SOME CONSEQUENCES OF THE DEVELOPMENT OF INSECTICIDE RESISTANCE IN STORED-PRODUCT INSECTS

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ABSTRACT: Resistant strains of 18 insects infesting stored products are known, and in the United Kingdom 9 species are resistant to lindane and 7 species resistant to malathion. Discriminating dose tests are suitable for surveying the problem, and regular monitoring for resistance in certain species may be valuable in some areas e.g. malathion resistance in *Oryzaephilus surinamensis* in Britain. Simple bait traps are useful for collecting living insects. Fumigant resistance is rare in field populations so fumigants remain of value against most resistant strains. A resistance specific to malathion is known in *Tribolium castaneum*, *T. confusum* and *Rhyzopertha dominica*. It can be detected by using triphenyl phosphate as a malathion synergist, and such strains can be controlled by alternative organophosphorus compounds. Non-specific organophosphorus resistances occur in at least seven species, and the patterns of cross-resistance to other insecticides are different in the different species. These strains with non-specific organophosphorus resistance present the most difficult control problems.

INTRODUCTION: The widespread occurrence of insecticide-resistant strains of 8 storage pests has been revealed by the global survey set up by F.A.O. in 1972. At present, only a preliminary report of that survey is available[1] but this, and other sources[2,3,4] indicate that resistance occurs in at least 13 species of beetles, 5 moths and 2 mites which attack stored products. The changes in control measures necessitated by the occurrence of resistant strains will vary in different countries according to differences in storage facilities and practices. To some extent the problems with exports and imports are different. Our experience inevitably reflects conditions in the U.K., an island which imports much of its food. In the U.K. we have had a well-trained inspectorate examining infestation in imports for over 25 years. We know much about our infestation problems, where they are coming from, and how they are changing[5]. However, as yet we have limited experience of dealing with resistant strains in the field. On the other hand, resistance problems must be diagnosed in the laboratory because there are many other possible causes of control failure. We hope our laboratory studies may be of interest to workers from countries whose storage problems differ from those of the U.K.

The problems of insecticide resistance can be completely avoided if non-chemical methods of control are used. We shall not consider these methods as they will be discussed elsewhere at this

Conference. In any event non-chemical control should always be the first line of defence against storage pests and only when they are not feasible should insecticides be used. We ought not to be involved with insecticide resistance problems if non-chemical approaches are possible.

RESISTANCE SURVEYS: The dissemination of resistant strains through International Trade is well established[1,6,7]. The F.A.O. Survey has given a broad picture of the distribution of resistant strains, but relatively few strains of each species were examined from each country, and the need to survey the extent of resistance in individual countries and industries remains. Some such surveys have been done[2] but several of these need updating. Some results from surveys made in the U.K. during 1973 are shown in Tables I and II in relation to the known occurrence of malathion and lindane resistance in other countries. We were concerned with the 8 species studied in the F.A.O. Survey and none of the samples came from imports on ships. Few strains of *Oryzaephilus mercator*, *Rhyzopertha dominica*, and *Sitophilus zeamais* were obtained as these species are not commonly found in established infestations inland in the U.K. However, strains resistant to lindane were found in all 8 species, and with four the percentage of strains found to be resistant was high: - *T. castaneum* 86%, *O. surinamensis* 77%, *S. oryzae* 80%, and *S. zeamais* 100%. With malathion the situation was less depressing. Only in the two species of *Tribolium* was the incidence of resistance high (*T. castaneum* 83%, *T. confusum* 50%) and in the two species of *Oryzaephilus* no malathion-resistant strains were detected.

TABLE I. Occurrence of Lindane Resistance in Stored Product Insects and Incidence in the U.K. in 1973.

Species	Countries with resistant strains*	UK situation in 1973	
		Strains tested	Strains resistant
Tribolium castaneum (Herbst)	71	28	24
Tribolium confusum Duv.	16	23	5
Oryzaephilus surinamensis (L.)	36	39	30
Oryzaephilus mercator (Fauv.)	7	7	3
Rhyzopertha dominica (F.)	32	8	2
Sitophilus granarius (L.)	11	43	2
Sitophilus oryzae (L.)	48	15	12
Sitophilus zeamais Motsch.	31	6	6
Dermestes maculatus Deg.	UK	not examined	
Caryedon serratus (Oliv.)	Gambia	"	"
Sitotroga cerealella (Oliv.)	Ceylon	"	"
Phthorimaea operculella (Zell.)	Australia	"	"

* Data from [1] supplemented by [8] [9] [10] & [11]

TABLE II. Occurrence of Malathion Resistance in Stored Product Insects and Incidence in the U.K. in 1973.

Species	Countries with resistant strains*	UK situation in 1973	
		Strains tested	Strains resistant
Tribolium castaneum	70	29	24
Tribolium confusum	25	24	12
Oryzaephilus surinamensis	8	52	nil
Oryzaephilus mercator	6	7	nil
Rhyzopertha dominica	25	7	2
Sitophilus granarius	5	43	3
Sitophilus oryzae	12	16	1
Sitophilus zeamais	UK	6	1
Dermestes maculatus	UK	not examined	
Trogoderma granarium Everts	Tunisia	"	"
Latheticus oryzae Waterh.	1	"	"
Plodia interpunctella (Hub.)	USA	"	"
Ephestia cautella (Walk.)	USA	"	"

* Data from [1] supplemented by [3] [8] [12] & [13]

These results, like those in the F.A.O. survey, were obtained by discriminating dose tests. That is, each strain is tested with a dose which is known to knock down all members of a susceptible strain. Those yielding survivors are classed as resistant. This type of test has several advantages over the determination of LD50 values when the aim is to detect rather than measure resistance [14], and it can also be undertaken with smaller samples. The reliability of these tests can be checked in several ways. One way is to keep any survivors and breed from them. If the resulting progeny are tested at the same dose level we might reasonably expect more of them to survive. Table III shows some examples of this. We can also measure the level of resistance by obtaining probit mortality lines. Some examples from tests on *Oryzaephilus surinamensis* are given in Table IV. We used 8 strains from 7 different countries which had yielded survivors in a discriminating dose test. These 8 strains were maintained in culture without selection and the discriminating dose tests were later repeated and also probit mortality lines obtained and LD50s calculated. Not only did all the repeated tests confirm the presence of resistance but there is a reasonable correlation between the rate of survival in the discriminating dose tests and the resistance factors obtained. Results like these give us confidence in the value of discriminating dose tests in surveys of the incidence of resistant strains.

REGULAR MONITORING FOR RESISTANCE: Resistance surveys delineate the extent of the problem. It may be worth keeping a continual watch on the situation in some species by regular monitoring for resistance, but in others this may be a waste of time.

TABLE III. Responses Obtained With Adult Beetles in Discriminating Dose Tests Together With Those Obtained With the F_1 Progeny of the Survivors.

Species and Origin	Insecticide	Per cent knock-down Parents	F_1 progeny of survivors
Tribolium castaneum			
Australia	lindane	94	45
Zambia	"	84	35
Australia	malathion + TPP*	17	3
Malawi	"	96	23
Tribolium confusum			
Poland	malathion	97	65
Senegal	"	87	27
UK	"	73	60
Oryzaephilus mercator			
Italy	malathion	67	20
Swaziland	"	73	63

*TPP = triphenyl phosphate

TABLE IV. Comparison of Responses in Discriminating Dose Tests With Resistance Factors in Eight Malathion-Resistant Strains of *Oryzaephilus surinamensis*.

Strain	Per cent knock-down in discriminating dose tests	Resistance Factor
Susceptible	100, 100	x1
Asia - 1	6, 5,	x43
Asia - 2	2, 5,	x27
Asia - 3	52, 32,	x7
Asia - 4	86, 42,	x7
Asia - 5	88, 96, 70	x6
South America - 1	95, 98, 89,	x4
South America - 2	86, 94, 77,	x3
South America - 3	99, 99, 94	x3

In the UK the high incidence of malathion and lindane resistance in *T. castaneum* (Tables I and II) is comparable to the high incidence of both these resistances found in this species in other countries during the F.A.O. Survey[1]. We know the vast majority of strains of this species arriving on imports are resistant to both these compounds[6] so regular monitoring for resistance in this species is no longer of any value. However, monitoring the type of malathion resistance present can be useful as described below.

The situation with *Oryzaephilus surinamensis* is very different. This is the most important pest of home-grown grain in the UK. We rely on malathion for control and the prospect of malathion-resistance occurring is very disturbing. In addition to the 52 samples tested in 1973 and found to be malathion-susceptible (Table II) a further 38 samples from inland sites were examined in the first half of 1974 and again all were malathion-susceptible. This indicates that we do not have malathion-resistant strains of *O. surinamensis* on our farms. However, this species is regularly imported into the UK, and in view of the risk of importing malathion-resistant strains which might become established we are regularly monitoring for resistance in all imported strains which are detected.

So far we have detected malathion-resistance in two strains of *O. surinamensis* from ships. Both were on Indian vessels bringing cargoes from that country. In the first case the infestation was in a ship store and did not involve goods to be unloaded. This did not create any problems. The second case, however, involved a mixed cargo and by the time the insects had been found to be malathion-resistant most of the goods had been unloaded into a port warehouse and some had been distributed. Wherever possible these goods were fumigated with methyl bromide. However, a lot of work in tracing imported goods and visits to premises inland were necessary so as to ensure that all infested cargo had been dealt with.

USE OF BAIT TRAPS: For collecting living insects for resistance testing we have found simple bait traps very useful. These consist of plastic mesh bags, about 10x20 cm, made of nylon with 1.5 mm apertures. Margins are heat-sealed except for one short edge which is closed with staples (those normally used for papers) when the bag has been filled with bait. An effective bait for most storage insects consists of one part carobs (locust beans), one part broken groundnuts, and one part wheat. Before use the baited traps are sterilized for 6 hours at 70°C. This ensures the initial absence of living insects, and tends to bring out the aromatic flavour of the carobs. The trap bags can be placed in warehouses, ships' holds etc, and collected after one or preferably two days. They can then be shaken over a tray (without being opened) and the living beetles drop on to the tray. Bait traps can be used more than once provided they are sterilized between each usage. They are also of value for detecting survivors of treatments.

CROSS-RESISTANCE STUDIES: When resistant strains are detected it may be necessary to use an alternative means of control. This is not always the case if disinfestation rather than protection is required. The levels of insecticide mixed with grain to give a prolonged protection in store are often more than sufficient to eliminate existing infestation. If resistance is not at a high level, its effect is likely to be a shortening of the period of protection in store rather than immediate control failure. This, of course, is why resistance is often not detected in the early stages of its spread.

On the other hand there are many instances where storage pests can tolerate doses far in excess of those permitted by international and national regulatory authorities. There are also many situations where admixture is impossible or unacceptable to trade interests, and the control of cross-infestation by fabric treatments may soon fail when resistance develops. In these cases a change in pesticide is essential and the extent of resistance to other pesticides is important. In this respect, it is helpful to distinguish true cross-resistance, where a single resistance mechanism confers protection to several compounds, from the multiple resistance which may occur when a particular strain has developed independent mechanisms for resisting several insecticides. Much more work is required on the cross-resistance of storage pests. Such knowledge as we have is in most cases based on very few strains from only one or two countries. For example, the spectrum of resistance to other compounds in only one malathion-resistant strain of *Oryzaephilus mercator* has been studied[15]. This strain came from Senegal, and it would be very unwise to advise those concerned with malathion-resistant strains of this species in Italy or Swaziland to accept the work on the West African strain as relevant to their local problems.

In view of this, and since some aspects of the subject are discussed elsewhere[2], we propose only to consider a few tentative generalisations which may have fairly wide applicability.

Resistance to fumigants in field populations is rare[1,16]. From the data available there is no reason to suspect that resistance to phosphine inevitably involves resistance to methyl bromide or vice versa. Moreover, it seems that fumigant-resistant strains may be fully susceptible to lindane or malathion. They may not be, of course, if the incidence of malathion or lindane resistance is very high as it is in *Tribolium* spp.[1], but this appears to be due to multiple resistance rather than a mechanism governing resistance to both fumigants and insecticides. Since fumigant resistance is rare, and there is no reason to suspect that resistance to the commoner contact insecticides confers any resistance to fumigants we can continue to use methyl bromide or phosphine for the control of most strains resistant to contact insecticides.

The cross resistance of organochlorine-resistant strains of storage pests had been little studied[2], probably because few organochlorine compounds except lindane are suitable for use on grain intended for food (rather than as seed for next year's crop), and even lindane is not permitted in some countries. With all eight species covered by the F.A.O. Survey, strains resistant to lindane and not to malathion were found, and this is also the case with *Dermestes maculatus* [8]. It seems therefore that mechanisms governing resistance to lindane and not malathion exist in all these species.

There is, however, some circumstantial evidence indicating that a mechanism governing resistance to both malathion and lindane may occur in *Oryzaephilus* spp. We have tested 45 strains of *O. mercator* and 150 strains of *O. surinamensis* for both malathion and lindane resistance, and the incidence of lindane resistance was 62% and 73% respectively. The incidence of malathion resistance was lower, being respectively 29% and 9% in the two species. Now, on the basis of the overall incidence of lindane resistance we should expect that about two-thirds of the malathion-resistant strains would also be lindane resistant and about one-third of them lindane susceptible. In fact all of the malathion-resistant strains were also lindane resistant, and we failed to detect any strains which were resistant to malathion alone. This can be interpreted in several ways, but it seems possible that in both of these species resistance to malathion can confer a true cross resistance to lindane. This possibility would appear to merit further investigation.

USE OF TRIPHENYL PHOSPHATE: The most extensive studies on malathion resistance have been made with *T. castaneum*. It has been possible to distinguish a malathion-specific type of resistance which gives a cross resistance only to malathion analogues, from another type or types with a very wide spectrum of cross resistance involving organochlorines, organophosphates, carbamates, pyrethroids, juvenile hormone analogues, and even the synergist piperonyl butoxide[7,17,18,19]. The malathion-specific type of resistance is completely suppressed by the synergist triphenyl phosphate[20] and this can be used in the diagnosis of resistance[6,21,22]. When malathion resistance is detected it can be useful to determine whether or not the malathion-specific type of resistance is present because with strains of this type most commercially available organophosphorus compounds are fully effective[23].

In the UK malathion resistance is widespread in this species but the malathion-specific type predominates. In 1969-70, tests on 17 strains intercepted on imports showed that 15 were malathion-resistant but only one of these was not of the malathion-specific type[6]. With UK strains collected in 1973, 23 of 28 tested were malathion resistant (Table II) and all but four of these had the malathion-specific type of resistance. Under these circumstances we find it well worthwhile undertaking resistance tests with malathion synergized by triphenyl phosphate before deciding on control recommendations.

Because this test has been valuable with *T. castaneum* it is reasonable to examine whether it can be applied to other species. Resistances which appear to be malathion-specific and which are overcome by the synergist triphenyl phosphate have also been found in *Tribolium confusum* and *Rhyzopertha dominica*. However, in other malathion-resistant strains of *T. confusum* and in strains of *Sitophilus oryzae*, *S. granarius*, *S. zeamais*, *Oryzaephilus surinamensis* and *O. mercator* malathion resistance is associated with resistance to other organophosphorus compounds[15,24] and is not overcome by the synergist triphenyl phosphate.

The interpretation of tests with malathion synergized by triphenyl phosphate is complicated by the fact that this compound is an effective synergist for malathion in susceptible strains of some species e.g. *Dermestes maculatus*[25]. This means that the toxicity of malathion to a resistant strain in the presence of the synergist may be unrelated to the resistance mechanism. This is illustrated by data on *Rhyzopertha dominica* in Table V. At the LD90 level there is a fivefold synergism of malathion by triphenyl phosphate in the susceptible strain. This means that the discriminating dose used for malathion needs to be reduced by this factor for the accurate determination of non-specific malathion resistance in this species[26].

TABLE V. Synergism of Malathion by Triphenyl Phosphate in Susceptible and Malathion-Resistant Strains of *Rhyzopertha dominica*.

	Susceptible	Resistant	Factor of Resistance
	LD50 Value		
malathion	0.58	1.31	x2
malathion + TPP	0.13	0.11	x<1
factor of synergism	x4	x12	
	LD90 Value		
malathion	1.23	2.91	x2
malathion + TPP	0.21	0.19	x<1
factor of synergism	x5	x15	

NON-SPECIFIC ORGANOPHOSPHORUS RESISTANCE: While malathion-specific resistance can be distinguished from non-specific organophosphorus resistance we do not have any means as yet for distinguishing the different types of non-specific resistance which may occur. The cross resistance spectra obtained with different species[2] show few similarities, and because few strains have been investigated we cannot be certain whether more than one mechanism of non-specific resistance occurs in any given species. Many of these non-specific resistant strains show some resistance to all the contact insecticides currently permitted or recommended by such international bodies as the F.A.O. Codex Alimentarius, the International Marine Consultative Organization and the European Economic Community.

In the UK *Tribolium castaneum* currently presents problems of this type. This species is of increasing importance as a pest in maltings and mills producing flour or animal foodstuffs. Most strains are resistant, and when the non-specific type of resistance occurs no permitted contact insecticides are fully effective. With fabric treatments insects are normally in contact with treated surfaces for very short periods, but to kill resistant strains very long exposures to high doses are required (Table VI). The problem is made even more difficult by changes in the behaviour of the insects[27].

This outline of the way in which resistance has affected our work in the UK shows that we have not got answers to all our problems. We hope, however, that some of our experiences will be of value to those dealing with resistance problems in other countries. Since the UK imports much of its food, the resistance problems of other countries are likely to become those of the UK in the course of time.

TABLE VI. Mortality of Susceptible and Resistant Strains of *Tribolium castaneum* Five Days After Short Exposures on Wettable Powder Formulations of Malathion or Fenitrothion.

	Time Exposed (Hours)	Per cent mortality Susceptible	Resistant
Malathion	2	100	21
at 1600 mg/m^2	6	100	47
	24	100	95
Fenitrothion	2	100	39
at 950 mg/m^2	6	100	94
	24	100	100

ACKNOWLEDGMENTS: We are grateful for help from many members of the laboratory and regional staff of the Ministry's Pest Infestation Control service, and particularly to Miss D. G. Blackman, P. M. Cogan, Mrs. R. Forster and K. B. Wildey.

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