TOXICITY OF INSECTICIDES USED IN CITRUS TO *APHYTIS MELINUS* DEBACH (HYMENOPTERA: APHELINIDAE) AND *RHIZOBIUS LOPHANTHAE* (BLAISD.) (COLEOPTERA: COCCINELLIDAE)

T.S. BELLOWS, JR. and J.G. MORSE

Department of Entomology, University of California, Riverside, California, USA 92521

Abstract

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The 48-h toxicities of freshly deposited residues of 28 insecticides against *Aphytis melinus* DeBach and *Rhizobius lophanthae* (Blaisd.) were evaluated at various rates. Rates as high as 4-fold the recommended field rate of several materials (formulated plant alkaloids, amitraz, formulated *Bacillus thuringiensis* Berliner endotoxin or exotoxin, and cryolite) revealed little mortality to either species. Concentration–mortality regressions were quantified for the remaining materials, which included five carbamates, a macrocyclic lactone, eight organophosphates, and four pyrethroids. Of these, pyrethroids in general were most toxic, followed by carbamates, and then organophosphates, to both species. Most materials tested were more toxic to *A. melinus* than to *R. lophanthae*

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Résumé

La toxicité des résidus de 28 insecticides fraîchement appliqués en diverses doses a été mesurée après 48 h chez *Aphytis melinus* DeBach et *Rhizobius lophanthae* (Blaisd.). Des doses même quatre fois plus élevées que la dose d'application recommandée en nature de plusieurs substances (diverses préparations d'alcaloïdes végétaux, amitraz, préparation d'endotoxine ou d'exotoxine de *Bacillus thuringiensis* Berliner, cryolite) ont causé peu de mortalité chez l'une ou chez l'autre espèce. Des régressions concentration–mortalité ont permis d'évaluer la toxicité d'autres substances, cinq carbamates, une lactone macrocyclique, huit organophosphates et quatre pyréthroïdes. Ce sont les pyréthroïdes qui se sont avérées les plus toxiques, suivies des carbamates, puis des organophosphates, chez les deux espèces. La plupart des substances étaient plus toxiques chez *A. melinus* que chez *R. lophanthae*.

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Introduction

Beneficial insects in many agricultural systems are affected by applications of insecticides targeted at a variety of pests. The effects of such applications can be both immediate and longer term. Immediate effects generally are in relation to the amount of insecticide deposited during application, the most severe of which is mortality of the beneficial organisms. Longer term impacts result from the amount of toxic residues present following treatment. Some insecticides dissipate rather quickly and have short residual impact, others decompose into compounds more toxic than the parent material, and others have longer residual half-lives and can affect natural enemy populations for considerable lengths of time (Bellows et al. 1985; Morse et al. 1987; Bellows and Morse 1988; Bellows et al. 1988, 1992, 1993). These longer term effects include both mortality and sublethal ones.

Insect pest management in citrus agroecosystems relies on both natural enemies and insecticides and is perhaps typical of many such ecosystems (Haney et al. 1992). A number of pests are suppressed by natural enemies, but other pests are subject to chemical treatments. The long-term residual effects of several insecticides used in citrus production on a number of beneficial organisms, including parasitic Hymenoptera and predaceous mites and beetles, have been examined (Bellows et al. 1985; Morse et al. 1987; Bellows and Morse 1988; Bellows et al. 1988, 1992, 1993). The immediate effects of many of these insecticides have not been studied, and must be quantified to understand fully the significance of insecticides selected for use in any particular management scheme. This paper compliments an earlier report (Morse and Bellows 1986) on immediate toxicity of selected materials by evaluating the toxicity of 28 insecticides to *Aphytis melinus* (DeBach), a parasitic wasp important in biological control of diaspidid scales in many parts of the world, and to *Rhizobius lophanthae* (Blaisd.), a coccinellid predator of diaspidid scales.

Materials and Methods

Aphytis melinus adults were obtained from a commercial insectary where the population was maintained on oleander scale, *Aspidiotus hederae* (Ballot), on squash. Adult *R. lophanthae* were collected from a colony maintained in a greenhouse at the University of California, Riverside, on the same scale host.

Insecticide evaluations were conducted in a manner similar to that described by Morse et al. (1986) and Morse and Bellows (1986). The 28 compounds evaluated (together with their commercial formulations and sources) were the pyrethroids esfenvalerate (Asana 1.9 EC, E.I. DuPont, Wilmington, DE), bifenthrin (Brigade 10% WP, FMC, Princeton, NJ), fenpropathrin (Danitol 2.4 EC, Valent, Walnut Creek, CA), and fluvalinate (Spur 2 EC, Sandoz, San Diego, CA); the macrocyclic lactone abamectin (Agri-mek 0.15 EC, Merck, Sharp & Dohme, Three Bridges, NJ); the carbamates carbosulfan (Advantage 4 EC, FMC), formetanate (Carzol 92% SP, Nor-Am, Naperville, IL), methomyl (Lannate 1.8 EC, E.I. DuPont), thiodicarb (Larvin 3.2 EC, Rhone Poulenc, Research Park Triangle, NC), and carbaryl (Sevin 80% SP and Sevin XLR 4F, Union Carbide, Research Park Triangle, NC); the organophosphates naled (Dibrom 8 EC, FMC), trichlorfon (Dylox 80% SP, Mobay, Kansas City, MO), dimethoate (Cygon 4 EC, American Cyanamid, Wayne, NJ), azinphos-methyl (Guthion 50% WP, Mobay), chlorpyrifos (Lorsban 4EC, Dow, Midland, MI), acephate (Orthene 75% SP, Valent), parathion (Parathion 25% WP, Platte, Greeley, CO), mevinphos (Phosdrin 4 EC, E.I. DuPont), and methidathion (Supracide 2 EC, Ciba Geigy, Greensboro, NC); *Bacillus thuringiensis* Berliner exotoxin [Dibeta 1.5% (w/w, 17.26 g AI per L), Abbott, North Chicago, IL] and endotoxin [Dipel 2X (0.032 billion International units (BIU) per g), Abbott; Javelin WG (0.032 BIU per g, 0.053 billion spodopteran units (BSU) per g), and Javelin SC (8.45 BIU per L, 16.91 BSU per L), Sandoz]; the inorganic compound cryolite (Kryocide 96% WP, Penwalt, Philadelphia, PA); the organo-tin compound amitraz (Mitac 1.5 EC, Nor-Am); ryanodine plant alkaloids (Ryan 50 0.1%, Dunhill, Rosemead, CA) and sabadilla plant alkaloids (Veratran D 0.2%, Dunhill).

Commercial insecticide formulations were prepared at various dilutions in water in a high speed blender. Citrus leaves were collected from untreated lemon groves at the University of California, Riverside, Citrus Experiment Station. Leaves were selected arbitrarily from leaves in the outer canopy of the trees between 1 and 2 m from the ground. Leaves selected were mature, fully sized, and dark green. Leaves were dipped into a solution and allowed to dry for 1–2 h. These leaves were placed in modified Munger cells (Munger 1942; Morse and Bellows 1986), which provided a closed test arena 3.2 cm in diameter and 0.9 cm high with the dorsal surface of the treated leaf as the base. Air was passed through screening at the side of the test arena at an average rate of 5.2 mL per s to reduce fumigation effects. The sides of the Munger cells were provisioned with minute drops of honey as a food source. One or more initial trials were conducted at various rates for each material to determine the range of rates that would incorporate 20–80% mortality; separate initial ranges were determined for each of the two test species.

Aphytis melinus adults, 0–24 h old, were introduced into the cells after anesthesia with CO_2. *Rhizobius lophanthae* adults of undetermined age were introduced into the cells without anesthesia. Five test insects (of one species) were placed together in each Munger cell with 10–20 replicate cells per insecticide rate. For compounds showing little effect on

TABLE 1. Toxicity of selected insecticides to adult *Aphytis melinus*

Insecticide	Field rate (FR) (g AI per L)	No. adults tested	Slope ± SE	LC_{50} (g AI per 100 L)	LC_{50} 95% FL	LC_{50} vs FR
Pyrethroids						
Bifenthrin	0.060	952	1.090 ± 0.249	0.049	0.025–0.146	0.0082
Esfenvalerate	0.015	1330	1.609 ± 0.085	0.016	0.010–0.027	0.0109
Fenpropathrin	0.060	510	2.646 ± 0.218	0.006	0.005–0.007	0.0010
Fluvalinate	0.060	1338	1.400 ± 0.288	6.503	3.429–13.451	1.08
Macrocyclic lactone						
Abamectin	0.0070	521	1.576 ± 0.232	0.165	0.100–0.251	0.2357
Carbamates						
Carbaryl 80S	1.20	809	1.908 ± 0.339	2.191	1.434–3.281	0.0180
Carbaryl XLR	1.20	1548	1.064 ± 0.313	2.914	1.227–21.764	0.0240
Carbosulfan	1.80	1013	1.054 ± 0.204	0.051	0.016–0.098	0.0003
Methomyl	1.08	891	1.602 ± 0.366	1.493	0.518–2.710	0.0138
Thiodicarb	1.08	899	1.549 ± 0.312	56.364	29.423–96.828	0.5227
Organophosphates						
Azinphos-methyl	0.240	1350	1.216 ± 0.162	0.371	0.253–0.553	0.0083
Mevinphos	2.40	499	1.329 ± 0.232	9.648	4.258–16.786	0.0403
Naled	3.00	920	1.444 ± 0.349	47.445	21.030–108.05	0.1584
Trichlorfon	4.79	479	2.641 ± 0.742	29.510	11.492–52.774	0.0616

the test species, evaluations were conducted at the field rate and at 2× and 4× the field rate. For the remaining materials, 6–10 rates were tested (within the initially determined range) to obtain concentration–mortality relationships. Mortality was evaluated by observing treated insects through a microscope 48 h after each experiment was initiated. All experiments were conducted at ambient laboratory conditions of 20–25°C and 30–50% RH. Data were adjusted for control mortality (which varied from 0 to 5%) in cells bearing untreated leaves using the formula of Abbott (1925), and relationships between log rate and mortality were quantified with probit analysis (SAS Institute 1985).

Results and Discussion

There were significant relationships between rate and mortality for 14 of the 20 compounds tested against *A. melinus* and for 12 of the 23 compounds tested against *R. lophanthae* (Tables 1 and 2). Slopes of these relationships ranged between 1.0 and 2.6, and were similar to those reported in tests of other insecticide residues (Morse and Bellows 1986; Bellows et al. 1992, 1993). In the present tests, the insects were exposed to recently deposited insecticide by walking and other activity on the leaf surface as well as through evaporation of the insecticide into the atmosphere, as opposed to being treated directly with the insecticide, and this difference in mode of contact may account for the lower slopes. *Aphytis melinus* adults were regularly observed to be walking about on the leaf surface during the trials. *Rhizobius lophanthae* appeared less active, and occasionally rested on the sides of the container.

Nearly all of the synthetic insecticides tested against *A. melinus* showed significant rate–mortality relationships. There was a wide range of toxicities among the materials tested. Comparison among calculated LC_{50} values indicates several orders of magnitude differences in toxicity even among the same class of compounds. Among the pyrethroids, fenpropathrin was most toxic (LC_{50} of 0.006 g AI per 100 L) and fluvalinate was least toxic (LC_{50} of 6.50 g AI per 100 L), a difference in toxicity of over 1000-fold. Similar ranges occurred among the carbamates and, to a lesser degree, among the organophosphates. As a group, the pyrethroids tested were most toxic, followed by the carbamates, and then the organophosphates. These

TABLE 2. Toxicity of selected insecticides to adult *Rhizobius lophanthae*

Insecticide	Field rate (FR) (g AI per L)	No. adults tested	Slope ± SE	LC_{50} (g AI per 100 L)	LC_{50} 95% FL	LC_{50} vs FR
Pyrethroid						
Fluvalinate	0.060	449	1.530 ± 0.216	5.643	3.670–8.493	0.9418
Macrocyclic lactone						
Abamectin	0.0070	375	1.740 ± 0.337	8.935	4.790–23.487	12.764
Carbamates						
Carbaryl 80S	1.150	419	1.003 ± 0.210	13.70	3.159–29.896	0.1192
Carbosulfan	1.797	401	1.649 ± 0.096	3.502	2.023–5.671	0.0195
Methomyl	1.078	573	2.320 ± 0.450	2.022	1.308–3.209	0.0188
Thiodicarb	1.078	1125	1.120 ± 0.161	1.363	0.738–2.194	0.0126
Organophosphates						
Acephate	0.599	412	1.793 ± 0.236	26.188	17.687–36.313	0.4371
Azinphos-methyl	0.449	754	1.066 ± 0.075	6.089	4.780–7.619	0.1356
Dimethoate	1.198	425	1.182 ± 0.271	121.89	57.998–420.80	1.0175
Methidathion	0.300	602	1.378 ± 0.249	4.830	2.470–7.968	0.1610
Parathion	0.719	293	1.857 ± 0.261	551.11	454.48–697.14	7.6649
Inorganic						
Cryolite	11.504	491	1.051 ± 0.212	4241.8	2241.8–7810.4	3.6872

results are similar to those reported by Bellows and Morse (1988) for a different set of compounds, where carbaryl was found to be more toxic than most organophosphates tested. Six materials showed little toxicity to *A. melinus* (Table 3). These included three formulations of *B. thuringiensis* toxins, the inorganic compound cryolite, the organo-tin compound amitraz, and the ryanodine plant alkaloids. These materials caused less than 12% mortality when tested at the field rate, and little relationship between rate and mortality. Earlier studies also indicated that the sabadilla plant alkaloids had little toxicity to *A. melinus* (Morse and Bellows 1986).

Fewer compounds showed high toxicity to *R. lophanthae* (Tables 2, 3), and these were in general less toxic to *R. lophanthae* than to *A. melinus*. There was a somewhat smaller range in measurable toxicities among the compounds tested. The minimum (thiodicarb) and maximum (carbaryl 80S) LC_{50} among carbamates varied 10-fold, and the range for organophosphates (methidathion and parathion) was 114-fold. The carbamates were more toxic in general than the organophosphates. Eleven materials showed little toxicity to *R. lophanthae* (Table 3), including three organophosphates, one pyrethroid (fenpropathrin), one carbamate (formetenate), the formulated *B. thuringiensis* toxins, the organo-tin compound (amitraz), and the plant alkaloids. These results are similar to those obtained for another coccinellid, *Cryptolaemus montrouzieri* (Mulsant), which also was measurably affected by fewer materials than *A. melinus* (Morse and Bellows 1986). Some exceptions to the general trend of lower toxicity to the beetle occurred, however, most notably with thiodicarb, which was the least toxic material to *A. melinus* and the most toxic to *R. lophanthae*.

In evaluating an insecticide for natural enemy selectivity, it is perhaps more important to evaluate its potential impact relative to the application rate in the cropping system than to compare its toxicity directly with other compounds. For example, a highly toxic material applied at a low rate in the field may cause less mortality than a less toxic material used at a higher application rate. For this reason, Tables 1 and 2 include a column that lists the LC_{50} as a fraction of the recommended field rate (cf. Morse and Brawner 1986) for each material [from Bailey and Morse (1988), for materials yet to be registered, the anticipated field rate is listed]. A low value of this parameter indicates a greater toxicity at the field rate than does

TABLE 3. Compounds showing little effect on adult *Aphytis melinus* and *Rhizobius lophanthae*

Insecticide	Field rate (g AI per L)	% corrected mortality (*N*) at rate indicated		
		Field rate	2 × field rate	4 × field rate
		(*a*) *A. melinus*		
Amitraz	1.797	11.56 (53)	5.86 (52)	32.48 (54)
B. thur. exotoxin (DiBeta)	17.26	0.00 (50)	0.00 (52)	0.00 (47)
B. thur. endotoxin (Dipel)	0.60*	0.00 (51)	4.21 (49)	3.43 (56)
B. thur. endotoxin (Javelin SC)	5 mL per L*	4.04 (50)	5.62 (53)	7.73 (52)
Cryolite	11.50	2.12 (49)	1.96 (51)	2.12 (49)
Ryanodine alkaloids	0.0042	0.00 (49)	15.62 (52)	33.97 (51)
		(*b*) *R. lophanthae*		
Amitraz	1.797	0.00 (25)	0.00 (25)	8.00 (25)
Chlorpyrifos †	0.449	0.00 (25)	28.29 (50)	35.46 (50)
B. thur. exotoxin (DiBeta)	17.26	4.43 (47)	2.08 (49)	8.13 (50)
B. thur. endotoxin (Dipel)	0.60*	0.00 (50)	0.00 (50)	2.08 (49)
B. thur. endotoxin (Javelin WG)	1.2*	4.04 (50)	5.62 (53)	7.73 (52)
Fenpropathrin	0.060	10.00 (51)	18.40 (50)	16.36 (50)
Formetanate	0.600	12.24 (50)	16.70 (49)	4.08 (50)
Naled	2.996	8.00 (25)	0.00 (25)	0.00 (25)
Ryanodine alkaloids	0.0042	0.00 (40)	0.00 (40)	0.00 (39)
Sabadilla alkaloids	0.024	0.00 (26)	0.00 (25)	0.17 (24)
Trichlorfon	4.793	0.00 (25)	4.00 (50)	12.00 (50)

*Rate is g formulated product per L.
†Rates tested for chlorpyrifos were 7.19, 14.38, and 28.76 g AI per L, approximately 16, 32, and 64 × the field rate.

a high value; we use it here to compare the potential effects among materials of immediate toxicity following an agricultural application.

With reference to the ratio of LC_{50} : field rate, there was a wide variety of toxicities both within and among insecticide groups. For *A. melinus* (Table 1), values range from 0.0010 for fenpropathrin to 0.564 for thiodicarb. Some materials that were toxic in an absolute sense are applied in the field at very low rates and in consequence have high LC_{50} : field rate values, indicating that they might have less impact when used in the system than their toxicities alone indicate. Abamectin, for example, is more toxic to *A. melinus* than any of the organophosphates tested here, but has a much lower application rate and consequently a higher LC_{50} : field rate ratio than any organophosphate. Seven materials had LC_{50} : field rate ratios less than 0.020 for *A. melinus*, indicating high toxicity at field rates. The general trends among groups of compounds for *A. melinus* indicated that the pyrethroids were most toxic at the field rate, followed by the carbamates, and then the organophosphates. Values for *R. lophanthae* similarly indicated a range of potential toxicities at the field rate, with values of LC_{50} : field rate ranging from 0.0126 for thiodicarb to 12.76 for abamectin. The carbamates as a class appeared more toxic at the field rate than the organophosphates. No values for LC_{50} : field rate were calculated for the materials that had little measurable toxicity (Table 3); the value in these cases is probably sufficiently large that it would serve only to emphasize the relatively small impact expected from these materials on these two species.

The results presented here indicate that there is a wide range of toxicities, and therefore of range of potential in preserving natural enemies, among the materials currently available for use in managing insect pests in citrus. We stress the comparative, rather than absolute, value of these results; the information in Tables 1–3 can indicate a relative ranking in terms of likely impact on these beneficial organisms, but probably cannot predict the actual toxicity immediately following a treatment. This caution is due partly to the potential difference in the actual amount of insecticide deposited on citrus foliage by the technique used here and by a field treatment. The comparative results, however, will be relatively independent of this difference.

For the case of *A. melinus*, several compounds (*B. thuringiensis* toxin formulations, cryolite, amitraz, and ryanodine alkaloids) showed little toxicity and probably would have minimal impact on field populations of this parasitoid. Among the more toxic compounds, most of the insecticides were highly toxic at field rates. The exceptions were fluvalinate, which was 100- to 1000-fold less toxic at the field rate than the other three pyrethroids, and abamectin, thiodicarb, and naled, all of which were 10- to 100-fold less toxic than the remaining materials.

For the case of *R. lophanthae*, 11 materials (Table 3) showed little toxicity even above field application rates, and could likely be used with minimal impact on field populations. Among the more toxic compounds (Table 2), abamectin, parathion, and cryolite were the least toxic at field rates. The remaining materials had toxicities approximately equal to or greater than LC_{50} at the field rates, and field rate applications would likely be toxic to populations of this predator.

In addition to the immediate toxicity of a material, the longevity of residual action may play a significant role in determining the suitability of a compound for use in an integrated pest management program. The residual toxicity of a material arises from a combination of mechanisms which include not only insecticide toxicity but also insecticide application rate, dissipation rates, oxidation, and toxicity of oxidation products (Bellows et al. 1985). The residual half-life and residual toxicity of many of the compounds tested here for immediate toxicity have been examined (Bellows et al. 1985, 1993; Morse et al. 1987; Bellows and Morse 1988). The length of time a material remains toxic following field treatment may be ranked (number of days following treatment that mortality of adults exposed to leaves remained above 20%) and these ranks plotted against immediate toxicity expressed in an increasing scale relative to the rate applied in the field (field rate : LC_{50}) (Fig. 1). Such a plot can serve to integrate much of the information we have on immediate [from this study and from Morse and Bellows (1986)] and residual impact of various materials to a particular natural enemy.

Several issues arise from such a plot for *A. melinus* (Fig. 1). First, there are several materials that have little direct toxicity and no or little residual impact to *A. melinus*; these include sabadilla, formulated *B. thuringiensis* toxins, and cryolite. Second, the synthetic materials all had immediate toxicities which were near or exceeded the LC_{50} at the field application rate. Treatments made with low-volume concentrations are plotted at higher toxicities : LC_{50} than dilute treatments of the same material because of the greater concentration of active material per volume of water applied. The pyrethroids, carbamates, and many of the organophosphates generally had the most persistent residues, ranging from 17 to >32 days. Applications made during the spring generally had longer residual impact than the same or related materials applied in the summer; the higher temperatures in summer probably contributed substantially to greater dissipation rates and shorter residual toxicities. Combining this information with the degree of immediate toxicity (Fig. 1), we find that sabadilla, the formulated *B. thuringiensis* toxins, and cryolite appear to offer the most promise for selectivity or conservation of the natural enemy; these materials would be excellent candidates for use in integrated management programs where protecting *A. melinus* populations from insecticide effects was important. Naled and abamectin also offer some possibility for conservation or at least early reestablishment of the natural enemy populations because of their short residual activity, as do methomyl and dilute summer treatments of some organophosphates and carbaryl. The remaining materials were more toxic at field rates (all of the synthetic pesticides but methidathion were more toxic than the LC_{50} at the field rate) and persisted at toxic levels for 2 weeks or more (Fig. 1), indicating less opportunity

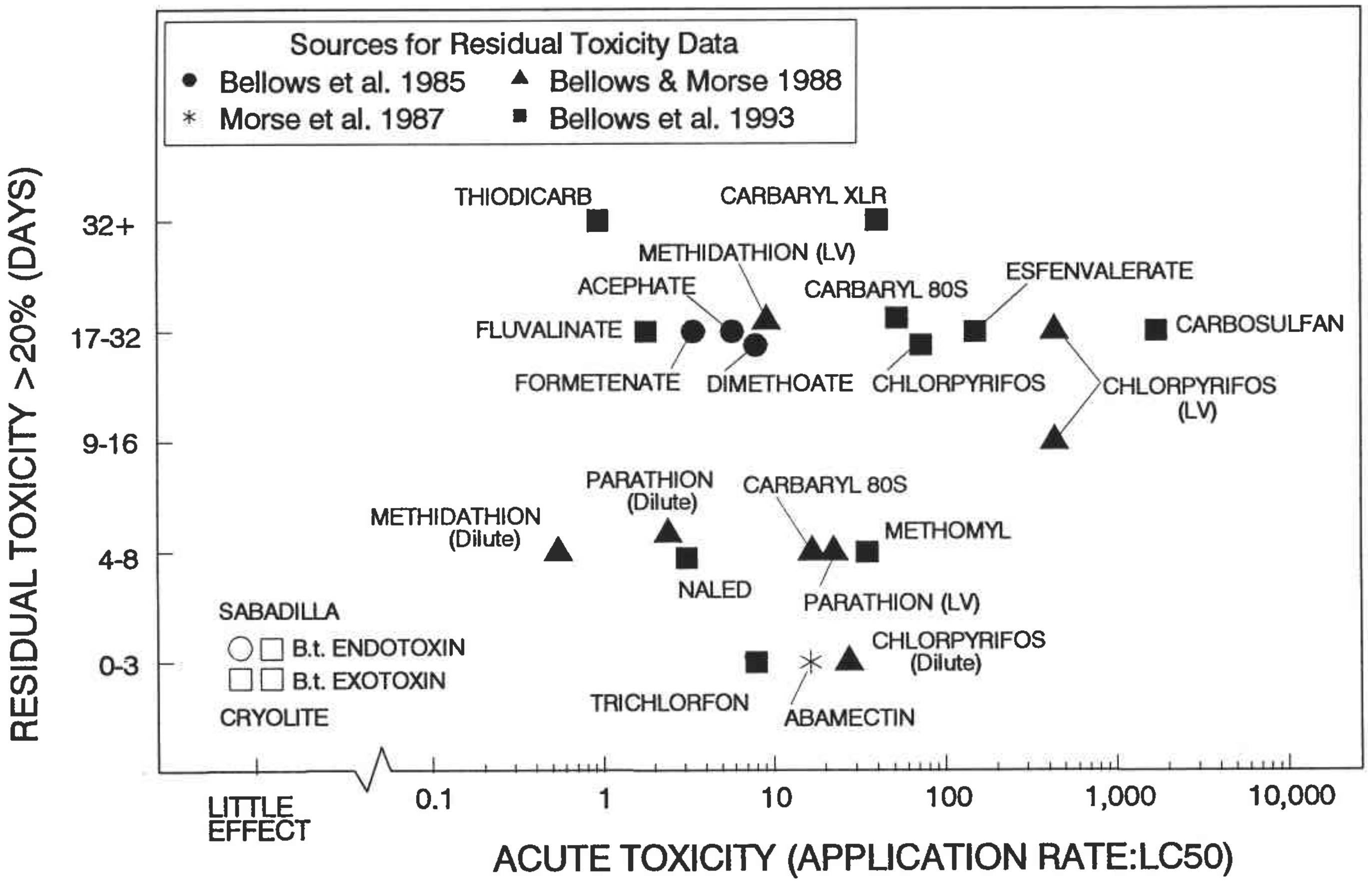

FIG. 1. Length of time residual mortality exceeded 20% plotted against field rate as a fraction of the LC_{50} of selected insecticides for *Aphytis melinus*. Hollow symbols are for materials that showed little or no immediate toxicity at 4 times the field rate. Data on immediate toxicity are from this paper (Table 2) and from Morse and Bellows (1986).

for conserving *A. melinus* populations. Materials most toxic or with the longest residual mortality were esfenvalerate, carbaryl (XLR), carbosulfan, thiodicarb, and low-volume chlorpyrifos; these materials should be avoided in management of orchards where biological control by *A. melinus* is a key part of the overall pest suppression program. Less information is available regarding residual toxicities for these materials for *R. lophanthae*, but information based on immediate toxicities indicates that several compounds may provide for conservation of this natural enemy, including formulated *B. thuringiensis* toxins, sabadilla and ryanodine alkaloids, and a few synthetic materials (Table 3).

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