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MALATHION RESISTANCE IN LARVAE OF SOME SOUTHERN MINNESOTA POPULATIONS OF THE INDIANMEAL MOTH, PLODIA INTERPUNCTELLA (LEPIDOPTERA: PYRALIDAE), INFESTING BULK-STORED SHELLED CORN

W. A. Sumner II, P. K. Harein, and Bh. Subramanyam

ABSTRACT

Larvae of 21 field collected populations of the Indianmeal moth, Plodia interpunctella, infesting stored shelled corn in southern Minnesota were tested for their susceptibility to malathion in the laboratory. A population that was a composite of the 21 populations and a malathion susceptible population were also tested for their susceptibility to malathion, pirimiphos-methyl and chlorpyrifos-methyl. Comparison of the LD₅₀ values of the field populations with the malathion susceptible population indicated that the field populations were ca. 33- to 625-fold resistant to malathion. The composite field population was ca. 243-fold resistant to malathion, and this population was 3.2-fold cross-resistant to pirimiphos-methyl, but was highly susceptible to chlorpyrifos-methyl.

The Indianmeal moth, Plodia interpunctella (Hübner), is a destructive pest of stored-grain whose larvae cause damage by feeding on the germ and endosperm of grain (Demianyk & Sinha 1981, Madrid & Sinha 1982). Wandering last instar larvae of P. interpunctella produce abundant webbing on the grain surface, and in severe infestations the entire grain surface is covered with a layer of webbing. In Minnesota, P. interpunctella is the third most commonly occurring insect infesting stored shelled corn (Barak & Harein 1981). The severity of this pest in the United States is compounded by the development of resistance in larvae to malathion, an organophosphate insecticide registered for use on grain since the late 1950's (Zettler et al. 1973, Bansode et al. 1981, Beeman & Schmidt 1982, Beeman et al. 1982, Zettler 1982).

Zettler et al. (1973) reported about 206-fold malathion resistance in P. interpunctella larvae infesting peanut- and grain-storages in Georgia, Alabama, Florida, Kansas, and Illinois. Armstrong & Soderstrom (1975) reported malathion resistance in 3 of 5 field populations of P. interpunctella infesting dried fruits and nuts in California, and in the laboratory they showed the stability of this resistance in the absence of malathion selection. Bansode et al. (1981) reported > 227-fold resistance to malathion in a composite field population of P. interpunctella. Beeman et al. (1982) found > 17-fold malathion resistance in 39 of the 43 populations of P. interpunctella collected from grain storages in the northcentral United States. Ten of 12 populations of P. interpunctella infesting peanuts in the Southeastern United States were > 114-fold resistant to malathion (Zettler 1982).

Some P. interpunctella populations collected from grain storages in Minnesota were tested for their resistance to malathion (Beeman & Schmidt 1982, Beeman et al. 1982).

1 Mention of a proprietary product does not constitute an endorsement by the University of Minnesota.
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Beeman & Schmidt (1982) tested susceptibility of 13 field populations of *P. interpunctella* from Minnesota to malathion at a discriminating dose of 20 μg per larva (LD₅₀ × 4). Mortality of 11 of the 13 populations varied from 0–69% indicating resistance. Two populations were susceptible showing 91–100% mortality.

In this paper, we report on the resistance to malathion in several populations of *P. interpunctella* larvae infesting shelled corn in southern Minnesota. In addition, we evaluated cross-resistance of a composite field population to two new organophosphate grain protectants—pirimiphos-methyl and chlorpyrifos-methyl.

**MATERIALS AND METHODS**

**Grain sampling and insect collection:** Corn storages located in Cottonwood, Dakota, Redwood, Scott, Steele, Stevens, Swift, and Waseca counties were visited during the summer of 1981 to remove grain samples for collecting *P. interpunctella*. Corn samples were removed with a 1.7 m, 12 compartment trier, and a deep cup probe. In each storage, 6 locations were probed with a trier vertically (4 in each of the N, S, W, E cardinal directions & 1 near storage center) and horizontally (1 sample from an undisturbed area of storage). The middle of the grain mass in the center of storage was sampled with a deep cup probe using 1 m extensions. Grain samples collected from different locations within a storage were pooled to form a composite sample. Composite samples were then placed in 0.95-liter plastic jars fitted with air-tight lids. Samples brought back to the laboratory were sifted through a 0.48-cm diam. sieve to separate larval, pupal, or adult stages of *P. interpunctella*. At the time of grain sampling, any live wandering last instar larvae and/or adults of *P. interpunctella* on the grain surface and storage walls were collected; populations collected from each storage were placed in separate 0.946-liter jars containing *P. interpunctella* rearing diet (see below).

**Insect rearing:** A turkey-mash diet (Subramanyam & Cutkomp 1987) was used to rear *P. interpunctella*. *Plodia interpunctella* populations from each storage were reared separately by seeding the diet with live larvae or adults. A few larvae from each storage were pooled and reared on the diet. This population was the composite of all field populations. We were successful in maintaining 21 field populations and a composite field population. In addition, a known malathion susceptible population of *P. interpunctella* obtained from the Stored-Product Insects Research and Development Laboratory, Savannah, Georgia, was reared similarly. *Plodia interpunctella* cultures were held in a controlled temperature and humidity (CTH) chamber set at 25°C and 75% RH. Insects were reared for a minimum of 3 generations before testing. Wandering last instar (5th instar) larvae of *P. interpunctella* weighing about 14 ± 2 mg were used in bioassays.

**Bioassays:** Larvae of *P. interpunctella* were treated topically with malathion (93% purity) using a microtopical applicator. Individual larvae were treated on the dorsum with 1 μl amounts of malathion solutions in acetone or acetone alone (control). A minimum of 5 dosages was used for each population. Three groups (replicates) of 15 larvae each were treated at each dosage, and each group after treatment was placed in a 9 cm diam glass petri dish fitted with a wet filter paper (9 cm diam). Petri dishes with larvae were then held in the CTH chamber for 96 h posttreatment, after which each petri dish was observed for dead larvae. Larvae that failed to move when prodded gently with a fine camel brush were recorded as dead. Mortality was calculated from the proportion of larvae dead out of the total exposed (p), and was expressed as a percentage (100 × p). Average mortality of replicates was corrected for mortality in the control (< 10%) (Abbott 1925). Probit analysis (Finney 1971) was performed on the corrected dosage-mortality data to estimate the dosage required to kill 50% of larvae (LD₅₀). The magnitude of malathion resistance (relative resistance) in the field populations was calculated as a ratio of LD₅₀ of a field population to the LD₅₀ of the malathion susceptible population; resistance was indicated if the ratio exceeded 1.

The susceptibility of the composite field and malathion susceptible populations of *P.
Figure 1. Toxicity of topically applied malathion to 21 field populations of *Plodia interpunctella* infesting stored shelled corn in southern Minnesota. Values in italics indicate the magnitude of malathion resistance in field populations compared to a malathion susceptible population, and was calculated as LD$_{50}$ of field population/LD$_{50}$ of malathion susceptible population (1.6 µg/ml per larva).

*interpunctella* to pirimiphos-methyl (92% purity) and chlorpyrifos-methyl (98%) was evaluated following procedures similar to that described for malathion.

**RESULTS**

Figure 1 shows the LD$_{50}$s of malathion to the 21 field populations of *P. interpunctella*. The LD$_{50}$ [95% confidence limits (CL)] of malathion to the malathion-susceptible population was 1.6 (1.2–2.1) µg of active ingredient (AI) per larva. The 21 field populations of *P. interpunctella* varied in their susceptibility to malathion, with the LD$_{50}$ values ranging from a low of 52 µg of AI per larva to a high of 1000 µg of AI per larva. The LD$_{50}$ (95% CL) of malathion to the composite field population was 389 (187–818) µg of AI per larva. Field populations of *P. interpunctella* were about 33- to 625-fold more resistant compared to the malathion susceptible population; the composite population was about 243-fold malathion resistant.

The LD$_{50}$s of pirimiphos-methyl and chlorpyrifos-methyl to the composite field population was 1.6 and 0.3 µg of AI per larva, respectively (Table 1). In comparison the LD$_{50}$s of these two insecticides to the malathion susceptible population was 0.5 and 0.3 µg of AI per larva, respectively. The composite field population in comparison to the malathion susceptible population was 3.2-fold resistant to pirimiphos-methyl, but was highly susceptible to chlorpyrifos-methyl.

**DISCUSSION**

Larvae of all 21 field populations of *P. interpunctella* infesting stored shelled corn in southern Minnesota were found to be resistant to topically applied malathion. Fifteen field
Table 1: Toxicity of pirimiphos-methyl and chlorpyrifos-methyl to the composite field and malathion susceptible populations of *P. interpunctella*.

<table>
<thead>
<tr>
<th>Insecticide/Population</th>
<th>µg/µl larva</th>
<th>LD₅₀ (95% CL)</th>
<th>Relative resistance*</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Chlorpyrifos-methyl</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Field composite</td>
<td>0.3 (0.1-0.4)</td>
<td>1.0</td>
<td></td>
</tr>
<tr>
<td>Malathion susceptible</td>
<td>0.3 (0.2-0.4)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Pirimiphos-methyl</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Field composite</td>
<td>1.6 (1.2-2.1)</td>
<td>3.2</td>
<td></td>
</tr>
<tr>
<td>Malathion susceptible</td>
<td>0.5 (0.3-0.6)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Relative resistance = LD₅₀ of composite population / 0.3 or 0.5.

populations were 33- to 365-fold resistant to malathion compared to a malathion susceptible population, while six populations were ca. 625-fold resistant to malathion. Malathion resistance has been well documented in *P. interpunctella* infesting grain in the northcentral United States (Bansode et al. 1981; Beeman et al. 1982; Beeman & Schmidt 1982), California (Armstrong & Soderstrom 1975), and southern United States (Zettler et al. 1973; Zettler 1982). These studies showed the malathion resistance to range from a low of 17-fold (Beeman & Schmidt 1982) to a high of > 200-fold (Zettler et al. 1973, Bansode et al. 1981).

Our composite field population of *P. interpunctella* was 3.2-fold resistant to pirimiphos-methyl in comparison to the malathion susceptible population. However, the composite and malathion susceptible populations were equally susceptible to chlorpyrifos-methyl (Table 1). Malathion resistant larvae of *P. interpunctella* infesting grain in southern United States (Zettler 1974) and in North Carolina (Bansode et al. 1981) were not cross resistant to pirimiphos-methyl. However, Attia et al. (1980) reported a 5-fold resistance to pirimiphos-methyl in larvae of two malathion resistant populations of *P. interpunctella* infesting grain in Australia. Resistance to malathion in *P. interpunctella* larvae in the United States was due to elevated levels of a malathion-degrading enzyme, carboxylesterase (Bansode et al. 1981, Beeman & Schmidt 1982). However, in two malathion resistant populations of *P. interpunctella* in Australia, studies with insecticide synergists indicated that enzymes other than carboxylesterase were involved in conferring resistance to malathion (Attia et al. 1980). In addition, these two malathion resistant populations were also cross resistant to pirimiphos-methyl (Attia et al. 1980). These results suggest that *P. interpunctella* larvae with malathion detoxifying enzymes other than carboxylesterase (e.g. oxidases and non-specific esterases) could develop resistance to pirimiphos-methyl.

Malathion resistance in larvae of *P. interpunctella* was widespread and severe in southern Minnesota. A low level of susceptibility to pirimiphos-methyl in the field composite population suggested that repeated use of pirimiphos-methyl on grain may result in development of resistance in *P. interpunctella*. Resistance to chlorpyrifos-methyl was not detected in the field composite population. However, pirimiphos-methyl and chlorpyrifos-methyl should be used on grain with caution to reduce the likelihood of development of cross resistance in malathion resistant populations of *P. interpunctella* infesting grain in Minnesota.

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