at 27°C and 60% RH until sufficient numbers were available for testing.

Probit Analysis Tests

Probit regression lines (SAS Institute, 1987) for phosphine were calculated for each susceptible laboratory strain in 2.5-liter dessicator jars (Anon., 1975) according to the procedure of Zettler et al. (1989). To produce the various required concentrations of phosphine, jars were dosed with the appropriate volume of gas from a cylinder containing 495 ppm (v/v) phosphine in nitrogen. Actual concentrations in the treatment jars were measured using a Varian Model 3700 gas chromatograph equipped with a flame photometric detector in the phosphorus mode (Zettler et al., 1989).

Discriminating Dosage Tests

The presence of resistance to the pesticides was determined by the discriminating dosage (DD) method. This method involves testing a population with a dose of pesticide, called the discriminating dosage, which will kill all susceptible individuals in a population but none of the resistant ones. The proportion of survivors following such treatment gives as estimate of the frequency of resistance in that population. The DD for each pesticide was estimated from its corresponding probit regression line. Adults were tested in 2.5-liter dessicator jars (Zettler et al., 1989). For each DD test, at least 100 adults of each strain were tested and the resulting mortalities were recorded 1 wk after treatment. Data were subjected to Chi-Square analysis (SAS Institute 1987).

RESULTS AND DISCUSSION

Response to Discriminating Dosages

The toxicity data shown in Table I agree with earlier data for the confused flour beetle (Lindgren and Vincent, 1966; Attia and Greening, 1981), and the cigarette beetle and red flour beetle (Lindgren and Vincent, 1966). Data for the lesser grain borer differed but only by a factor of less than 2X at the LD99.9. The data for Indianmeal moth differed in the slope of the regression line by 13X at the LD99.9. Generally, phosphine was more toxic to the susceptible Savannah laboratory strains than to the susceptible strains tested by Lindgren and Vincent (1966) and Attia and Greening (1981).

Table I. Toxicity and discriminating dosages of phosphine-susceptible laboratory strains of stored product insects to phosphine.

Species	Reference	No. insects	Slope±S.E.	LD50 (95%CL)	LD99 (95%CL)	DD
T. castaneum	1	900	7.18±0.37	4.4 (4.2-4.5)	9.2 (8.6-10.2)	10
T. confusum	2	550	8.27±0.35	7.2 (6.9-7.5)	14.0 (11.9-18.2)	20
R. dominica	1	1500	5.56±0.42	2.4 (2.3-2.5)	6.6 (5.7-8.0)	10
L. serricorne		550	3.79±0.51	5.1 (4.7-5.5)	21.0 (16.6-29.5)	25
P. interpunctella	3	480	7.86±0.36	3.7 (3.6-3.8)	7.3 (6.8-8.0)	10
C. cautella	3	480	8.40±0.35	7.2 (6.6-7.8)	13.6 (11.7-17.4)	15

References:
1. Zettler and Cuperus (in press)
2. Zettler (in press)
3. Zettler et al. (1989)

The DDs reported in Table I are lower than those which have been arbitrarily set at some multiple of the LD99 or LD99.9 (Suckling et al., 1987; Subramanyam et al., 1989). It has been shown that a DD is not necessarily diagnostic when it is arbitrarily set at such a dose (Halliday and Burnham, 1990). Indeed, these authors showed that the power and accuracy of the DD test decrease as mortality of the susceptible strain approaches 100%. Thus, the DDs reported here, although relatively low, may indeed be too high to detect resistance in some strains.

Nevertheless, phosphine resistance was detected in all species (Table II). Lesser grain borer showed the highest frequency of resistance. This is the first instance of phosphine resistance in the lesser grain borer in the U.S. Two-thirds of the strains tested were resistant to phosphine; of these, half showed that 50% or more of the individuals were resistant. Conversely, red flour beetles collected along with lesser grain borer were among those pests with the lowest incidence of resistance to phosphine (1 of 8 strains), and the frequency of resistance in that strain was only about 6%. The two moths and the red flour beetles collected from peanuts showed a high frequency of resistance. Of the resistant Indianmeal moth and red flour beetle, frequencies ranged from 8-22% and 4-36%, respectively. Of the two pests from flour mills, the red flour beetle showed the greater resistance with nearly half the strains being resistant. Only 3 of 17 strains of confused flour beetle were resistant and of those, the highest frequency was 7%. Only 1 of 9 strains of the cigarette beetle was resistant and this strain had a resistance frequency of 23%.

Table II. Phosphine resistance in strains of six species of insect pests collected from various commodities stored in the United States

Commodity	Reference	Species	No. species tested	No. strains resistant	Range of response	Percent strains resistant
Peanuts	1	P. interpunctella	7	4	(77-92)	57
		C. cautella	8	3	(93-96)	17
		T. castaneum	23	8	(64-96)	35
Wheat	2	R. dominica	12	8	(8-92)	67
		T. castaneum	8	1	(94)	13
Flour	3	T. castaneum	28	13	(45-96)	46
		T. confusum	17	3	(93-94)	18
Tobacco		L. serricorne	9	1	(77)	11

References:
1. Zettler et al. (1989)
2. Zettler and Cuperus (in press)
3. Zettler (in press)

Control Failures and Discriminating Dosages

Control failures following fumigation may not be caused by resistance. In many cases where survivors are found following treatment, the fumigation was done improperly, such as inadequate sealing of structures (Mills, 1983; Banks and Annis, 1984; Dyte and Halliday, 1985; Zettler et al., 1986; Zettler et al., 1989). Control failures due to resistance following a properly conducted fumigation, however, depend upon both the presence of genes which confer resistance and the degree to which resistance is conferred by those genes.

The percentage of survivors of a DD test reflects only the frequency of genes for resistance in that population and tells nothing at all about the level of resistance in the population. Genes which confer resistance may occur frequently in a population without causing control failures. Thus, a strain with high frequency of resistance might be susceptible to normal treatments with the pesticide to which it's genes confer resistance. Conversely, a strain with low frequency of resistant individuals could be difficult to control if the few genes for resistance conferred extremely high levels of resistance. Thus, while the laboratory DD test is beneficial in detecting early development of resistance, it plays little or no role in predicting control failures in the field.

Indeed, it would be extremely useful if control failures could be correlated with a simple diagnostic assay for resistance. However, the relationship between control failures and frequency of resistance genes in a population is not known. Thus, one cannot make pest control decisions based solely on the results of resistance studies from DD tests. In fact, results of DD tests measuring resistance gene frequency in field strains and actual control efficiency of pesticides to resistant strains could not be correlated in a number of Tribolium sp. strains (Arthur and Zettler, unpublished results), although Denholm et al. (1984) successfully correlated these parameters in the housefly. However, it is likely that direct correlations of this nature will require evaluation of not only results of DD tests but also other factors including the level or intensity of resistance found in a particular population.

Intensity of Phosphine Resistance

In spite of the relatively high frequencies of resistance reported in these pests, only small increases in the fumigant dose were required to force the populations to extinction. For example, Table III shows that all individuals in phosphine resistant strains of confused and red flour beetles were killed with doses of 4X the DD; most succumbed to doses lower than 4X.

Table III. Mortality of phosphine-resistant strains of flour beetles at multiples of the discriminating dosages *

Strain	DD	Mortality (percent) at multiple of discriminating dosage			
		1.5	2	3	4
		Tribolium castaneum			
1	45	62	99		
2	80	100			
3	94	100			
4	74	90	98		
5	46	79	89	95	100
6	88	93	98		
7	96	99			
		Tribolium confusum			
8	94	100			
9	93	100			

* From Zettler (in press)

On the other hand, toxicity data for phosphine resistant strains of the moth species showed that these populations were more resistant than were the beetles. For example, probit regression lines for strains of phosphine resistant Indianmeal moths (Table IV), extrapolated to the LD99.9, showed that a dose 4X the DD would kill all individuals in the resistant populations, except for the G2 strain which would require about a 6X dose. Similar data for strains of almond moths showed that 2 of the 3 resistant strains of this insect would be killed by small increases in the DD, but the G1 strain would require about a 13X dose to kill all individuals. This level of resistance might indeed cause control failure with phosphine.

Table IV. Toxicity of phosphine to resistant strains of Indianmeal moth, Plodia interpunctella, and Almond moth, Cadra cautella.

Strain	No.	Slope±S.E.	LD50 (95%CL)	LD99 (95%CL)
			Plodia interpunctella	
G1	1765	3.15±0.31	3.5 (3.3-3.7)	19.0 (16.9-21.8)
G2	300	2.71±0.52	4.5 (3.7-5.6)	32.5 (24.6-42.8)
A1	420	3.63±0.41	5.2 (4.9-5.8)	23.1 (18.0-31.4)
A2	480	7.57±0.56	10.1 (9.6-10.5)	21.2 (17.4-28.8)
Lab*	480	7.86±0.36	3.7 (3.6-3.8)	7.3 (6.8-8.0)
			Cadra cautella	
G1	400	2.62±0.28	12.3 (10.5-14.7)	95.4 (87.3-104.6)
G3	400	11.07±0.47	19.1 (17.7-24.5)	34.8 (30.5-38.6)
A3	480	11.98±0.58	16.2 (14.0-19.1)	24.8 (22.2-28.1)
Lab*	480	8.40±0.35	7.2 (6.6-7.8)	13.6 (11.7-17.4)

*From Zettler et al. (1989)

CONCLUSIONS

The data reported and summarized here indicate that low level phosphine resistance is rather widespread among stored product pests in the United States. For the most part, severity of resistance appears to be small in spite of the fact that frequencies of genes for resistance are high in some populations. High frequencies notwithstanding, the phosphine resistances reported here are not as severe as those contributing to control failures in Pakistan (Taylor and Halliday, 1986), Bangladesh (Mills, 1983), and Australia (Attia and Greening, 1981). The control failures reported to be associated with phosphine resistance in the U.S. are more properly attributed to improper sealing of structures prior to fumigation or to some other inadequate fumigation procedure. The rather large resistance ratios for the moths notwithstanding, proper fumigation utilizing adequate sealing and other procedures to insure proper levels of phosphine for at least 5 days should provide adequate control of these and other phosphine resistant species.

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Footnote

1/ This paper reports the results of research only. Mention of a pesticide in this paper does not constitute a recommendation for use by the USDA nor does it imply registration under FIFRA as amended. Mention of a proprietary product does not constitute an endorsement by the USDA.

RESISTANCE A LA PHOSPHINE DES INSECTES DES DENREES STOCKEES AUX ETATS-UNIS

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RESUME

Des souches sauvages d'insectes des denrées stockées ont été récoltées dans différentes denrées présentes d'un bout à l'autre des Etats-Unis afin de mesurer leur résistance à la phosphine et à d'autres pesticides. Les mesures de doses discriminantes et les analyses de régression dose/réponse des données montrent que la résistance à la phosphine se développe rapidement et qu'elle peut grandement contribuer à l'échec des méthodes de lutte.