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STATUS AND MANAGEMENT OF PYRETHROID RESISTANCE IN THE PREDATORY MITE, AMBLYSEIUS FALLACIS (ACARINA: PHYTOSEIIDAE)¹

B. A. Croft²

ABSTRACT

Low levels of (5–15 fold) resistance to synthetic pyrethroid (SP) insecticides occur in unexposed apple orchard populations of the predatory mite, *Amblyseius fallacis* Garman. Permethrin resistance in one strain has been elevated 60–500 fold by selections in greenhouses. Multiple resistances to DDT and azinphosmethyl are present and cross-resistance to SP-related compounds is generic at 10–250 fold. Permethrin resistance appears due to both hydrolytic esterase and knock down resistance mechanisms. Permethrin resistance appears to be polygenic and more recessive than dominant; it is unstable in the presence of high densities of susceptible immigrant types, but is reasonably stable in the presence of unselected, resistant immigrant types. Successful establishment of SP-resistant mites into SP-treated, commercial apple orchards was monitored using electrophoretic finger-printing techniques over a two year period. Aspects of management of resistance in *A. fallacis* to improve IPM are discussed.

Amblyseius fallacis Garman is a phytoseiid predator of phytophagous mites which occurs on many agricultural crops and has developed strains resistant to a wide variety of pesticides. On apple, endemic strains are resistant to DDT (Smith et al. 1963, Croft et al. 1982), several organophosphates (OP's) (Croft et al. 1976), carbaryl (Croft and Meyer 1973) and permethrin (Strickler and Croft 1981). These developed resistances provide for predator survival in orchards and allow for increased biological control of the spider mites Panonychus ulmi (Koch) and Tetranchyus urticae (Koch) when insecticides are used to control a wide range of orchard insect pests other than mites.

To the synthetic pyrethroids (SP's) (which are only beginning to be used for control of apple pests), resistance in A. fallacis was developed by selection in laboratory experiments (Strickler and Croft 1981, 1982) before widespread use occurred in the field. This was done with intent of avoiding problems associated with development of highly resistant pest mites while predatory phytoseiid mites remained susceptible. Phytoseiid mites usually only develop resistances in the field after resistance in spider mites has occurred and only if the pesticide continues to be used for control of other pests (see Fig. 2, Croft 1982).

In this paper, research to genetically improve A. fallacis by developing and establishing resistant strains in orchards is summarized including (1) background status of resistance in endemic orchard populations of predators, (2) baseline data on the susceptibility and variability of SP resistance, (3) selection of SP resistance in field and laboratory populations, (4) multiple and cross resistances to permethrin, (5) selectivity of SP compounds, (6) the inheritance and stability of SP resistance, (7) the mechanisms of SP resistance, and (8) possible procedures for release and management of SP resistant phytoseiids in the field.

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BACKGROUND STATUS OF RESISTANCE TO SP'S IN A. FALLACIS

Considering the variety of insecticides used in commercial apple production in the USA over the past 30 years (see types in Croft 1981), it was expected that a previously selected potential for resistance to SP's would be present in field populations of *A. fallacis*. It is known for example, that the knock down resistance (kdr) mechanism of DDT resistance is also common to pyrethroid-resistant house flies, certain mosquitos, and the cattle tick. For many years, *A. fallacis* was directly exposed to DDT in the field and resistance eventually developed to this compound (e.g. Oatman 1976, Croft et al. 1982). Other resistance mechanisms such as esterases and mixed-function oxidase (MFO) detoxification enzymes which contribute to OP and carbamate resistance in *A. fallacis* may influence SP resistance (Scott et al. 1983).

To study these resistance relationships, the variability of permethrin susceptibility in 12 field strains of A. fallacis was evaluated to identify strains for use in SP selection experiments (Strickler and Croft 1981). Results of this survey are given for two groups of strains including seven recently collected field populations and five earlier collected, susceptible (S) populations in Table 1. The intrinsic susceptibility level of A. fallacis to permethrin is in the range of .00017-.00043% AI as indicated in seven of 12 strains tested (Table 1). The field collected Fennville, Graham, and Monroe colonies showed only low levels of resistance. The Kleins strain which had never received any exposure to SPs in the field showed a 7.7 fold resistance which likely was due to the use of other insecticides applied in the past (possibly DDT). The Geneva strain which had been exposed to the SPs fenvalerate and permethrin in an experimental orchard for three previous years, showed a near 15-fold permethrin resistance increase. Undoubtedly it, along with the other field strains with more limited resistance, provided the basis for the higher resistance levels achieved by selection in greenhouse experiments (see later discussion).

Table 1. Permethrin LC₅₀ and dosage mortality curve statistics for 12 Amblyseius fallacis populations (adapted from Strickler and Croft 1981).

	Permethrin							
	LC ₅₀ % A.I.	LC ₅₀ Confidence Limit	Slope	Intercept	χ^2	df	Sig. (∝ = .05)	Resistance ^a Ratio
Field Collec	ted Colo	nies						
Collins	.00017	.0000900025	1.31	9.94	5.00	3	N.S.	1
Paw Paw	.00030	.0001500051	.99	8.48	3.46	3	N.S.	1.8
Hudson	.00043	.0002000074	1.17	8.93	6.05	3	N.S.	2.5
Fennville	.00063	.0002500096	2.45	12.86	.59	3	N.S.	3.7
Graham	.00075		1.23	8.84	17.65	3	P> .005	4.4
Kleins	.0013	.000960018	1.42	9.09	1.26	3	N.S.	7.7
Geneva	.0025	.00200032	3.27	13.51	4.31	3	N.S.	14.7
Laboratory	Colonies							
Rose Lake	.00018	.0000900027	1.21	9.56	5,48	3	N.S.	1.1
Rasch &								
Klackle	.00033	.0001700055	1.10	8.81	7.26	3	N.S.	1.9
Garden	.00038	.0002300054	1.84	11.30	.40	3	N.S.	2.2
Composite	.00043	.0002600066	.86	7.90	2.01	3	N.S.	2.5
Monroe	.00083	.000450013	1.30	9.01	1.14	3	N.S.	4.9

^a Fold resistance as compared with the most susceptible colony (Collins).

The results shown in Table 1 shed light on the intrinsic potential and expected selection time or number of treatments required for A. fallacis to develop SP resistance under field conditions. While resistance rates of development would obviously be influenced by previous selection histories, number of applications, degree of orchard isolation, etc, after three years the Geneva strain demonstrated a resistance level of 15-fold when exposed to regular field programs of 4-7 SP applications/season. This resistance level allowed for some survival of these mites at a field dosage (.005-.05%) A.I. range. Hull and Starner (1983) observed similar levels of resistance in Pennsylvania populations of A. fallacis exposure to multiple SPs applications/season for five consecutive years in the field.

SELECTION FOR SP RESISTANCE IN A. FALLACIS

In 1979, a laboratory selection experiment in greenhouses was begun with three populations. The initial two were started with a mix of the strains listed in Table 1 and tests included a repeated permethrin selection (GH-1) and an alternating permethrin-azin-phosmethyl (GH-2) selection test. A third population (Geneva, Table 1), established from a single colony having the highest initial resistance level to permethrin, was also selected repeatedly with permethrin (Strickler and Croft 1982).

In Figure 1, the results of the selection experiments are illustrated; the permethrin treated population (GH-1) showed a 64-fold increase in resistance after about 12 applications, but

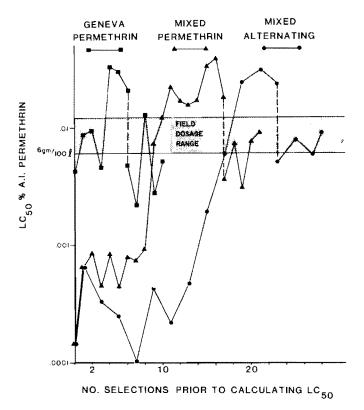


Fig. 1. Selection of permethrin resistance in three strains of Amblyseius fallacis when selected with permethrin and alternating permethrin and azinphosmethyl in greenhouse experiments (after Strickler and Croft 1982).

thereafter leveled off for the next 10 selections. Mites in the alternating, SP-OP selection experiment (GH-2) achieved a similar SP resistance in 18–20 selections (10 permethrin selections). The Geneva strain did not greatly increase its already moderate level of permethrin resistance after 10 selections with permethrin (Fig. 1, Strickler and Croft 1982). Both the permethrin-alone and alternating permethrin-azinphosmethyl (GH-1 and Geneva) selected strains maintained relatively high levels of azinphosmethyl resistance throughout the experiments (Fig. 2, Strickler and Croft 1982).

In summary, the selection results presented in Figure 1 demonstrate increased resistance to permethrin to a level which would provide for survival of mites in the field at the low end of the recommended field rate (.005–.01% A.I.). Unfortunately resistance levels plateaued for each treatment and higher levels were not obtained. While these results indicate a limit to selection of SP resistance in the laboratory, they do not preclude that higher levels might not be achieved in the field when selection was continued over longer time periods and as other modifying mechanisms of resistance became involved.

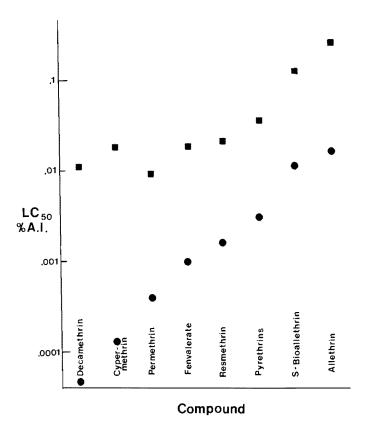


Fig. 2. Cross resistance relationships in a permethrin-resistant and susceptible strain of *Amblyseius fallacis* to seven pyrethroid type insecticides (after Croft et al. 1982).

FEATURES OF CROSS AND MULTIPLE RESISTANCE TO PERMETHRIN

In the comparisons made by Strickler and Croft (1981) there were no negative correlations between permethrin and azinphosmethyl resistance in A. fallacis. In fact, Strickler and Croft (1982) showed a slow increase in azinphosmethyl resistance in the presence of permethrin (-only) selection, indicating a slight positive correlation and suggested that a common mechanism of resistance occurs between these two compounds (possibly esterases? see later discussion).

In Table 2, the levels of multiple resistance to the insecticides azinphosmethyl, DDT and carbaryl in relation to pyrethroid resistant strains of A. fallacis are shown (Croft et al. 1982). The GH-1 strain (permethrin only of Strickler and Croft 1982) had relatively high levels of resistance to azinphosmethyl and DDT in addition to permethrin, but virtually no cross-resistance to carbaryl. The Geneva strain was even more resistant to azinphosmethyl and DDT than GH-1 and moderately resistant to permethrin. Monroe, which had no resistance to azinphosmethyl, had low levels of DDT and permethrin resistance. Collins showed high levels of azinphosmethyl and relatively high DDT resistance, but virtually no permethrin resistance (Table 1). The Fennville strain had moderate to high levels of azinphosmethyl and DDT resistance, a low level of pyrethroid resistance and the highest LC50 value for carbaryl.

Cross-resistance from permethrin to seven pyrethroids in the GH-1 strain (as compared to the Rose Lake strain) was very broad as indicated by LC_{50} values (Fig. 2) ranging from 8–230 fold (Croft et al. 1982). Even a moderate cross-resistance to a mix of natural pyrethrins was manifest (11.8 fold). Comparing structure activity relationships between the various pyrethroids, a pattern was apparent (Fig. 2). In both the resistant and susceptible strains, the -cyanophenoxybenzyl esters of dihalovinyl-chrysanthemic acid (i.e. decamethrin, cypermethrin) were more toxic than the compounds more closely related to the natural pyrethroids (i.e. allethrin). The greatest resistance difference between the susceptible and resistant strains was manifested in the four most toxic pyrethroids.

Table 2. Levels of permethrin, azinphosmethyl, DDT, and carbaryl resistance among nine strains of the predatory mite *Amblyseius fallacis* (after Croft et al. 1982).

Straina	Perme 3.4			osmethyl WP	DE 50 V	-	Carbaryl 50 WP	
	LC ₅₀	Fold-R	LC ₅₀	Fold-R	LC ₅₀	Fold-R	LC ₅₀	Fold-R
Rose Lake	0.00018c	1	0.007°	1	0.032c	1		_
Monroe	0.00083	5	0.010	1	0.208	7		
Composite	0.00043	3	0.013	2	0.088	3		
Collins	0.00017	1	0.18	26	>1.308	$>41^{d}$	0.0125	2
Graham	0.00075	4	0.24	34	0.840	26		
Kleins	0.0013	8	0.09	13	0.131	4		
Geneva	0.0025	15	0.21	30	>1.380	$>43^{d}$	0.0142	2
GH-1	0.022	129	0.11	16	$>1.440^4$	$>45^{d}$	0.0072	1
Fenneville	0.00063	4	0.13	19	0.930	29	0.0196	3

^a See history of strains in Strickler and Croft (1981, 1982).

^b Data taken from Strickler and Croft (1981, 1982).

c Percent A.I. of material in water.

d A conservative estimate based on mortality at 1.2% sol and assuming a maximum slope of 4.0 probits of change/10 fold increase in concentration. Tests with higher dosage solutions gave inconsistent results due to difficulties in maintaining uniform solutions.

In a related study, Croft and Wagner (1982) examined cross-resistance to permethrin between resistant (R) and S strains of A. fallacis to find pyrethroids with selective acaricidal activity on the principal prey of this predator, T. urticae. Figure 3 gives the ld-p line for four experimental pyrethroids in relation to both R and S strains of both species. Only small differences in LC50 values were observed between R (OP) and S strains of T. urticae indicating little cross-resistance to OP's; very low slope values for each compound to this pest species were also observed (Fig. 3, Croft and Wagner 1982). To R (permethrin) and S strains of A. fallacis, ld-p curves showed steeper lines and cross-resistances in the range of 2-46 fold with SD-57706 giving the largest difference.

With regard to selectivity, fluvalenate and especially NIC 85913 showed little potential; however, SD-57706 and ZR 3903 at the lower concentration levels of 0.001-.025% A.I. were more toxic to adult prey than predatory mites which demonstrated a potential for selectivity with these types of pyrethroid compounds. Additional research is needed to identify SP

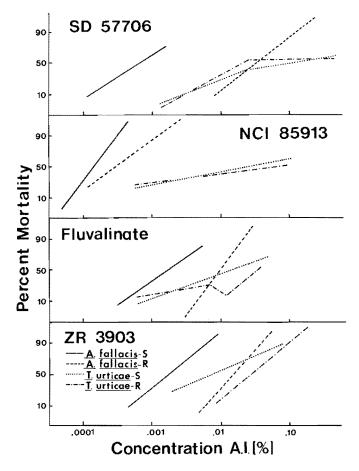


Fig. 3. LC₅₀ response curves for permethrin-resistant (R) and -susceptible (S) strains of A. fallacis and organophosphate-resistant and -susceptible strains of T. uritcae to four pyrethroid insecticides.

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compounds that are more completely selective to the predators over their prey while at the same time maintaining their useful insecticidal properties for controlling other key pests of agricultural crops (see further discussion of means to accomplish this selectivity for apple pests in Croft and Wagner [1982], Croft [1981, 1982]).

MECHANISMS OF RESISTANCE

Data of Table 2 indicate that several mechanisms of insecticide resistance are present in field strains of A. fallacis. For example, several biochemical mechanisms of DDT (e.g. DDT-dehydrochlorinase, Mixed-Function Oxidase [MFO] and pyrethroid (kdr, MFO, esterase) resistance are known. Only kdr and MFO are common to both groups of compounds. In the data shown in Table 2, there are strains which are both DDT + pyrethroid resistant (GH-1) and others which are DDT resistant, but SP susceptible (Collins). Also, other mechanisms of resistance common to OP's could confer contributing mechanisms to pyrethroid resistance (e.g. esterase, MFO). Such diversity in mechanisms of resistance might be expected in populations from orchards which have been exposed for 30 years to a wide range of insecticides.

One apparent conclusion from the data in Table 2 is that the MFO's, which are common mechanisms of carbaryl resistance, are not a major factor in these strains of A. fallacis (Table 2). LC₅₀ values to carbaryl were similar for all strains tested and near the values observed for a wide variety of S strains as reported by Croft and Meyer (1973) and Croft and Hoying (1975).

To evaluate possible mechanisms of resistance in A. fallacis, Scott et al. (1983) studied toxicity and synergized responses of several strains of this predator (i.e. those listed in Table 2). Using the synergists pipernyl butoxide and DEF in LC50 studies with methoxychlor, they found that most strains showed only low levels of oxidative activity further indicating that MFO's were not the major mechanism of resistance observed (with the possible exception of carbaryl resistance in the Fennville strain, Table 3). They concluded that resistance in certain strains to both DDT and permethrin was due primarily to the kdr mechanism (e.g. GH-1, Fennville) while in others resistances to these compounds may be related to DDT dehydrochlorinase (e.g. Geneva, Collins). Hydrolytic esterases played a significant role in both OP and SP resistance in the GH-1 strain and to a lesser degree in the Fennville and Geneva strains.

In related biochemical experiments, Mullin et al. (1982) examined basic differences in whole body enzyme levels in S and R strains of T. urticae (OP) and A. fallacis (SP) (Table 4). As measured by conversion of aldrin to dieldrin epoxidase, MFO activity was ca. 5 times lower in the susceptible strains of predator than prey mites. However, there were no differences in degradation of aldrin epoxidase between R (GH-1) and S (Rose Lake) strains of A. fallacis which is consistent with the data from synergist studies with respect to MFO activities (Table 4). With cytosolic cis epoxide hydrolase (that fraction usually associated with food substrate detoxification), prey levels of enzyme were significantly higher than predator levels. Between SP R and S strains of predators there also were significant differences in enzyme levels (Table 4). Glutathione transferase was much more common in predator vs. prey mites with significantly higher levels present in resistant vs. susceptible strains of the predator. With esterase levels, S prey and predator strains had similar amounts, but the R predator strain showed much higher levels than the S strain again confirming that either SP or OP resistance or both resistances is due to higher levels of these enzymes.

Considering the differential response of *T. urticae* vs. *A. fallacis* in detoxification potentials (Table 4), data indicate possible factors contributing to resistance and to the overall generally greater susceptibility of the entomophagous species as compared to its prey. Data also give clues to possible means for exploiting selectivity differences between the two species (e.g. compare MFO vs. glutathione transferase, see further discussion in Mullin et al. [1982]).

Table 3. Toxicity and synergized compound studies of mechanisms of insecticide resistance in six strains of the predatory mite Amblyseius fallicis.

	LC50 Toxicity Studies						LT ₅₀ Synergist Studies					
	Compounds					Compound and Synergist						
	DDT	Methoxy chlor	Permethrin	Azinphos- methyl	Carbaryl	DDT + PBB	Methoxy. + PBB	Azinphos. + PBB ^c	Azinphos. + DEF	Permethrin + DEF		
Rose Lake	1.0(.032)	1.0(.068)	1.0(.0002)	1(.018)	1(.009)	NSb	NS	NS	NS	NS		
Collins	4 1a	6	1	26		NS	NS	*	*	NS		
Kleins	4	2	8	13		NS	NS	NS	*	NS		
Geneva	43	3	15	30	2	NS	NS	+e	*	+		
GH-1	45	18	50	16	1	NS	NS	NS	*	*		
Fennville	29	13	4	19	3	*d	*	NS	NS	*		

a Fold resistance level over the susceptible—Rose Lake strains (LC50 in percent A.I.)

b Significance of difference between synergized/non-synergized compound at a 1:3 ratio of synergists to toxicant, NS = non significant from 1:1 ratio.

cLT25 values instead of LT50.

d Significantly greater than 1.0 at P ≤ 0.05 level

e Significantly greater than 1.0 at P ≤ 0.10 level

Table 4. Detoxification capability of the predatory mite *Amblyseius fallacis* and its prey *Tetranychus urticae*. Activity is expressed in pmol/min-mg protein for a combined microsomal plus cytosolic fraction; $\bar{x} \pm SD$ from 3-5 separate enzyme preparations.

Mite Species		F	Enzyme Activ	ity						
Strain	Aldrin	Epoxide I	Hydrolase	Glutathione	Esterase					
	Epoxidase	trans	cis	Transferase	X 10-3					
A. fallacis										
S	0.27	278	431a	1095 ^b	318					
	± 0.22	± 49	± 109	± 495	± 68					
R	0.23	600 ^f	834c	1683 ^d	1788 ^f					
	± 0.14	± 55	±268	± 249	\pm 341					
T. urticae										
S	1.44 ^b	1587¢	117	102	389d					
	± 0.25	± 74	± 20	± 19	± 30					
R	1.60	1613	124	219e	263					
	± 0.36	± 178	± 32	± 27	± 78					

Interspecific difference between susceptible strains: ^a for P < 0.01; ^b for P < 0.005; ^c for P < 0.001. Intraspecific difference: ^d for P < 0.01; ^e for P < 0.05; ^f for P < 0.005. Significance is indicated for strain with the higher enzyme level.

GENETIC ANALYSIS

In Figure 4, the ld-p lines to permethrin for individual R and S parent strains, the F or parent cross (combined) and backcross (combined) matings of R and S strains of A. fallacis are presented; statistical properties of these lines are summarized in Table 5 (from Croft and Whalon 1982). Parental strain LC50 values were 0.017 and 0.026% A.I. and 0.000072 and 0.00039% permethrin in water, respectively for R and S strains at the beginning and end of the test. The F cross hybrid LC50 was 0.00051% A.I., which was intermediate in susceptibility, but more close to the susceptible parent line than to the resistant strain response curve (Fig. 4). The backcross ld-p line gave an LC50 value of 0.00062% A.I. which was very near the value of the susceptible strain indicating a recessive genetic basis for the resistance. The ld-p line for the backcross did not show a flattened slope in the mid-dosage range, indicative of a monogenic relationship (i.e. reflecting a 1:2:1 ratio in the resistance response).

These data are very similar to those reported for crosses made between permethrin resistant and susceptible strains of *Metaseiulus occidentalis* (Nesbitt) by Hoy et al. (1980). They observed a polygenic, recessive type response in parent and the subsequent backcrosses and hypothesized the SP resistance would likely be unstable in the field in the presence of large populations of immigrant susceptible types.

RELEASE EXPERIMENTS INTO COMMERCIAL APPLE ORCHARDS—1980

Three strains of A. fallacis including the two permethrin resistant strains GH-1 and Geneva, and a susceptible strain (Composite, Table 1) were released into a 0.8-ha commercial apple orchard near Fennville, Michigan, in 1980. Release treatments were replicated in six trees and there were four release treatments including a check which contained populations of the indigenous strain of predators. Predator and prey were followed during the growing season by randomly selecting 50 leaves/tree at 7-14 day intervals throughout the season. A total of 2,500 mites were released/tree in late June to early July. A total of three permethrin and one fenvalerate sprays at relatively high rates were applied to the trees and surrounding vegetation during the growing season (Whalon et al. 1982).

In Table 6, the densities of predatory mites found at each sample date are given for the four treatments. In early season (May-June), virtually no indigenous mites were present except on non-treated vegetation surrounding the orchard.

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Table 5. LC₅₀ values to permethrin of genetic cross between resistant and susceptible strains of the predatory mite *Amblyseius fallacis*.

Strain-Date Cross	LC ₅₀ ^a	Slope	(C.I. (95%))a	Fold Rb
Resistant 24/6/81 (GH-1)	.017	1.53	.012023	435.9
Resistant 16/9/81 (GH-1)	.026	1.65	.019036	666.7
Susceptible 24/6/81 (Collins)	.000072	1.80	.00004700010	1.8
Susceptible 16/9/81 (Collins)	.000039	1.75	.000025000053	1.0
Parent Cross 24/6/81 $R \stackrel{\circ}{\circ} \times S \stackrel{\circ}{\circ} \text{combined}$ $R \stackrel{\circ}{\circ} \times S \stackrel{\circ}{\circ}$.00051	1.90	.0004100065	13.1
Backcross 16/9/81 (combined)	.000062	1.40	.000039000085	1.6

^a Express as percent A.I. concentration in water.

The first application (197.6 ml/ha) of permethrin of 23 July, after the predator releases, represented approximately 1/8 the maximum recommended field rate or 1/2 the average recommended full rate. This treatment reduced the susceptible predatory mite populations from an average of 2.60 to 0.16 mites/leaf by 4 August. Several predator mites were collected in the indigenous or check trees on 25 July. Microelectrophoresis analysis indicated that they were either GH-1, Geneva, or a hybrid of GH-1 and Geneva strains (see later discussion). Both resistant released strains were generally unaffected by the first low-rate application of permethrin (Table 6).

The next application of permethrin (1780 ml/ha) on 7 August represented a full recommended field rate and it virtually eliminated the susceptible strains from all six replicate trees (Table 6). Predators collected in these trees after 11 August were either GH-1, Geneva, or possible hybrids of these two strains. This full-rate permethrin application reduced both the GH-1 and Geneva strains. There was no significant ($P \le 0.05$) difference between the GH-1 and Geneva strains before or after the permethrin treatments. Since both strains survived the field application rate of permethrin, it is likely that they could survive similar rates in commercial orchards.

Fenvalerate was applied at the recommended full field rate of 1482 ml/ha on 3 September (Table 6). The GH-1 strain survived and increased from 0.59 to 0.68 mites/leaf after this application, but the Geneva strain declined and was all but eliminated by 22 September. It is not clear if the Geneva strain lacked the fenvalerate cross-resistance potential exhibited by the GH-1 strain (Table 1) (Croft et al. 1982) or whether other circumstances like a disadapted diapause response or possibly rapid reversion to susceptibility contributed to this poor survival.

The characteristic microelectrophoresis-carboxylesterase banding patterns for each of the predatory strains is presented in Figure 5. Measurements were made from the sample start position to the approximate center of each band. In all, seven strains tested produced seven distinct bands: B at 4.4 cm, C at 4.8, D at 5.3, E at 5.7, F at 6.1, G at 6.6, and H at 8.2–10.0 cm. The run front was at 19.0 cm and the gel end at 20.0. Bands at A (between 2.6–4.3 cm) have been associated with T. urticae prey enzymes. While there was some variation between individual mites of the same strain (particularly in intensity), all strains except the Composite Susceptible, produced bands at position D. The Indigenous strain was similar to

^bIn comparison to susceptible (Collins strain) 16/9/81 test.

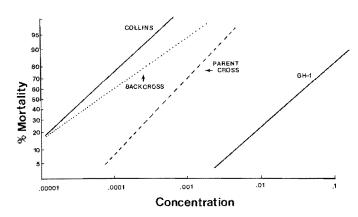


Fig. 4. Log-concentration mortality responses of parent resistant (GH-1) susceptible (Collins), parent cross and backcrossed populations of *Amblyseius fallacis* to permethrin (percent A.I.).

Table 6. The synthetic pyrethroid application dates and rates, predatory mite release dates, mite sample dates and average predatory mite numbers/leaf following releases of *A. fallacis* (SP resistant) into a commercial apple orchard.^a

		Predatory Mite Strains ^b							
Date	Application or Release	GH-1	Geneva	Susceptible	Indigenous				
10-7	SAMPLE	0.85 ± 0.42	0.82 ± 0.24	0.42 ± 0.63	0.0				
16-7	SAMPLE	1.43 ± 1.23	1.76 ± 1.33	2.60 ± 1.96	0.0				
23-7	PERMETHRIN		*****						
	200 ml/ha								
25-7	SAMPLE	1.85 ± 1.53	0.59 ± 1.84	0.22 ± 0.15	$0.01 \pm (-)$				
4-8	SAMPLE	1.33 ± 0.77	1.73 ± 1.55	0.16 ± 0.78	0.00				
7-8	PERMETHRIN								
	780 ml/ha								
11-8	SAMPLE	0.86 ± 0.67	0.63 ± 1.34	0.00	0.41 ± 0.33				
27-8	SAMPLE	0.49 ± 0.21	0.43 ± 1.41	0.21 ± 0.61	0.17 ± 0.63				
3-9	FENVALERATE								
	1482 ml/ha								
12-9	SAMPLE	0.59 ± 0.75	0.16 ± 0.71	0.11 ± 0.70	0.11 ± 0.48				
22-9	SAMPLE	0.63 ± 0.38	$0.05 \pm (-)$	0.01 ± 0.25	0.0				

^a Permethrin (1780 ml/ha) was applied on 29/5 and 21/6/1981 while Fenvalerate (2500 ml/ha) was applied on 15/6. Mites were sampled on 21/6 and 28/6 after these applications and no Amblyseius fallacis were found in the trees or orchard ground cover.

^b GH-1 was released on 28/6/1981, while Geneva and the susceptible strains were released on 3/7.

the susceptible, but produced a faint but definite band at D. The Composite Susceptible strain demonstrated almost no esterase activity at all, except for a variable intensity at C and H. The two resistant, released strains, Geneva and GH-1, as well as the overwintered strain (1981 Field Collected), produced very similar patterns with bands at C, D, E, F, and G. Several of the strains (Composite Susceptible, 1981 Field Collected and Geneva) also produced a clear band at H, however the bands of the resistant strains were much darker than in the Indigenous, Composite Susceptible or Susceptible, indicating a greater quantity of enzyme or perhaps two or more unseparated bands.

The microelectrophoretic technique provided a useful tool in identifying the resistance origins of individual predatory mites. We were able to confirm the survival of the mites originating from the Geneva or GH-1 strains (these strains could not be separated by enzyme banding studies) in all the replicated release trees following both applications of permethrin. From population studies it appeared that only the GH-1 strain persisted following a late season fenvalerate treatment. The Indigenous predatory mites were not detected within the synthetic pyrethroid block after 21 June, but were readily collected from surrounding azinphosmethyl treated blocks. The Susceptible strain survived the first application of permethrin at low rates, but was not detected after the second full-rate permethrin spray or first full-rate fenvalerate applications. The predators found in the Indigenous and Susceptible treatment trees (Table 2) after 25 July and 27 August, respectively, exhibited banding patterns characteristic of the GH-1 or Geneva strains. These individuals were probably dispersing from nearby GH-1 and Geneva release trees. Several of the predatory mites found especially late in the growing season exhibited uncharacteristic banding patterns and we hypothesize that these individuals were either long range dispersers or hybrids of the various strains.

FIELD STUDIES-1981

Predator populations in the experimental release plots were again followed both by electrophoretic esterase evaluation and dosage-mortality assessments in 1981 (Croft and Whalon 1983). Again SP's were applied to the block, but only once in early season in the entire block. Predator assessments were made in early, mid, and late season.

In Table 7, the LC₅₀ values for four strains:(1) released SP resistant (GH-1), (2) released Susceptible, (3) Indigenous, and (4) subsequently Field Collected mites, from the perme-

Table 7. Summary of LC₅₀ values for indigenous, released and subsequently collected field populations of *Amblyseius fallacis* occurring in a commercial apple orchard treated with permethrin (Fennville, MI, 1981).

Strain (Date of Collection)	LC ₅₀ a	CI (95%)a	Slope	Resistance Levelb
Released and Indigenous St	rains (1980))		
Resistant-Released	.0090	0.0070011	1.94	53
Susceptible-Released	.00017	.0000900025	1.31	1
Indigenous	.0003	.0001100057	1.73	2
Field Collected Strains (198	1)			
08/5	.0034	.00240043	1.96	20
15/6	.016	.013024	1.25	94
05/9	.00077	.0010005	1.08	4

^a In percent A.I. permethrin in water.

b As compared to the susceptible released strain.

1983

thrin-treated, commercial apple orchards are summarized. In early spring 1981, resistant mites were present in the release orchard following the overwintering period; the LC_{50} response of 0.0034%, A.I. permethrin was somewhat lower than that of the original release population, but still 20 and 10 fold higher than the Susceptible released and Indigenous strains, respectively. By midseason following selection with a field application of permethrin at (70.2g A.I./ha.), the LC_{50} value of the released mites was even higher than that of the original release strain (Table 7, LC_{50} value = 0.016% A.I. permethrin).

Johnson and Croft (1981) have shown that orchards typically are exposed to high densities of migrating A. fallacis in late season. In the absence of subsequent sprays, predator populations in the experimental orchard were undoubtedly influenced by a large influx in SP-susceptible immigrant predators which hybridized with the resistant mites. Thereafter the recessive nature of the resistance when hybridization occured was manifested in the field LC_{50} of collected mites (LC_{50} value = 0.00077% A.I., Table 7). Electrophoretic studies to fingerprint the esterase detoxification enzymes associated with each of these three groups of predators collected in the field provided further evidence of these conclusions.

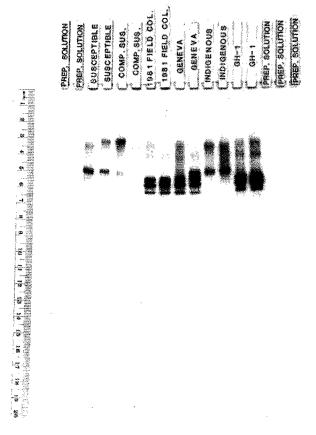


Fig. 5. Microelectreophoretic banding patterns in susceptible, released -susceptible, released-resistant indigenous and recovered populations of the predatory mite, Amblyseius fallacis, made in a commercial apple orchard (1980–1981).

LABORATORY STUDIES

SP-resistant predators when surrounded by similar resistant, but unexposed predator populations, immigrated to plants without predators at daily rates of 0.005–0.03 individuals/leaf (these levels would be somewhat higher than those found in orchards, except late in the growing season [Johnson and Croft 1981]). Resistance in these colonies to both permethrin and azinphosmethyl (a resistance which is due to a relative dominant, single gene factor in A. fallacis [Croft et al. 1976]) in the absence of selection remained relatively high for at least 25 generations or ca. one year (Fig. 6). In laboratory tests where immigration did not occur (Fig. 6), a similar pattern of resistance was observed. This further confirmed that SP resistance in A. fallacis was reasonably stable over time in the absence of hybridization with S types. These data indicate that there was no lack of fitness or other properties of genetic instability in the SP resistant mites tested in these experiments under the conditions of laboratory rearing and selection. Possibly similar degrees of fitness and resistance would be observed in the field provided that large regional SP resistant predator populations were present (as in the case of OP resistant strains).

In conclusion, permethrin resistance in our selected strain of A. fallacis appears to be a recessive, polygenic trait which is highly susceptible to reversion toward susceptibility following hybridization with immigrant susceptible types. In this study, reversion occurred even after applying a single permethrin application early in the growing season before most spider mite-predator interactions had occurred. This type of limited use would be preferable

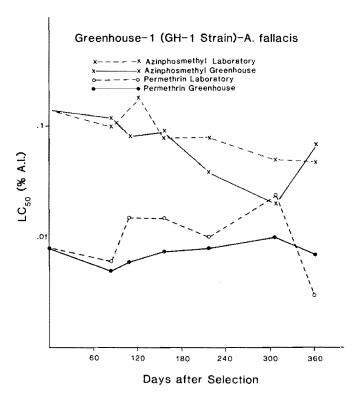


Fig. 6. LC₅₀ values for the Greenhouse-1 (GH-1) strain when left untreated and in the presence and absence of immigrant permethrin-resistant A. fallacis over a one-year period. (percent A.I. permethrin).

for resistance management and IPM programs (Croft and Hoyt 1978, Croft 1982). If SPs were used more extensively on a routine basis (a use not currently recommended for IPM), then large regional populations of SP resistant predators would probably develop in 3-5 years (Strickler and Croft 1981, 1982; Hull and Starner 1983); however, it is feared that pest resistances in a species like the codling moth Cydia pomonella L. could also develop due to more extensive use (Croft and Hoyt 1978). A more desirable compromise might be to use the SP's twice during the season; once in early season to control such troublesome pests as the spotted tentiform leafminer (Phylonorcycter blancardella Fabricius) and tarnished plant bug (Lygus lineolaris Palisot deBeauvois) and once later after the principal predator-prey mite interactions have occurred in mid to late season (i.e. during early August). At this time, second generation codling moth and apple maggot (Rhagoletis pomonella Walsh) populations could be suppressed with the SP's. This late-season spray should also be timed, if possible, to affect susceptible immigrant populations of A. fallacis which usually disperse into the orchard in great numbers in late August and September (Johnson and Croft 1981). The early and late spray would thus bracket the intense period of susceptible predator influx and insure that SP resistance in predatory mites was maximally maintained in the orchard, hopefully without inducing high levels of SP resistance in other orchard pests.

Another possibility for improving and managing SP resistance in A. fallacis would be to select out a single-gene, more dominant basis for this trait. Geneticists have suggested that by using high SP concentrations (equivalent to field rates) from the beginning of selection trials, rather than slow incremental increases in concentration as were used in these studies (see Strickler and Croft 1982), a more stable resistance of hybridization could be obtained. Further research is needed to evaluate this hypothesis as well as to survey field populations for more stable SP resistance traits which may be developed as these compounds are more widely used.

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