Manure accumulations are major sources of house fly problems in the Chino Basin, where 375 large dairies are concentrated. Baited jug traps (right) were used to measure fly populations resulting from treatments.

Effect of permethrin on house fly resistance

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Resistance has developed on several southern California dairies that have had to rely on regular permethrin treatments

The Chino Basin in southern California is one of the most concentrated dairying areas in the world. Approximately 375 dairies, averaging 620 cows each, are crowded into about 30 square miles. This area also supports one of the fastest growing human populations in the United States.

All the dairies practice confinement-type management, which is undoubtedly the most efficient method of producing milk. But the manure accumulations are tremendous and provide ideal sites for development of large populations of the house fly, *Musca domestica* L. Fly control on most dairies is accomplished with insecticides, often applied by a licensed pest control operator. The typical house fly control program consists of permethrin sprays every week or two weeks applied to animal shelters, storage buildings, and other fly resting sites; synergized pyrethrin mists are occasionally applied near mangers and calf stalls.

Permethrin was a welcome addition to existing fly control chemicals in the late 1970s, since it represented a new class of insecticide that had long residual life and was effective against flies resistant to organophosphates. After several years of commercial use, however, reports began to be heard of failure of permethrin to control house flies. Fly resistance to pyrethroids has since been documented in a number of countries, and in the state of Georgia.

In view of the intensive permethrin pressure on the house fly population in the Chino Basin, and the practical consequences of resistance in this densely populated area, we designed a study to evaluate the susceptibility of these flies to permethrin.

Resistance tests

Adult house flies were sampled from eight dairies in the Chino Basin on November 30, 1984. The dairies, referred to here by initials, contained varying numbers of cows, ranging from 650 to 1,400.

Two of these dairies (VB, CD) practiced little or no chemical fly control, except for periodic use of fly baits formulated with methomyl or bomyl. They served as controls.

The remaining six dairies (J1, J2, TP, DD, PC, EX) were under routine treatment for adult flies, consisting of residual sprays of permethrin wettable powder and space sprays of synergized pyrethrins (containing an added compound to overcome resistance). Pest control operators applied all treatments: the residual sprays to the outside walls of buildings and other fly resting areas; pyrethrin mist sprays near feed storage areas and calf mangers. During 1983 and 1984, each of the six dairies had received ten such treatments at approximately two-week intervals between early May and mid-October. Each dairy had been treated in this manner by the same pest control operator.

We determined permethrin resistance levels in the laboratory, treating first-generation adult females by topical application of insecticide solutions in acetone, under

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carbon dioxide anesthesia, according to standard methods. All tests were repeated five times, using at least five different dosages of insecticide or of insecticide and piperonyl butoxide synergist. Identical tests were performed on a susceptible laboratory strain of house flies.

Resistance ratios at LD_{95} (dosage that killed 95 percent of the test population) ranged from 15.2 on dairy CD to 185.2 on J1 (table 1). The low resistance level at CD was expected, since this dairy had not received any applications of permethrin for at least three years before sampling. The exceptionally high resistance level at J1 was surprising, because this dairy had not received a larger number of treatments than the other dairies. Furthermore, J1 is only a mile from J2 or TP, well within the dispersal range of house flies. The sampling at J1 was not biased in favor of resistant survivors, since the collection was made six weeks after permethrin treatments had been discontinued for the season.

Several mechanisms can be responsible for house fly resistance to permethrin. These include increased rate of permethrin breakdown by enzymes, insensitivity of the fly's target site (its nervous system), and decreased penetration through the fly's cuticle. The respective house fly populations were tested with permethrin in combination with piperonyl butoxide to determine indirectly whether an enzyme system was involved in the observed resistance. The use of piperonyl butoxide considerably increased the toxicity of permethrin to each population tested and significantly increased the dose-mortality relationship (table 1), indicating that the major portion of the resistance was due to enzyme action. Synergism ratios ranged from 12.4 on dairy DD to 34.3 on J1, and were significantly correlated to the resistance ratio (r=0.91). One or more additional mechanisms are presumed to be responsible for the resistance (13.5-fold on J1) remaining after synergism by piperonyl butoxide.

Stability of resistance

House flies collected from dairy J1 in November 1984 were reared in the laboratory without selection pressure until July 1985, when dose-mortality was again compared to determine the stability of resistance. The results indicated that resistance had decreased moderately during that interval.

We then selected this strain with permethrin at about the 85 percent mortality level for two consecutive generations to learn if the strain can quickly develop higher resistance. The bioassay procedures in the selection studies were the same as those previously described. Mortality determinations were made 24 and 48 hours after treatment. Permethrin resistance levels were determined for the parental, first, second, and fourth generations. Data from all bioassay and selection studies were subjected to statistical (probit) analysis.

This selection process increased the lethal dosage required (LD_{95}) in this strain to 22.90 micrograms per fly in the first generation and 53.82 micrograms in the second generation, about a 12.5-fold increase.

Rearing a subcolony of the second generation without selection again revealed partial instability of resistance. The LD_{95} declined within two generations to 15.05 micrograms per fly.

The owners of the J1 dairy chose not to use permethrin during 1985, primarily because of inadequate control. In fact, they abandoned all chemical treatments except for the use of fly baits made with methomyl. Their elimination of permethrin that year gave us the opportunity to test for a possible decrease in permethrin resistance. Adult house flies were sampled in early December 1985, a year after the initial collection.

We also collected adult flies from the PC dairy in late November 1985. Collections a year earlier had shown the PC dairy to have the second highest resistance level after J1. PC, however, remained under permethrin treatment by the same pest control operator during 1985, and so had been under selection pressure for three consecutive years (1983-85).

Bioassays were conducted on these two collections, and on the standard susceptible laboratory strain. The results revealed a very strong decrease in the resistance level of the J1 population, with a drop in the LD_{95} value from 9.26 to 1.84 grams per fly between 1984 and 1985. In contrast, the PC strain showed little change in resistance after an additional year of permethrin use; the LD_{50} (permethrin dosage lethal to 50 percent of the test population) increased slightly from 0.21 to 0.29 microgram per fly, but the LD_{95} decreased from 1.65 to 1.27 micrograms per fly.

Mortality at 24 vs. 48 hours

The results reported thus far are based on mortality counts 24 hours after treatment. As a possible indicator of the presence of a

TABLE 1. Toxicity of permethrin and and permethrin plus piperonyl butoxide (PB) (1:5) to various strains of house flies collected from dairies in the Chino Basin, California

Dairy, and herd size	Permethrin		Permethrin + PB		
	LD_{95}	RR*	LD_{95}	RR*	SR†
	µg/fly		*µg/fly*		
NAIDM‡	0.05	—	0.02	—	2.5
CD, 650	0.76	15.2	0.04	2.0	19.0
EX, 750	0.80	16.0	0.06	3.0	13.3
DD, 1,080	0.87	17.4	0.07	3.5	12.4
VB, 725	1.14	22.8	0.06	3.0	19.0
J2, 950	1.39	27.8	0.09	4.5	15.4
TP, 650	1.63	32.6	0.12	6.0	13.6
PC, 1,400	1.65	33.0	0.08	4.0	20.6
J1, 720	9.26	185.2	0.27	13.5	34.3

* RR (resistance ratio) = LD_{95} field strain/LD_{95} susceptible strain. ‡ Susceptible laboratory strain of house flies.
† SR (synergism ratio) = LD_{95} unsynergized/LD_{95} synergized.

nervous-system-insensitivity type of resistance, mortality counts for the tests on the J1 population were also taken 48 hours after treatment. In every case, the flies exhibited a higher degree of survival at 48 hours, indicating a significant degree of recovery from knockdown. The average LD_{95} was about 50 percent greater at 48 than at 24 hours.

It is presumed that, by raising the threshold at which knockdown is initiated or terminated, the insensitivity mechanism extends the period over which recovery is possible through metabolic breakdown of the chemical. Thus, results of permethrin treatment would also be misleading in the field, if estimates of kill were limited to the first 24 hours after an application.

Synergized permethrin

The results of these laboratory studies suggested that synergizing permethrin with piperonyl butoxide might offer a means of restoring the toxicity of permethrin against field populations of house flies. The 1:5 mixture of permethrin and piperonyl butoxide was toxic even to the most resistant fly strain.

We selected three dairies from the Chino Basin (J1, PC, CD) and three from the Sun City area (MP, AB, DJ), 37 miles southeast of Chino Basin, to compare the effects of permethrin and synergized permethrin on fly density and resistance levels. One dairy in each area received periodic applications of either permethrin wettable powder or synergized permethrin. The third dairy in each area received no insecticidal treatments other than periodic applications of fly baits formulated with sugar and 1 percent methomyl (table 2).

TABLE 2. Relative herd size and treatment interval for dairies cooperating in field evaluation of synergized permethrin

Dairy, and herd size	Treatment	No. applications each treatment*
Chino Basin		
J1, 720	Permethrin	6
PC, 1,400	Permethrin + PB	22
CD, 650	Control	—
Sun City		
MP, 350	Permethrin	6
AB, 1,150	Permetrhin + PB	7
DJ, 980	Control	—

* Treatments applied at periodic intervals between June and October.

Treatments were begun on each facility in mid-June and continued periodically until late October 1986. Samples of adult house flies were collected from each dairy before and after the treatments. Resistance levels were determined for each collection according to the previously described methods. Adult fly populations were measured every week with baited jug traps, three of which were placed on each dairy near areas where flies congregated.

In Chino Basin, the CD (control) dairy generally had greater numbers of flies per trap than either J1 (permethrin) or PC (synergized permethrin) (fig. la). The addition of piperonyl butoxide to the permethrin had no obvious effect on fly densities on the PC dairy over the life of the study.

Results from Sun City dairies were quite different. The MP (permethrin) dairy had relatively low numbers of flies compared with the AB (synergized permethrin) and DJ (control) dairies (fig. 1b). The addition of piperonyl butoxide had no effect on the AB fly population. which tended to coincide with population fluctuations at the CD dairy.

Permethrin resistance levels remained relatively stable between June and November, dropping slightly at four of the test sites. Resistance ratios for each collection in June were similar, ranging from 17.3 to 30.6 (table 3). Results from November bioassays were surprisingly similar, with resistance levels dropping on five of six dairies, even after multiple applications of permethrin.

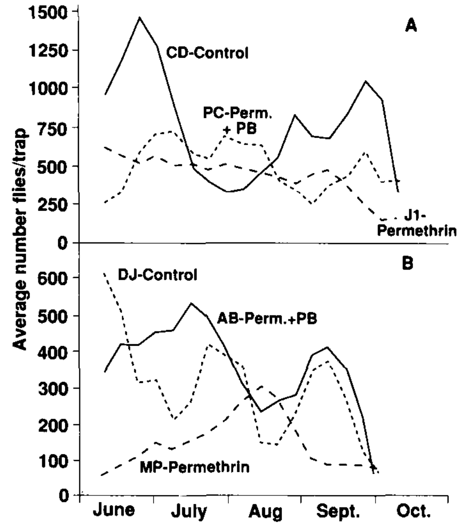

Fig. 1. Although results differed in the Chino Basin (A) and Sun City (B), the addition of piperonyl butoxide (PB) to permethrin had little effect on house fly densities at either location.

TABLE 3. Resistance level of six field strains of house flies, bioassayed (female flies) with permethrin and permethrin plus piperonyl butoxide (PB) (1:5), before (June) and after (November) various field treatments

Dairy	Treatment	Sample date	Permethrin LD_{95}	Permethrin Slope	Permethrin RR*	Permethrin + PB LD_{95}	Permethrin + PB Slope	Permethrin + PB RR*
			μg/fly			μg/fly		
NAIDM†	—	June	0.066	3.26	—	0.002	6.66	—
		November	0.072	3.85	—	0.006	3.46	—
J1	Permethrin	June	2.021	2.36	30.6	0.018	2.75	6.9
		November	1.050	1.92	15.0	0.011	2.98	1.8
PC	Permethrin + PB	June	1.143	2.14	17.3	0.017	3.48	6.5
		November	1.531	1.89	21.8	0.013	3.31	2.2
CD	Control	June	1.471	1.73	21.1	0.009	3.21	3.5
		November	1.138	1.79	16.3	0.015	2.82	2.5
MP	Permethrin	June	1.475	1.83	22.4	0.012	3.41	4.6
		November	1.527	1.76	12.4	0.007	2.56	1.2
AB	Permethrin + PB	June	1.390	1.80	21.1	0.017	2.42	6.5
		November	0.906	1.97	13.0	0.006	2.49	1.0
DJ	Control	June	1.218	1.91	18.5	0.005	3.95	1.9
		November	0.869	1.86	12.4	0.005	3.63	0.8

* RR (resistance ratio) = LD_{95} field strain/LD_{95} susceptible strain.
† Susceptible laboratory strain of house flies.

Conclusions

The addition of piperonyl butoxide to permethrin gave no indication of improved control of field populations of house flies, despite the enhanced toxicity shown in laboratory tests. The ineffectiveness of piperonyl butoxide on the dairy may be attributed to its chemical instability under field conditions. Permethrin has a long residual life and is not affected by light, whereas piperonyl butoxide is easily broken down by sunlight.

The current resistance problem is a direct result of the lack of effective integrated pest control technologies for dairies. The size, intensity, and concentration of dairies in the Chino Basin almost preclude the reliance on manure management for fly control. Biological control of house flies on dairies with commercial parasitic wasps may be impractical because of the large numbers of parasites necessary to reduce fly populations. Also, integrated pest management programs would have to be implemented on an areawide basis, making it necessary to have the cooperation of several hundred dairies. Continued research with present IPM technologies, or new technologies as yet unforeseen, may offer some workable solutions to dairy fly problems.

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