EFFECTS OF DENGUE-1 INFECTION IN AEDES ALBOPICTUS ON ITS SUSCEPTIBILITY TO MALATHION

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Whether the presence of a viral infection in a vector may vary the susceptibility of the invertebrate host to a pesticide is not known. However, the presence of a virus, such as dengue, in a mosquito may alter the mosquito's susceptibility to an insecticide, such as malathion. If this were to occur, the traditional use of ultra low volume (ULV) malathion adulticide during a dengue outbreak could alter the proportions of infected and uninfected mosquitoes in the surviving population.

Mosquitoes used in the study were 3-day-old female Aedes albopictus (Skuse) from a laboratory colony in the F_6 to F_8 generation that originated from specimens obtained in Southern Espirito Santo, Brazil in 1986. The virus used was a dengue-1 isolate from human serum from Puerto Rico (PR-1679 isolated by Duane Gubler, San Juan Laboratories).

The isolate had been cultured in Aedes albopictus cells (C6/36), and the harvested tissue culture fluid (with 20% fetal calf serum added) was stored at -65° C until needed. Experimental mosquitoes were injected with about 0.25 ml of a dengue-1 suspension containing $10^{3.2}/0.10$ mL plaque-forming units, using the method of Rosen and Gubler (1974). One group of control mosquitoes was given a sham injection of fetal calf serum and another was left uninjected.

Infected and control mosquitoes were placed in 0.473-liter paper cartons (25 to 30 per carton) fitted with mesh at both ends and given access to a 10% sucrose solution. The cartons were housed in screened enclosures and maintained at 80% relative humidity (RH) and 26.7°C for 8 days.

The infection rate in the virus injected group of mosquitoes was estimated on the eighth day by examining head squashes for viral antigen using the direct fluorescent antibody test (Kuberski and Rosen 1977). Malathion susceptibility of infected and noninfected mosquitoes was estimated using aerosol exposures in a windtunnel after the method of Mount et al. (1976). Three replicates of 25 to 30 insects were each exposed to 0.1 mL of a range of concentrations of malathion dissolved in acetone in the pesticide wind tunnel (Model D2), supplied by the American Biological Supply Company, Baltimore, MD.³ The mosquitoes were then transferred from the mesh cartons to clean petri dishes fitted with filter paper and moistened with 10% sucrose solution. Twenty-four hours later, the mortality was determined, and the data probit was analyzed (Finney 1968) using a computer program by Daum (1969). The mean mortalities were plotted along with the computer estimated lines for the concentrations of the malathion. These tests were repeated three times.

Mortality of 5 to 30%, due to the trauma of the intrathoracic injection occurred during a 24 hour period following injection in both the dengue- and sham-inoculated mosquitoes. Thereafter, survival in the mosquitoes remained steady. The mortality in the sham-inoculated controls was similar to that in the noninjected controls. The infection rate in the dengue-inoculated mosquitoes was 87%. This was different from the variable infection rate obtained in orally infected insects (30% to 52%) when the insects were fed on infected blood through a membrane (B. Miller, personal communication, 1987).

Mortality. No significant difference was observed between the dengue-infected and the control groups of mosquitoes in their response to malathion (Fig. 1). With LC_{50} s of 0.17 (shaminoculated group) and 0.19 (dengue-inoculated group), there was a large degree of overlap of the fiducial limits. Since both the dengue-infected and uninfected mosquitoes responded equally to malathion, it would appear unlikely that use of this insecticide during a dengue outbreak would result in selection in favor of either group.

There have been a few publications on the effects of insecticides or growth regulators on the susceptibility of mosquitoes to microrganisms (Prasittisuk and Curtis 1982, Spencer and Olson 1982, Rasnitsyn and Zharova 1985), but we have been unable to find data on possible changes in susceptibility to insecticides as a

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³ Use of trade names or commercial sources is for identification only and does not constitute endorsement by the Public Health Service or by the U.S. Department of Health and Human Services.



Fig. 1. Computer-generated mortality/malathionconcentration lines and observed points for dengueinfected and control *Aedes albopictus*.

result of infection with a microorganism. In this respect, our findings may well be the first.

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REFERENCES CITED

- Daum, R. J. 1969. A revision of two computer programs for probit analysis. Bull. Entomol. Soc. Am. 16:10-15.
- Finney, D. J. 1968. Probit analysis. A statistical treatment of the sigmoid curve. Cambridge Univ. Press. 325 pp.
- Kuberski, T. T. and L. Rosen. 1977. A simple technique for the detection of dengue antigen in mosquitoes by immunofluorescence. Am. J. Trop. Med. Hyg. 26:533-537.
- Mount, G. A., N. W. Pierce and K. F. Baldwin. 1976. A new wind-tunnel system for testing insecticidal aerosols against mosquitoes and flies. Mosq. News 36:127-131.
- Prasittisuk, C. and C. F. Curtis. 1982. Absence of effects of insecticides on susceptibility of anophelines to *Plasmodium yoeli nigeriensis*. Southeast Asian J. Trop. Med. Public Health 13:127-132.
- Rasnitsyn, S. P. and A. N. Zharova. 1985. Effect of contact of mosquitoes with DDT. (In Russian). Parazitologiya (Leningrad) 19:287-289.
- Rosen, L. and D. Gubler. 1974. The use of mosquitoes to detect and propagate dengue viruses. Am. J. Trop. Med. Hyg. 23:1153–1160.
- Spencer, J. P. and J. K. Olson. 1982. Evaluation of the combined effects of methoprene and the protozoan parasite Ascogregarina culicis on Aedes mosquitoes. Mosq. News 42:384–390.