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ORGANOPHOSPHORUS TOLERANCE IN *CULEX QUINQUEFASCIATUS* IN TEXAS

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ABSTRACT. Twelve collections of *Culex quinquefasciatus* from separate areas of Galveston County, Texas and 8 from various parts of Brazoria County were established as temporary strains in the laboratory for 2 or 3 generations to provide sufficient material for insecticide susceptibility tests. As compared with a standard susceptible strain (UTMB), the LC_{50}'s for malathion of 9 strains of larvae from Galveston County ranged from 0.0058 to 0.280 ppm or a maximum of 7X less susceptible. The LD_{50}'s of the adults ranged from 0.9% to > 5.0% malathion. Five strains of larvae from Brazoria County were 2X to 9X more tolerant to malathion than the standard, whereas the LD_{50}'s of the 8 strains of adults ranged from 1.3% to > 5.0%. Larvae from both counties exhibited tolerance to chlorpyrifos, ranging from 2X to 13X as compared with a susceptible strain. The larvae of 2 strains tested against a battery of other organophosphorus insecticides not used in mosquito control, exhibited decreased susceptibility which in the case of fenitrothion and parathion reached 16X and 20X, respectively, less susceptible than the UTMB strain.

Despite the rather intensive use of organophosphorus (OP) insecticides for the control of various mosquito species by mosquito control districts along the Texas Gulf Coast, and the presence of organophosphate resistance in *Culex pipiens quinquefasciatus* in California (Georghiou et al. 1975) and tolerance in Louisiana (Steelman and Devitt 1976), there have been no reports of resistance in Texas since the finding 15 years ago that this and several other species were resistant to the BHC-dieldrin group (Micks et al 1961). Shortly thereafter the use of malathion as an adulticide was initiated, sometimes supplemented by certain other OP insecticides. In addition, chlorpyrifos has been used as a larvicide for approximately the past 7 years for the control of *Cx. quinquefasciatus*.

Because of the fact that *Aedes sollicitans* and *Ae. taeniorhynchus* have been the principal target species of control programs, and that malathion-resistance was reported in *Ae. taeniorhynchus* in Florida after approximately 10 years of use (Gahan et al 1966), much of the insecticide susceptibility monitoring was concentrated on these *Aedes* species. However, the occurrence of outbreaks of St. Louis encephalitis in Harris County in 1975 and 1976 stimulated us to make a thorough investigation of the insecticide susceptibility status of *Cx. quinquefasciatus* in 2 adjacent coastal counties, i.e., Brazoria and Galveston, during 1976.

MATERIALS AND METHODS. During the last half of 1976, a total of 20 batches of larvae were collected from various areas of Brazoria and Galveston Counties. Each was given a specific strain designation and reared through at least one cycle in the laboratory in order to have sufficient material for testing. Insecticide susceptibility determinations were done using the standard larval and adult test procedures of the World Health Organization. All larval susceptibility tests were conducted with F_1 larvae. Initial tests with larvae of several strains included a battery of insecticides: Abate®, bromophos, chlorpyrifos, fenthion, fenitrothion, malathion and parathion. Thereafter, the larvae were tested against chlorpyrifos and malathion only since those are the only insecticides used for mosquito control in these counties. Two replicates of 25 early 4th instar larvae each were used for each insecticide concentration (chlorpyrifos—0.0002, 0.001, 0.0025, 0.005, 0.0125 and 0.025 ppm; malathion—0.025, 0.062, 0.125, 0.312, 0.625 and 3.125 ppm). Tests with adults utilized from 3 to 5 replicates with 25 adult females per tube. Only malathion test papers (0.5, 3.2 and 5.0%) were used. Although all of the data reported herein are based on standard 1 hr exposures, some of the most tolerant strains were also exposed for 2 hr.

The test methods described above were also employed with the UTMB strain of *Cx. quinquefasciatus* which has been reared continuously in this laboratory for more than 20 years and is used by many investigators as a standard insecticide-susceptible strain. A number of tests of this strain were set up in parallel with field-collected strains from time to time, using both larvae and adults. Such comparisons were most useful in determining levels of insecticide susceptibility.

RESULTS AND DISCUSSION. The most interesting finding of this study was the marked variation in susceptibility to a particular insecticide between strains of *Cx. quinquefasciatus* larvae and adults collected from different localities within a single

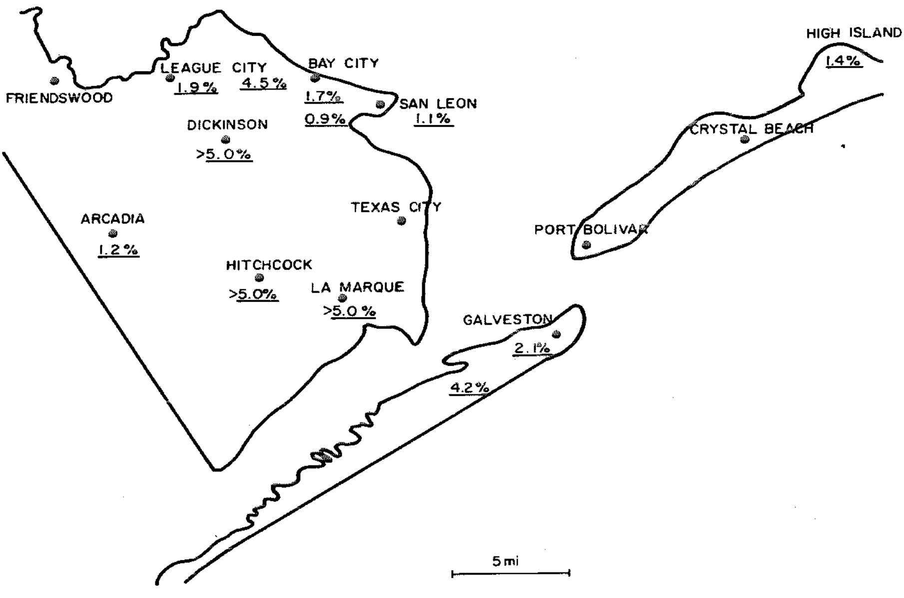

Fig. 1. Source and malathion susceptibility (LD_{50}) of twelve strains of *Cx. quinquefasciatus* from Galveston County, Texas in 1976 tested as adults.

county, and even within areas only several miles apart. The sources and corresponding LD_{50}'s for twelve Galveston County strains tested as adults against malathion are shown in Figure 1. The widest range in LD_{50}'s (0.9 to > 5.0%) was found in collections from the mainland, the largest of the 3 county land masses. The highly malathion-tolerant strains were largely from the central portion on the mainland, and the most susceptible collections were from the peripheral population centers. Doubling the exposure time of the strains with LD_{50}'s of greater than 5% malathion did not materially increase the mortality over that obtained in 1-hr exposures. The LD_{50} of the standard susceptible strain (UTMB) was 1.0% malathion.

Similar results were obtained with susceptibility tests of adults reared from larvae collected from 8 locations in Brazoria County, the LD_{50}'s ranging from 1.3 to > 5.0% malathion (Figure 2). Half of these strains were highly tolerant (> 5.0%). The most susceptible ones were collected almost entirely from the northeast corner of

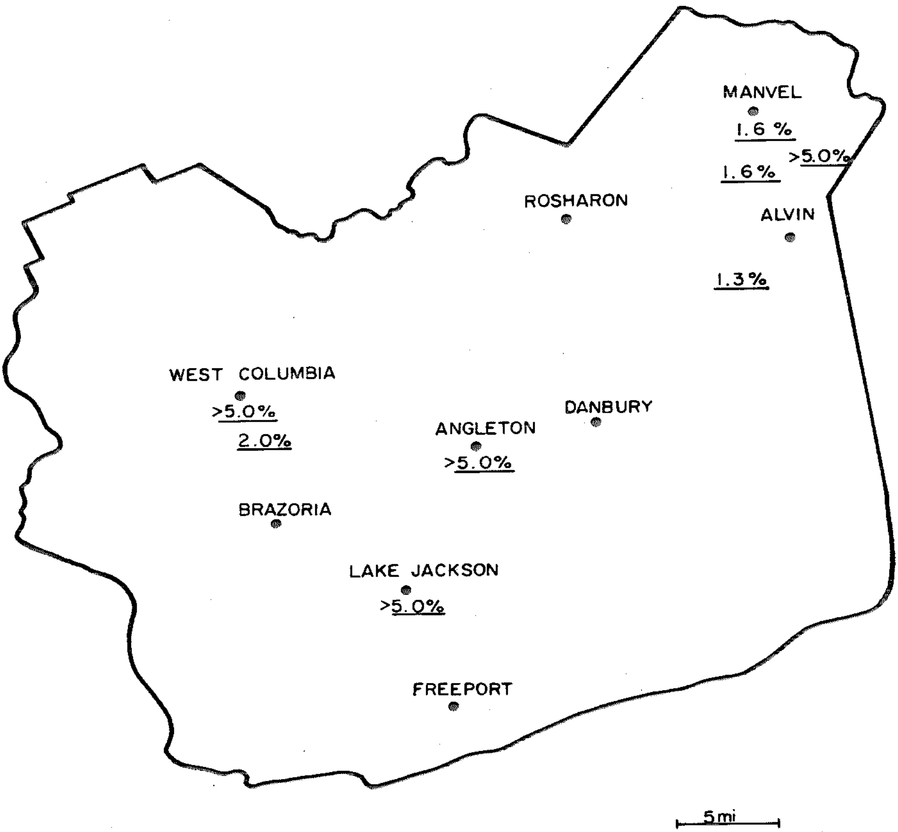

Fig. 2. Source and malathion susceptibility (LD_{50}) of eight strains of *Cx. quinquefasciatus* from Brazoria County, Texas in 1976 tested as adults.

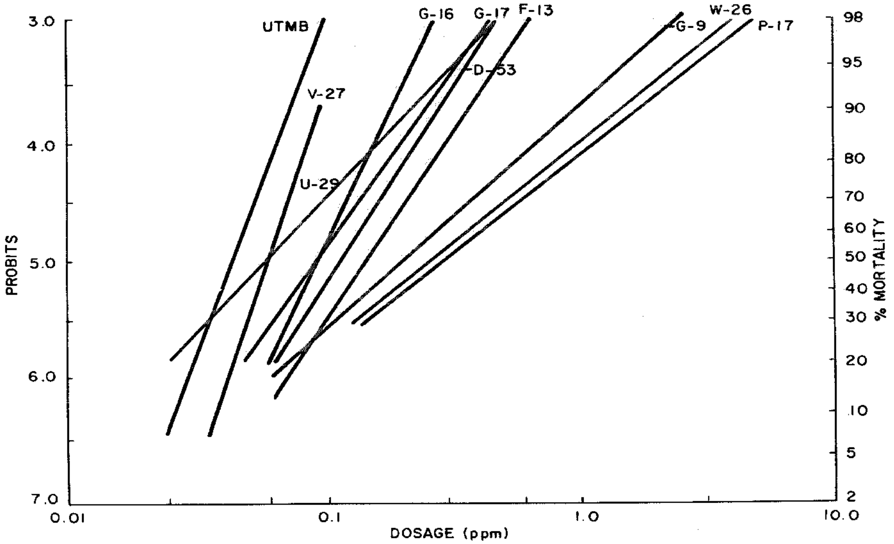

Fig. 3. Dosage-mortality regression lines comparing the malathion susceptibility of the UTMB (susceptible) strain of *Cx. quinquefasciatus* larvae with nine strains of this species collected from Galveston County in 1976.

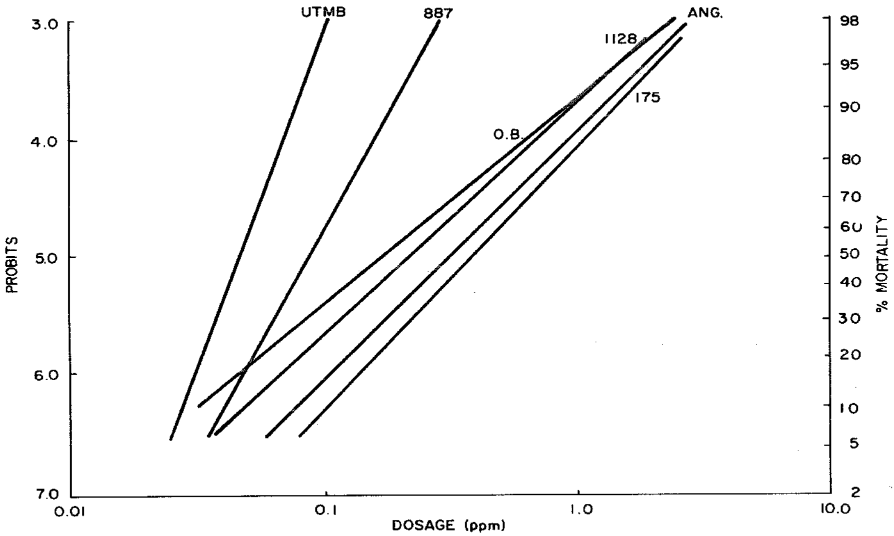

Fig. 4. Dosage-mortality regression lines comparing the malathion susceptibility of the UTMB (susceptible) strain of *Cx. quinquefasciatus* larvae with five strains of this species collected from Brazoria County in 1976.

the county where less malathion had been used by the mosquito control district.

Nine collections of larvae from Galveston County exhibited malathion tolerance. The LC_{50}'s ranged from 0.058 to 0.280 ppm with a maximum of 7X less susceptibility than in UTMB larvae (Fig. 3). For the most part, the strains of larvae with the greatest tolerance were also those showing the least susceptibility to malathion as adults. However, there were several exceptions. The 5 strains of larvae from Brazoria County ranged from 2X to 9X less susceptible than the UTMB strain (Fig. 4). Those strains with the lowest larval susceptibility were also the least susceptible to malathion as adults.

Chlorpyrifos susceptibility of *Cx. quinquefasciatus* larvae in both counties generally reflected the length and intensiveness of use of this insecticide in the control of larvae in septic ditches. Tests with the 8 strains from Galveston County, where chlorpyrifos had been used for approximately 5 years, showed insecticide tolerance in all strains; the range was 2X to 5X greater than the UTMB strain (Fig. 5). Larval collections from Brazoria County, following 7 years of use of chlorpyrifos, ranged as high as 13X less susceptible than the standard susceptible strain but exhibited substantial differences between strains in LC_{50}'s (Fig. 6). The 2 populations with the highest tolerance to this insecticide were also the least susceptible to malathion.

The complete spectrum of susceptibility to all insecticides included in the WHO larval test kit was obtained with 2 of the more malathion-tolerant strains from Galveston (G-9) and Brazoria (175) Counties (Table 1). Both of these field-collected strains showed substantial levels of tolerance to the only 2 insecticides used in the county mosquito control programs, i.e., chlorpyrifos and malathion. The collections from the 175 and G-9 areas were 11X and 8X, respectively, less malathion-susceptible, and 9X and 7X less susceptible to chlorpyrifos than the UTMB strain.

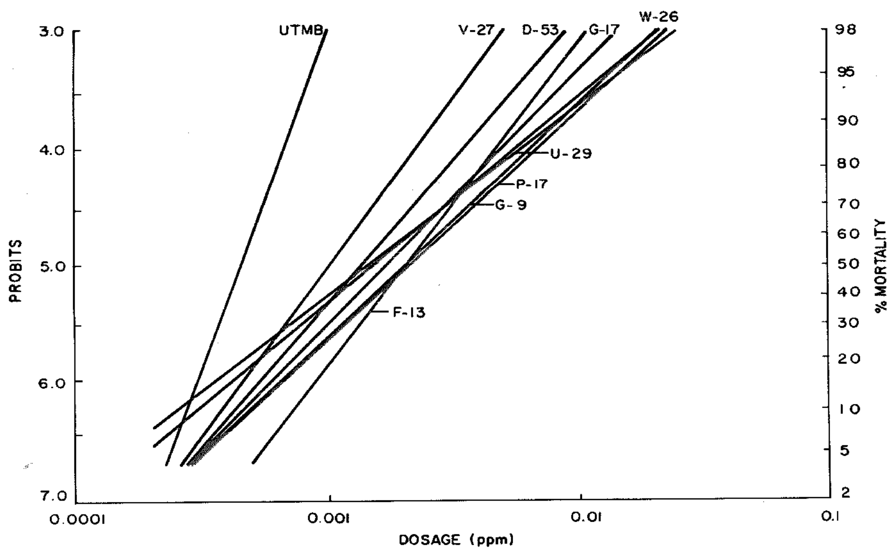

Fig. 5. Dosage-mortality regression lines comparing the chlorpyrifos susceptibility of the UTMB (susceptible) strain of *Cx. quinquefasciatus* larvae with eight strains of this species collected from Galveston County in 1976.

Table 1. Insecticide susceptibility of field-collected *Cx. quinquefasciatus* from Brazoria (175) and Galveston (G-9) Counties in 1976 as compared with a standard susceptible laboratory strain (UTMB)

Insecticide	UTMB Strain	175 Strain		G-9 Strain	
	LC_{50} (ppm)	LC_{50} (ppm)	Tol.*	LC_{50} (ppm)	Tol.*
Malathion	0.045	0.480	11	0.380	8
Chlorpyrifos	0.0004	0.0035	9	0.0028	7
Bromophos	0.0036	0.0260	7	0.0125	3
Fenthion	0.005	0.045	> 9	0.0120	2
Fenitrothion	0.011	0.170	16	0.0210	2
Abate	0.005	0.022	> 4	0.0165	3
Parathion	0.001	0.011	>11	0.0250	20

* Tolerance=LC_{50} field-collected strain÷LC_{50} UTMB Strain.

The 175 strain exhibited a surprisingly high level of tolerance to the remaining 5 insecticides whereas the LC_{50}'s for the G-9 strain were substantially lower with one exception, i.e., parathion. These findings reflect the indirect exposure of *Cx. quinquefasciatus* to agricultural chemicals. Furthermore, the differential between counties is very likely explained by the fact that Brazoria County is much more agricultural and is one of the major rice-growing areas in Texas. The high levels of fenitrothion-tolerance (16X) in the 175 strain, and parathion-tolerance (20X) in the G-9 strain were completely unexpected.

Perhaps the central point to emerge from these studies is that one cannot rely

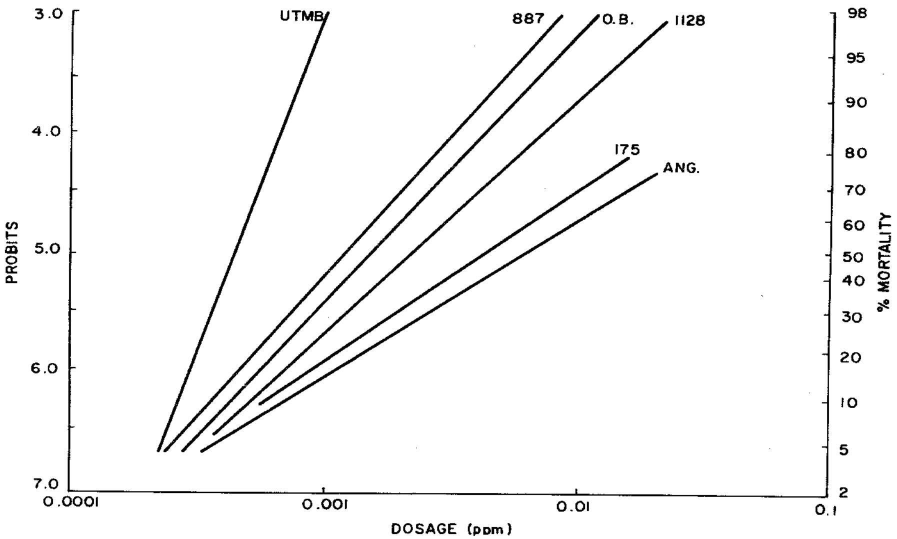

Fig. 6. Dosage-mortality regression lines comparing the chlorpyrifos susceptibility of the UTMB (susceptible) strain of *Cx. quinquefasciatus* larvae with five strains of this species collected from Brazoria County in 1976.

upon insecticide susceptibility test data from one or even several collections of *Cx. quinquefasciatus* to be accurately representative of an area the size of a county, or even smaller. Although insecticide tolerance represents a warning signal to the mosquito control program and assists the field worker in more effectively evaluating control procedures, inadequate sampling and testing may lead either to a false sense of security or unnecessary alarm. All the more dangerous are attempts to extrapolate the known insecticide susceptibility status of the species in one part of a state to another part without testing.

In spite of the presence of chlorpyrifos and malathion tolerance in a number of strains, due to their use as a larvicide in ditches and as an adulticide, respectively, good control of *Cx. quinquefasciatus* in Brazoria and Galveston Counties is still being obtained at recommended dosages.

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ON THE BIONOMICS OF BROMELIAD-INHABITING MOSQUITOES. IV. EGG MORTALITY OF *WYEOMYIA VANDUZEEI* CAUSED BY RAINFALL[1]

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ABSTRACT. In southern Florida the immature stages of the mosquito *Wyeomyia vanduzeei* Dyar and Knab are found in water held in the leaf axils of bromeliads. Correlation of data resulting from weekly estimates of the numbers of eggs, larvae and pupae vs. rainfall data indicated possible loss of eggs, but not of larvae or pupae, in heavy rain. This was confirmed by direct experimentation. Heavy rainfall appears to be an important mortality factor for the eggs of this mosquito.

INTRODUCTION. We have demonstrated (Frank et al. 1976) that presence of water in the leaf axils of tank bromeliads is a determinant of oviposition by *Wyeomyia vanduzeei*. Because the presence of water in leaf axils is necessarily determined by the quantity of recent rainfall, it would be logical to examine the relationship between, on the one hand, rainfall quantity and, on the other, change in number of eggs of *Wy. vanduzeei*. We wished to examine this relationship in the natural population in our study area (Frank et al. 1976),

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