

Insecticide bioassays for western flower thrips (*Frankliniella occidentalis*) (Thysanoptera: Thripidae) and greenhouse whitefly (*Trialeurodes vaporariorum*) (Hemiptera: Aleyrodidae)

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Abstract Bioassays were tested for their suitability to determine the resistance of western flower thrips (*Frankliniella occidentalis*) and greenhouse whitefly (*Trialeurodes vaporariorum*) to insecticides. Adult female greenhouse and lupin strains of western flower thrips were exposed to bean leaf discs treated with insecticide solutions for 24 h at 25°C. The susceptibility of greenhouse strain western flower thrips was further assessed following exposure for 48 h at 25°C to treated bean leaf discs and plastic Petri dishes, the inside surfaces of which had been sprayed with insecticide. The susceptibility of adult greenhouse whitefly was compared when exposed to leaf discs sprayed with insecticide, leaf discs dipped in insecticide solutions, and insecticide-sprayed Petri dishes. Whitefly mortality on leaf discs was assessed after 48 h at 25°C and in plastic dishes after 24 h at 15°C in the dark. A further series of bioassays compared the effect of insecticide on whitefly nymphs after 7 days at 25°C when exposed to various orientations of buprofezin-sprayed leaf discs, and whole leaves dipped into buprofezin solutions. The lupin strain of western flower thrips was

more susceptible than the greenhouse strain to fipronol, maldison, methiocarb, and methamidophos with resistance factors of 14, 19, 26, and 45, respectively. The greenhouse strain of western flower thrips was more susceptible to dichlorvos, lambda-cyfluthrin, maldison, and methamidophos in the Petri dish bioassay than in the leaf disc bioassay, but the thrips were less susceptible to maldison in 1996 bioassays than in 1993 and 1999. Adult greenhouse whitefly were similarly susceptible to endosulfan in the three bioassays, but were more susceptible to methomyl in the Petri dish tests than in the two leaf disc bioassays. Dipped leaf discs gave lower LC₅₀s of whitefly nymphs than other bioassays, but there was high variation between runs. The Petri dish bioassay was successfully adapted for adult western flower thrips and greenhouse whitefly, but its use is limited by the mode of action of different insecticides, and it is not suitable for insecticides that only affect juvenile whitefly.

Keywords diagnostic dose; concentration-mortality response

INTRODUCTION

In New Zealand, western flower thrips (*Frankliniella occidentalis* Pergande) (Thysanoptera: Thripidae) and greenhouse whitefly (*Trialeurodes vaporariorum* (Westwood) (Hemiptera: Aleyrodidae)) are pests of many vegetable and ornamental crops, particularly when grown in greenhouses. Overseas, both pests are known to be resistant to many insecticides (e.g., Broadbent & Pree 1997; Kontsedalov et al. 1998; Rufingier et al. 1999; Jensen 2000; Herron & Gullick 2001; Gorman et al. 2002). In New Zealand, western flower thrips from greenhouses are resistant to several insecticides compared with the outdoor lupin strain (Martin & Workman 1994), whereas resistance to buprofezin has been demonstrated in greenhouse whitefly (Martin & Workman 1995). The failure of an insecticide application to control a pest may be because of factors other than resistance

to insecticide. However, it can be difficult for growers or their advisors to distinguish between, for example, poor application technique and resistance and then to make appropriate decisions. The bioassays used to determine pesticide resistance in these pests in New Zealand have involved rearing colonies of the insects and test plants. They are time-consuming and expensive. Ideally, a simple test is required that can be used by a grower or advisor and gives a result within a day. Several such tests have been proposed for small invertebrates based on treating an inert surface with insecticide, e.g., glass vials (Cahill & Hackett 1992; Broadbent & Pree 1997), yellow sticky traps (Sanderson & Roush 1992), and plastic Petri dishes (Kabir et al. 1991). The simplest of these methods is the plastic Petri dish technique, which was developed for mites and offered the prospect of being able to prepare dishes in advance with a diagnostic dose of insecticide so people could test the insects on their property.

This paper reports on research to determine the suitability of the plastic Petri dish technique for western flower thrips and greenhouse whitefly. The paper also reports on the concentration/mortality response of the greenhouse strain and lupin strains of western flower thrips, and efforts to determine a diagnostic dose for buprofezin and greenhouse whitefly nymphs.

MATERIALS AND METHODS

Six experiments were carried out, two with western flower thrips, and four with greenhouse whitefly, using various insecticides (Table 1). Voucher specimens of each insect population were deposited in the

New Zealand Arthropod Collection, Landcare Research, Auckland. In all of the experiments, serial dilutions of the insecticides were made from commercial formulations. The aim was to have two to three dilutions on either side of the expected LC_{50} . Between 5 and 7 and dilutions were used per run (one insecticide treatment for a bioassay on 1 day). The maximum concentration varied between runs and between experimental treatments. Water was included as an untreated control in all runs of each experiment for each treatment. For each insecticide used, runs of each experimental treatment (a single bioassay comparing two strains of an insect, or one insect strain and comparing two bioassay procedures) were generally conducted at over a period of 4–8 weeks. However, runs for the different insecticides used in a bioassay were carried out at different times. All experiments were conducted between January 1996 and February 1999. Unless otherwise stated, all Petri dishes used in the bioassays were small plastic dishes (35 mm internal diam. and 10 mm high).

Western flower thrips

Two strains of western flower thrips were used in two experiments. The greenhouse strain, derived from an Auckland greenhouse capsicum crop, was reared on flowers of potted chrysanthemum 'Charm' in cages kept at 25°C and 16:8 h light (L) : dark (D). The lupin strain was collected from tree lupin (*Lupinus arboeus* Sims) Piha, West Auckland. Adult female thrips were used in all bioassays.

Experiment A: comparison between strains

Female thrips of both strains were exposed for 24 h to pesticide residues on bean leaf-discs in sealed

Table 1 Insecticides used in bioassays to determine concentration-mortality responses of western flower thrips (*Frankliniella occidentalis*) and greenhouse whitefly (*Trialeurodes vaporariorum*).

| Pesticide | | Recommended concentration | | Experiment |
|--------------------|------------------|---------------------------|--------------------|------------|
| Common name | Product | g a.i./litre | product/100 litres | |
| Thrips | | | | |
| Dichlorvos | Nuvan | 1 | 100 ml | B |
| Fipronil | Regent | 0.04 | 25 ml | A |
| Lambda-cyhalothrin | Karate | 0.01 | 20 ml | B |
| Maldison | Malathion | 1 | 200 ml | A, B |
| Methamidophos | Tamaron | 0.9 | 150 ml | A, B |
| Methiocarb | Mesuro | 0.75 | 100 g | A |
| Whitefly | | | | |
| Buprofezin | Applaud 25 W | 0.125 | 50 g | D, E, F |
| Endosulfan | Endosulfan 35 EC | 0.71 | 200 ml | C |
| Methomyl | Lannate L | 0.24 | 120 ml | C |

containers before mortality was assessed (Martin & Workman 1994). Four insecticides were each tested against both strains, with five (or six) dilutions of insecticide tested plus water controls. Between 2 and 5 runs were used for each strain and insecticide, with either 5 or 10 Petri dishes containing leaf discs used for each concentration for each strain within a run.

The pesticides were applied to both sides of the bean leaf discs (25 mm diam.) in a Potter's tower with 2 ml of solution at 69 Pa for 7 s. The surfaces of the leaf discs were allowed to dry and then placed individually on filter paper in the lids of Petri dishes. Each dish with a leaf disc was inserted into a plastic bag with flowers, and female thrips were tapped in until between 2 and 35 thrips (target 10) were present in each dish. The lid was closed and the dish sealed with Parafilm and held at 25°C. Numbers of live and dead thrips in each dish were recorded after 48 h. Mortality was assessed as no movement when gently touched.

Experiment B: comparison between bioassays

Four pesticides were used in a comparison of bioassays using leaf discs in Petri dishes (as in experiment A), and sprayed Petri dishes (Kabir et al. 1993). For the sprayed dishes, Petri dishes with tight-fitting lids (50 mm diam. and 9 mm high) were used. The internal surfaces of lids and bases were sprayed with insecticide solution using a Potter's tower as for leaf discs above. After drying, a small plug of 1.5% w/v agar was placed in the base of each dish to maintain sufficient humidity. Thrips were added to the dishes as above, giving up to 26 thrips in each dish. The lids were sealed and mortality assessed after 24 h. Only the greenhouse strain was used for these bioassays.

High concentrations of the product containing one insecticide, lambda-cyhalothrin, were difficult to spray through the Potter's tower so for this insecticide all the leaf discs were dipped in insecticide solutions for 5 s and dried.

Each of the four insecticides was used with both bioassays, with 3 or 4 runs used for each. In a run there were either 5 or 10 dishes per concentration.

Whitefly

Greenhouse whitefly were reared on tobacco at Mount Albert Research Centre, Auckland, and had only been exposed to dichlorvos and diazinon during the previous 10 years. For the first experiment (experiment C), adult whitefly were narcotised with carbon dioxide and transferred using a brush to each dish. For the three nymph experiments (experiments

D, E, F), adult whitefly were allowed to lay eggs for 24 h on the primary leaves of dwarf beans (*Phaseolus vulgaris* 'Topcrop'). Adults were removed by blowing with hot air. The plants were kept in a greenhouse until first instar nymphs emerged.

Experiment C: comparison between bioassays

Two insecticides were used in a comparison of three bioassays using adult whitefly. The Petri dish test, as in experiment B, was compared with sprayed leaf discs, as in experiment A, with leaf discs dipped in insecticide solution.

The sprayed Petri dishes were kept for 24 h in the dark at 15°C after sealing. For the other two bioassays, bean leaf discs were either sprayed on one side with a Potter's tower (as in experiment A above) or dipped in insecticide solutions for 5 s. After drying, the leaf discs were placed on a thin layer of 1% agar in small Petri dishes. Adult whitefly were added to each dish (target number 5, actual varied from 1 to 7). The dishes were sealed with Parafilm and kept at 25°C for 48 h. Mortality was assessed as for thrips.

Both insecticides were used in each of the bioassays, with 3–6 runs used for each. In a run there were five dishes per concentration.

Experiment D: nymphs, comparison of sprayed and dipped leaf discs

Two bioassays using whitefly nymphs with one insecticide (buprofezin) were compared. Bean leaf discs (25 mm diam.) with first instar nymphs were either dipped in insecticide solution for 5 s or the discs, with the normal underside facing upwards, were sprayed under a Potter's tower. Dry leaf discs were placed with the upper surface on agar in Petri dishes and closed with a lid lined with filter paper to absorb the excreted liquid. The dish was placed upside down in a cabinet at 25°C, 16:8 L:D photophase. The number of nymphs at each instar were recorded after 7 days. Nymphs that were still at instars 1 and 2 were classified as dead, whereas nymphs at instars 3 and 4 were classified as live. Nymph numbers ranged from 3 to 94 per dish.

Five runs of the three bioassays were carried out, with five dishes per concentration in each run for each bioassay.

Experiment E: nymphs, comparison of dipped leaf discs and whole leaves

Two bioassays using whitefly nymphs with one insecticide were compared. Dipped leaf discs were compared with whole dipped leaves. Dipped discs were prepared as above (experiment D). Detached

whole bean leaves with first instar nymphs were dipped in insecticide solutions for 5 s and allowed to dry. The petiole of each leaf was placed in a vial of water and kept in a cabinet at 25°C, 16:8 L:D photophase for 7 days when the number of nymphs at each instar were recorded. Nymphs that were still at instars 1 and 2 were classified as dead whereas nymphs at instars 3 and 4 were classified as live. Numbers ranged from 1 to 322 per dish or leaf.

Three runs of the leaf disc assay, and four of the intact leaf bioassay were carried out, with 10 dishes per concentration in each run for each bioassay.

Experiment F: nymph bioassays, leaf disc drying times and whole leaves

Five bioassays using whitefly nymphs with one insecticide were compared. Whole dipped leaves were compared with dipped leaf discs that were dried for four different times. Whole leaves and discs were prepared as above (experiments D and E). For discs, the Petri dishes were sealed when the leaf discs were wet, just dry, dried for 2 or 4 h. In this bioassay, only one run per bioassay was carried out, with five dishes per concentration for each bioassay. After 7 days, the number of nymphs at each instar were recorded. Nymphs that were still at instars 1 and 2 were classified as dead whereas nymphs at instars 3 and 4 were classified as live. Numbers ranged from 1 to 119 per dish or leaf.

Statistical analysis

For each experiment, data for each insecticide were analysed separately, with all bioassays and runs for an insecticide analysed together. Thus, there was no direct comparison between the insecticides used within each experiment. Data for each individual dish/leaf disc were used for all the analyses.

Insecticide concentration-response curves were fitted to the proportion of insects that died, taking into account the binomial nature of the data (McCullagh & Nelder 1989, chapter 4). A logistic-concentration-response curve of $\log(\text{concentration})$ was used:

$$\% \text{Dead} = P + \frac{100 - P}{1 + \left(\frac{\text{Conc.}}{\text{LC}_{50}} \right)^{-b}}$$

where b is related to the steepness of the curve at a concentration of LC_{50} , P is the natural % mortality (at Concentration = 0), and LC_{50} is the concentration that kills half of the remaining $(100 - P)\%$ insects. Curves were fitted using the non-linear generalised

model fitting facilities in GenStat, including those implemented in the PROBITANALYSIS command, which allow simultaneous estimation of all parameters (including P). The concentration killing 90% of the insects, including those that died naturally (D_{90}) was estimated from these fitted parameters. Testing for significant differences in the LC_{50} , b or P parameters between treatments or runs was carried out using parallel regression analysis (Ross 1984).

In addition to examining the data through curve-fitting, two binomial generalised linear models with a logit link (McCullagh & Nelder 1989) were used, one fitting to the means for each concentration for each treatment, and one fitting to the means for each concentration of each treatment within each run. Using analysis of deviance (McCullagh & Nelder 1989), a comparison of the fit of these two models allowed an assessment of whether the variation between runs was greater than that within runs for dishes treated similarly. A comparison of the fit of the concentration-response curve to the fit of the treatment mean model similarly allowed an assessment of lack of fit of the curve to the treatment means. For all of the bioassays, the run-to-run variability (i.e., between run mean deviance) was found to be significantly larger than that within runs. Thus, all the standard errors presented here, and tests for differences between bioassays in parameters of the concentration-response curves, were based on the run-to-run variability.

For some of the assays, a few extreme outliers were identified and excluded from the final analysis.

All analyses were carried out using Genstat 5 release 4.1 (Genstat 5 Committee 1997).

RESULTS

Control mortality P did not vary significantly ($P > 0.05$) between bioassays for any of the insecticides in any of the experiments. For most of the experiments, there were significant differences between concentration-mortality curves fitted separately to each run of a bioassay or insecticide, indicating substantial heterogeneity of the response. This is reflected in the standard errors of the fitted parameters of the curves presented here. Unless otherwise stated, there was no significant ($P > 0.05$) lack of fit between single curves (averaged over runs) fitted for each insecticide for each bioassay, and the means for each concentration averaging across runs, indicating that

the fitted curves presented give a good summary of the general responses found. However, because of the large differences between runs, these single curves are not necessarily useful in predicting responses in future experiments.

Western flower thrips experiments

Experiment A: comparison between strains

The lupin strain was more susceptible to the four insecticides than the greenhouse strain by 14–45 times (Table 2). The concentration-mortality curves for the insecticides, except fipronil, had similar slopes (*b*) for both strains, indicating that the main difference between the strains can be summarised as a sideways shift of the concentration response curve when plotted against log of concentration. Thus, the concentrations estimated to kill 90% of insects (D_{90}) were also substantially larger for the greenhouse strain than for the lupin strain.

Experiment B: comparison between bioassays

The greenhouse strain of western flower thrips was more susceptible ($P < 0.05$) to each insecticide in the Petri dish bioassay than in the leaf disc bioassay (Table 3). For one of the insecticides (dichlorvos) there was a significant ($P < 0.05$) lack-of-fit between the concentration-response curves and the means, indicating that the data did not follow a sigmoidal response. This means that the estimate of the concentrations required for the higher kill level (D_{90}) are less well estimated.

Based on the LC_{50} , the insecticides were 3.5× (dichlorvos) to nearly 150× (maldison) more toxic when used in sprayed Petri dishes than when using leaf discs. Even for 90% kill (D_{90}), all of the insecticides were substantially more toxic when sprayed on dishes rather than on leaves.

In these bioassays the greenhouse strain of western flower thrips was less susceptible to maldison

Table 2 Estimated and derived parameters for concentration-response curves for experiment A, comparing bioassays on two strains of thrips (*Frankliniella occidentalis*) for four insecticides (standard errors for parameters are in parentheses). (RF, a resistance factor calculated as the LC_{50} of the greenhouse strain divided by the LC_{50} of the lupin strain; d.f., degrees of freedom associated with the standard errors; *n*, total number of insects used over all runs and concentrations.)

| Insecticide | Thrips strain | <i>n</i> | LC_{50} (g/litre) | <i>b</i> | P (% mortality) | D_{90} (g/litre) | RF |
|------------------------------|---------------|----------|------------------------|--------------|--------------------|-----------------------|----|
| Fipronil (d.f. = 56) | Greenhouse | 2202 | 1.657 (0.115) | 1.65 (0.141) | 3.37 (1.63) | 6.127 | 14 |
| | Lupin | 1827 | 0.115 (0.011) | 1.31 (0.130) | 5.21 (2.16) | 0.591 | |
| Maldison (d.f. = 34) | Greenhouse | 624 | 0.397 (0.075) | 2.06 (0.547) | 1.12 (3.59) | 1.147 | 19 |
| | Lupin | 2638 | 0.021 (0.002) | 2.53 (0.402) | 8.40 (4.07) | 0.048 | |
| Methiocarb (d.f. = 39) | Greenhouse | 2272 | 1.154 (0.188) | 1.57 (0.300) | 5.15 (3.98) | 4.525 | 26 |
| | Lupin | 2950 | 0.044 (0.007) | 1.84 (0.430) | 12.82 (4.09) | 0.134 | |
| Methamidophos (d.f. = 12) | Greenhouse | 858 | 0.226 (0.052) | 1.50 (0.383) | 5.42 (5.03) | 0.941 | 45 |
| | Lupin | 860 | 0.005 (0.001) | 1.73 (0.612) | 14.25 (5.97) | 0.016 | |

Table 3 Estimated and derived parameters for concentration-response curves for experiment B, comparing two bioassays of thrips (*Frankliniella occidentalis*) for four insecticides (standard errors for parameters are in parentheses). (d.f., degrees of freedom associated with the standard errors; *n*, total number of insects used over all runs and concentrations.)

| Insecticide | Bioassay | <i>n</i> | LC_{50} (g/litre) | <i>b</i> | P (% mortality) | D_{90} (g/litre) |
|-----------------------------------|------------|----------|------------------------|---------------|--------------------|-----------------------|
| Dichlorvos (d.f. = 39) | Petri dish | 1193 | 0.543 (0.023) | 4.35 (0.613) | 6.94 (2.97) | 0.884 |
| | Leaf disc | 1106 | 1.883 (0.196) | 1.94 (0.280) | 9.14 (3.09) | 5.54 |
| Lambda-cyhalothrin (d.f. = 25) | Petri dish | 1170 | 0.144 (0.006) | 4.05 (0.559) | 6.43 (3.22) | 0.243 |
| | Leaf disc | 865 | 1.438 (0.208) | 1.28 (0.223) | 4.52 (3.39) | 7.73 |
| Maldison (d.f. = 56) | Petri dish | 1239 | 0.009 (0.001) | 0.866 (0.175) | 2.74 (3.31) | 0.171 |
| | Leaf disc | 1588 | 1.338 (0.232) | 1.31 (0.219) | 2.06 (3.06) | 7.02 |
| Methamidophos (d.f. = 42) | Petri dish | 1204 | 0.057 (0.002) | 5.35 (1.055) | 4.56 (3.39) | 0.085 |
| | Leaf disc | 1276 | 0.461 (0.053) | 2.43 (0.545) | 8.59 (2.88) | 1.10 |

and methamidophos than in the same leaf disc bioassay used when comparing thrips strains (experiment A) (Table 2). This probably reflects the large variation found between runs of the same bioassay. Maldison was 3 times as toxic in experiment A than in experiment B, and methamidophos was twice as effective.

Greenhouse whitefly experiments

Experiment C: comparison between bioassays

There was no significant difference between bioassays in the susceptibility of adult whitefly to endosulfan, but greenhouse whitefly were substantially more susceptible ($P < 0.05$) in the Petri dish bioassay to methomyl than in the two assays using leaf discs (Table 4). For this insecticide, the slope (b) was similar for both bioassays, indicating that the difference between the assays primarily represented a horizontal shift in the response curve, as in experiment A. However, there was some lack of fit ($P > 0.05$) between the fitted curves and the means,

indicating that the curve did not completely describe the concentration responses observed for methomyl.

Experiments D, E, F: nymph bioassays

The LC_{50} for the dipped leaf disc was lower ($P < 0.05$) than for the sprayed leaf disc (experiment D) and the dipped leaf (experiment E) (Table 5). The amount of drying of the leaf discs before sealing the dish with adult whitefly did not significantly affect the mortality of the whitefly (experiment F).

DISCUSSION

The bioassay based on the Petri dish with insecticide residues sprayed from a Potter's tower (Kabir et al. 1993) was successfully adapted for use with adult western flower thrips and adult greenhouse whitefly. A moisture source, a small plug of agar, was essential for survival of the control insects. Good survival of adult whitefly also required the dishes to be held

Table 4 Estimated and derived parameters for concentration-response curves for experiment C, comparing three bioassays of adult whitefly (*Trialeurodes vaporariorum*) for two insecticides (standard errors for parameters are in parentheses). (d.f., degrees of freedom associated with the standard errors; n , total number of insects used over all runs and concentrations.)

| Insecticide | Bioassay | n | LC_{50} (g/litre) | b | P (% mortality) | D_{90} (g/litre) |
|-------------------------|-------------------|-----|------------------------|---------------|--------------------|-----------------------|
| Endosulfan (d.f.=56) | Petri dish | 529 | 16.5 (5.16) | 0.693 (0.111) | 1.75 (2.81) | 382 |
| | Sprayed leaf disc | 678 | 26.2 (7.55) | 1.012 (4.198) | 11.23 (4.60) | 201 |
| | Dipped leaf disc | 676 | 11.1 (2.76) | 1.015 (0.167) | 4.58 (3.58) | 92 |
| Methomyl (d.f.=56) | Petri dish | 400 | 0.011 (0.000) | 4.27 (0.402) | 1.71 (1.62) | 0.018 |
| | Sprayed leaf disc | 807 | 0.534 (0.030) | 2.29 (0.189) | 5.74 (1.70) | 1.335 |
| | Dipped leaf disc | 803 | 0.427 (0.026) | 2.15 (0.185) | 7.75 (1.71) | 1.140 |

Table 5 Estimated and derived parameters for concentration-response curves for experiments D, E, and F, comparing bioassays of whitefly (*Trialeurodes vaporariorum*), using buprofezin (standard errors for parameters are in parentheses). (d.f., degrees of freedom associated with the standard errors; n , total number of insects used over all runs and concentrations.)

| Insecticide | Bioassay | n | LC_{50} (g/litre) | b | P (% mortality) | D_{90} (g/litre) |
|------------------------------|----------------------|-------|------------------------|--------------|--------------------|-----------------------|
| Experiment D (d.f. = 46) | Sprayed leaf disc | 5809 | 1.440 (0.121) | 2.13 (0.252) | 0.69 (1.12) | 4.03 |
| | Dipped leaf disc | 7053 | 0.324 (0.029) | 1.56 (0.177) | 2.01 (1.74) | 1.30 |
| Experiment E (d.f. = 30) | Dipped leaf disc | 15399 | 0.311 (0.053) | 1.79 (0.358) | 4.47 (3.90) | 1.03 |
| | Dipped leaf | 13476 | 0.604 (0.089) | 1.70 (0.331) | 3.63 (3.52) | 2.15 |
| Experiment F (d.f. = 149) | Wet leaf disc | 823 | 0.234 (0.014) | 6.10 (0.994) | 5.52 (2.75) | 0.332 |
| | Just dry leaf disc | 463 | 0.195 (0.028) | 2.78 (0.725) | 11.44 (4.38) | 0.409 |
| | 2 h drying leaf disc | 870 | 0.251 (0.014) | 4.71 (0.630) | 6.13 (2.45) | 0.394 |
| | 4 h drying leaf disc | 773 | 0.238 (0.024) | 3.08 (0.523) | 7.04 (3.06) | 0.473 |
| | Dipped leaf, dry | 2676 | 0.413 (0.024) | 1.88 (0.133) | 5.54 (1.57) | 1.284 |

at a lower temperature (15°C) and in the dark. The technique, however, is limited to insecticides that will kill the insects through contact or fumigant action and is probably dependent upon the interaction between the chemicals and the plastic of the dish. (Kabir et al. 1993) found that the precision of the Petri dish test varied compared with other bioassay methods, depending on the specific active ingredient and the formulation of the active ingredient used. Western flower thrips and greenhouse whitefly tended to be more susceptible to some insecticides when sprayed on to Petri dishes (Tables 3 and 4), but there was unacceptable variability between runs of each bioassay that made it difficult to define a diagnostic dose and makes the test unsuitable for one-off rapid insecticide resistance testing. We were not able to identify the source of this variability. The glass vial test appears to offer greater consistency (Broadbent & Pree 1997), but requires specialised equipment for rotating and drying the treated vials.

For western flower thrips, a comparison of the sprayed leaf disc data for maldison and methamidophos in Tables 2 and 3 and for maldison, methamidophos, and methiocarb in this paper and Martin & Workman (1994) shows that the susceptibility to each insecticide was similar for the lupin strain and greenhouse strain of western flower thrips. However, greenhouse strain western flower thrips appeared to be less susceptible to maldison when tested in 1996 (Table 3) than in 1993 (Martin & Workman 1994) and 1999 (Table 2), giving a resistance factor of 64 compared to 19.

For greenhouse whitefly nymphs, a comparison of sprayed leaf disc data showed that whitefly in the tests reported in this paper were less susceptible to buprofezin (LC₅₀ 1.44 mg/litre, Table 5) than suggested in earlier estimates (Martin & Workman 1995) (LC₅₀ 0.135 mg/litre). These differences could be because of the variability experienced between runs of experiments. High variability was also experienced in experiments using detached leaves (Table 5). Survival of whitefly nymphs is influenced by leaf quality and the use of leaves attached to plants (Cahill et al. 1996b) appears to be a better option, though this requires either controlled environment rooms with high quality lighting or an adequately cooled greenhouse.

For insecticides that only kill whitefly nymphs or where systemic activity is important, bioassays involving leaf discs, detached leaves, or attached leaves are essential (Martin & Workman 1995; Cahill et al. 1996a,b). Buprofezin kills juvenile whitefly when they moult. Although leaf discs can

be used to compare the susceptibility of whitefly strains to the chemical (Martin & Workman 1995), the use of detached leaves appeared promising (Cahill pers. comm.), but attached leaves are best (Cahill et al. 1996b). Detached bean leaves do not require as much space in controlled environment facilities as pots of bean seedlings and thus were chosen for our research. In our experiments, dipped leaf discs gave the lowest LC₅₀s, even lower than the dipped leaves (Table 5). This did not seem related to the dryness of the leaf discs when the Petri dish was closed (Table 5), but may be associated with the vapour action of buprofezin or the poorer quality of the leaves within the dish compared to the detached leaves. Use of leaves attached to seedlings in well ventilated chambers should prevent these problems.

For insects known to be prone to developing resistance to insecticides, it would be desirable to have a suite of diagnostic bioassays to test suspect insect populations for resistance to insecticides. The comparison of the concentration-response of western flower thrips to four insecticides (Table 2) confirms earlier findings (Martin & Workman 1994) that the greenhouse strain is less susceptible to maldison, methamidophos, and methiocarb than the lupin strain with resistance factors of 19, 45, and 26, respectively. However, all three products provide field control of western flower thrips. More recently, the discovery of onion thrips, *Thrips tabaci* Lindeman, in onion crops that were resistant to insecticides (Martin et al. 2003) stimulated research into baseline data for new insecticides registered for onion crops. However, these baseline bioassays are largely dependent on the input of the companies marketing the insecticides, or the cropping industries. The testing of populations of insects for resistance is likely only when control of the pest has become very difficult and companies marketing the products or groups of growers using the insecticides pay for the tests.

ACKNOWLEDGMENTS

For technical assistance, thanks to Tatjana Balint and Mark Bullians.

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