Israel Journal of Entomology

Vol. XXIX (1995) pp. 309-313

RELEASE OF PESTICIDE-RESISTANT *APHYTIS* STRAINS IN ISRAELI CITRUS ORCHARDS

ABRAHAM HAVRON[1], DAVID ROSEN[1] AND AMOS RUBIN[2]
[1]*The Hebrew University, Faculty of Agriculture, Rehovot 76100, Israel*
[2]*Biological Control Laboratory, 14 Rahavat Ilan, Givat Shrnuel, Israel*

ABSTRACT

The discovery in Israel of a field population of *Aphytis lingnanensis* Compere (Hymenoptera: Aphelinidae), partially resistant to pesticides, was followed by selection procedures that further increased resistance to azinphosmethyl, x80 relative to a susceptible strain. A field population of *A. melinus* DeBach was selected for carbaryl resistance in California. Both strains were repeatedly released on populations of *Aonidiella aurantii* (Maskell) (Homoptera: Diaspididae), infesting commercially managed citrus groves in various parts of Israel. Here , we report on release procedures, initial recoveries and their preliminary evaluation.

KEY WORDS: *Aphytis lingnanensis, A. melinus, Aonidiella aurantii,* pesticide resistance, release, selection, citrus.

INTRODUCTION

Genetic improvement of beneficial arthropods, and of biological control agents in particular, has long been advocated and repeatedly attempted. Resistance to pesticides has frequently been pointed out as a desirable property, enhancing the performance of natural enemies in integrated pest management (IPM) programs (Hoy, 1986; Roush, 1979). Significant increases of such resistance have been achieved by artificial selection in phytoseiid mites, green lacewings and parasitic Hymenoptera (Croft, 1990; Havron et al., 1991; Hoy, 1986, 1990; Rosenheim and Hoy, 1988).

Selection for pesticide resistance might profitably utilize partially resistant field populations. Unfortunately, high levels of tolerance to pesticides are rarely found in field populations of natural enemies of agricultural pests. This may be explained as an effect of the limited, intermittent selection pressure, resulting from pesticidal treatments timed according to host abundance and disregarding parasite life cycles. Also, parasites may be capable of developing resistance only after their hosts have become resistant and increased in number. However, this situation, amounting to control failure, typically leads to a change in pesticides, again slowing down the development of resistance in parasite species (Croft, 1990; Hoy, 1990).

Our selection experiments utilized the fortuitous discovery of a population of *Aphytis lingnanensis* Compere (Hymenoptera: Aphelinidae) in a citron orchard in Ra'nana, on the coastal plain of Israel. This orchard had been subjected to an extremely intensive spraying program, and the survival of the parasite [and of its host, the California red scale, *Aonidiella* *aurantii* (Maskell)] suggested possible resistance to one or several of the compounds utilized. Tolerance tests with azinphosmethyl indeed indicated a tolerance ratio of ×10.3 relative to a susceptible population of this species (Havron et al., 1991).

A. melinus DeBach, another effective natural enemy of the California red scale, was selected for resistance to carbaryl by Rosenheim and Hoy (1988) in California.

These resistant strains were released in, and recovered from, commercially managed citrus orchards in Israel. Here we describe the release procedures and initial recoveries and tests of establishment to date.

The resistant *A. lingnanensis* population was released in designated orchards along the coastal plain of Israel. Pre- and post-release populations were sampled and tested for azinphosmethyl resistance. The resistant *A. melinus* from California was released in various citrus-growing areas in Israel. This species, known to be highly effective under hot and dry climatic conditions, had previously been introduced and established in Israel (Kamburov, 1974; Rosen and DeBach, 1979). However, recent recoveries have been rare, and none were made in our pre-release surveys.

MATERIALS AND METHODS

Insects. *A. lingnanensis.* A field population had been found to persist in a citron orchard under an exceptionally heavy spraying schedule of organophosphorus and other pesticides. Laboratory tests (Havron et al., 1987a; 1991) showed a ca. 10-fold resistance to azinphosmethyl relative to a susceptible laboratory strain, as well as high variability of the resistance trait. Upon selection by both the mass- and male-selection procedure, this resistance was further increased 8-fold (at the LC_{90} level) after 18 selection cycles (Havron et al., 1987b, 1991; Javier et al., 1991).

A. melinus. A carbaryl-resistant population was received from California and maintained in our laboratory for releases. Field populations had been collected in California citrus orchards with relatively high pesticide use, and combined prior to selection. LC_{50} tests had indicated ca. 5-fold resistance to carbaryl relative to a susceptible strain, which was further increased 4-fold by 13 laboratory selection cycles (Rosenheim and Hoy, 1988; Spollen and Hoy, 1992).

Release sites. *A. lingnanensis* was released in the coastal plain, mainly in citron (*Citrus medica*) groves. Unblemished citron fruits are used in Jewish religious rites and command very high prices. Citron orchards are therefore often maintained under heavy pesticide regimes.

A. melinus was released in the southern coastal plain (Shuva, Be'eri, moderate climate), in the Upper Galilee (Gadot, hot and moderately dry) and near the Dead Sea (Ein Gedi, extremely hot and dry).

Sampling and testing. Pre-release samples were obtained from California red scale-infested fruits and held in the laboratory for parasite emergence and propagation. Tolerance tests were conducted and evaluated according to Havron et al. (1987a). In several instances, very low California red scale populations precluded recovery of *A. lingnanensis* for pre- or post-release assays (Table 1).

Post-release (recovery) samples were taken from release trees or from nearby trees, and emerging *Aphytis* were propagated and tested as described for pre-release populations.

TABLE 1

Aphytis lingnanensis releases and results of tolerance tests

No.	Location	Cultivar	No. of specimens released	Pre–release: CRS infestation (relative scale)*	Pre–release: *Aphytis* tolerance LC_{50} (95% confidence limit), ppm	Post–release: CRS infestation (relative scale)	Post–release: *Aphytis* tolerance LC_{50} (95% confidence limit), ppm
1	Kadima	Citron	183,000	2	–	3	7.2 (3.61–11.7)
2	Rishon Le Zion	Citron	257,000	1	–	3	44.5 (32.7–62.3)
3	Petach Tikva	Citron	2,640,000	0–1	–	3	31.1 (21.1–45.7)
4	Kefar Habad	Citron	250,000	3	3.1 (1.74–4.83)	3	9.8 (5.0–15.4)
5	Hibat Zion	Citron	110,000	2	9.7 (8.5–11.0)	0–1	No recovery
6	Kefar Saba	Citron	752,000	3	17.2 (15.4–19.3)	0–1	No recovery
7	Tzrifin	Valencia	4,000	3	34.5 (23.0–53.8)	2	31.9 (21.6–52.9)
8	Ra'anana	Citron	92,000	1	54.9 (47.8–63.6)	2	43.9 (40.2–47.7)
	Mean ± SD				23.9 ± 20.1		28.1 ± 16.2

*Relative scale of California red scale (CRS) infestation: 0 = no infestation, 1 = very light, 2 = light, 3 = medium, 4 = heavy, 5 = very heavy.

A. lingnanensis was recovered one or two years after releases, and *A. melinus* after 6 summer months (6–8 generations).

Test results were computed by the POLO-PC program for probit analysis (Russell et al., 1977).

Releases. *A. lingnanensis* was collected from large rearing cages (Havron et al., 1987a) after CO_2 anaesthesia, into gauze-covered plastic cups, and released in batches onto leaves or fruits of California red scale-infested trees.

A. melinus was released directly from collection tubes with honey as food, that had been mounted previously for 1–2 days on emergence cages (Havron et al., 1987a). At the release trees, collection tubes holding 100–200 wasps were tied to branches of trees moderately infested with California red scale and opened to allow escape, or else groups of parasites were "sprinkled" onto several adjacent leaves.

At the end of each summer, the *A. lingnanensis* stock population was subjected to an intense selection cycle (5% survival), in order to maintain colony resistance.

RESULTS AND DISCUSSION

A. lingnanensis. Results of pre-release tolerance tests show considerable variability between orchard populations (Table 1). LC_{50} values of several populations, notably No. 8 (from the citron orchard where the selection population originated), indicate partial resistance, acquired under selection pressure of frequent pesticide applications. Post-release tests also show high variability, as would be expected from a mixed population descended from native and introduced strains.

Mean LC_{50} values between orchards increased from 23.9 to 28.1 ppm, but this small difference was not statistically significant. Populations No. 7 and 8, which were relatively resistant in the field from the beginning, were not improved. The number of paired pre- and post-release observations is probably too small to be meaningful (Table 1).

Most releases were made onto low California red scale populations, and released wasps had to compete for hosts with existing unselected *A. lingnanensis* before their superior pesticide resistance could assert itself. Recovered populations may therefore represent small mixed samples of resistant, susceptible or intermediate types. Several seasons of commercial pesticide use may be required before the competitive advantage of the released strain can be ascertained.

TABLE 2
Aphytis melinus releases and recovery

Location	Cultivar	No. of specimens released	CRS infestation (relative scale)*		% of recovered CRS parasites (*A. melinus*)
			Pre–release	Post release	
Ein Gedi	Sweetee	2,500	2	2	72
Gadot	Sweetee	7,500	4–5	2	19
Shuva	Washington	3,500	2	2–3	6
Be'eri	Sweetee	4,000	1	1	0

*See Table 1.

A. melinus. This species, previously imported and established, has recently been reported only from the eastern Jezreel Valley (B.A. Peleg, personal communication). The re-imported, pesticide-resistant strain, released under three different climatic regimes, has so far been recovered from all of them. In hot, dry Ein Gedi the introduced *A. melinus* outnumbered other California red scale parasites (Table 2).

ACKNOWLEDGMENTS

This project was supported by the Citrus Marketing Board of Israel through the Agricultural Research Organization and by research grants from the Hebrew University of Jerusalem. We are grateful to Dr. Y. Rössler and the staff of the Israel Cohen Institute for Biological Control, Rehovot, for logistic support, to Mr. G. Perlman, Ra'anana, for cooperation in the collection of the field-resistant source population, and to Drs. M.A. Hoy and K.M. Spollen of the University of California, Berkeley, who provided the resistant strain of *A. melinus*.

REFERENCES

Croft, B.A. 1990. Arthropod Biological Control Agents and Pesticides. John Wiley & Sons, New York. 723 pp.

Havron, A., Kenan, G. and Rosen, D. 1991. Selection for pesticide resistance in *Aphytis*. II. *A. lingnanensis*, a parasite of the California red scale. *Entomologia Experimentalis et Applicata* 61:229–235.

Havron, A., Rosen, D. and Rössler, Y. 1987a. A test method for pesticide tolerance in minute parasitic Hymenoptera. *Entomophaga* 32:83–95.

Havron, A., Rosen, D., Rössler, Y. and Hillel, J. 1987b. Selection on the male hemizygous genotype in arrhenotokous insects and mites. *Entomophaga* 32:261–268.

Hoy, M.A. 1986. Use of genetic improvement in biological control. *Agriculture Ecosystems & Environment* 15:109–119.

Hoy, M.A. 1990. Genetic improvement of arthropod natural enemies: becoming a conventional tactic? In: New Directions in Biological Control. Edit. R.R. Baker and P.E. Dunn. Alan R. Liss, New York. pp. 405–417.

Javier, P., Havron, A., Rosen, D. and Morallo-Rejesus, B. 1991. Selection for pesticide resistance in *Aphytis*. III. Male selection. *Entomologia Experimentalis et Applicata* 61:237–245.

Kamburov, S. 1974. Studies on the parasites of California red scale, *Aonidiella aurantii* (Maskell), in Israel with emphasis on the introduced parasite *Aspidiotiphagus citrinus* (Craw). Ph.D. Thesis, Hebrew University of Jerusalem. iv + 101 pp. (in Hebrew with English abstract).

Rosen, D. and DeBach, P. 1979. Species of *Aphytis* of the World (Hymenoptera: Aphelinidae). Israel Universities Press, Jerusalem, and W. Junk, The Hague. 801 pp.

Rosenheim, J.A. and Hoy, M.A. 1988. Genetic improvement of a parasitoid biological control agent: artificial selection for insecticide resistance in *Aphytis melinus* (Hymenoptera: Aphelinidae). *Journal of Economic Entomology* 81:1539–1550.

Roush, R.T. 1979. Genetic improvement of parasites. In: Genetics in Relation to Insect Management. Edit. M.A. Hoy and J.J. McKelvey. Rockefeller Foundation Press, New York. pp. 97–105.

Russell, R.M., Robertson, J.L. and Savin, N.E. 1977. POLO: a new computer program for probit analysis. *Bulletin of the Entomological Society of America* 23:209–213.

Spollen, K.M. and Hoy, M.A. 1992. Carbaryl resistance in a laboratory-selected strain of *Aphytis melinus* DeBach (Hymenoptera: Aphelinidae): mode of inheritance and implications for implementation in citrus IPM. *Biological Control* 2:211–217.