

## LABORATORY STUDIES OF RESIDUAL TOXIC EFFECTS OF MALATHION GRANULES ON *CULEX PIPIENS* *QUINQUEFASCIATUS* SAY LARVAE<sup>1</sup>

LALLAN RAI AND LAWRENCE L. LEWALLEN

Consolidated Mosquito Abatement District, Selma, California

and

California State Department of Public Health, Fresno, California

The results reported by Gjullin and Lewallen (1958) suggest a long term residual effect of malathion granules on mosquito larvae when the granules are applied at a dosage much larger than is usually used. This situation was encountered in a pasture in the Fresno Mosquito Abatement District, Fresno, California. The usual spray application of 0.5 pound of malathion to an acre began to prove ineffective against *Culex tarsalis* Coq. larvae in the pasture in the summer of 1956. The pasture was heavily treated with malathion pellets in August after which spray applications of malathion at 0.5 lb./acre were found to be effective and remained so for the rest of the mosquito season. Water from this pasture was also toxic to *Aedes nigromaculis* (Ludlow) larvae in September of that year.

Samples of water from the pasture which were taken in June of the following year also were toxic to *C. p. quinquefasciatus* larvae in laboratory tests. The results of several kinds of experiments on this water led the authors to the conclusion that the toxic effect was due to some kind of chemical rather than to a disease agent.

The present paper presents the results of further tests on water from this pasture. The hypothesis that the reported toxic effect was due to the malathion granules applied is also examined in the light of experiments on the residual action of this compound.

**MATERIALS AND METHODS.** The mosquitoes used in these tests were 4th instar

*Culex pipiens quinquefasciatus* Say larvae which are designated the Bakersfield susceptible strain. They were originally collected in Kern County in 1952 and were colonized at the Kern Mosquito Abatement District, Bakersfield, California. They have been under cultivation at the Fresno Field Station of the Bureau of Vector Control since 1955.

Two techniques were used for exposing larvae to test solutions, the paper cup technique and the residual technique. In the paper cup method, 25 larvae were placed into 100 ml. of test solution in 4 oz. paper cups.<sup>2</sup> Equal numbers of "control" larvae were placed in an equal volume of tap water. Exposure was for 24 hours after which mortality was judged by lack of movement of the larvae. These and all other tests reported here were done at 23° to 28° C.

The residual technique was used to determine how long insecticidal granules were toxic to mosquito larvae. The tests were carried out with the granules exposed to mud to simulate conditions in the field. A circular enamel pan 35 cm. in diameter and 12.5 cm. deep was used for this purpose. The pan was filled with mud from the suspected breeding place to a depth of 5 cm. The mud was then covered with water from the same breeding place to a depth of about 5 cm. The weighed amount of granules to be used was sprinkled evenly over the water surface with the aid of forceps. The larvae to be exposed were placed in wire baskets (tea strainers) which were then lowered into the test medium. The baskets were

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<sup>2</sup>Dixie Cup Company, Anaheim, California.

supported from sticks placed across the top of the pan. Twenty-five larvae were placed in each of three baskets for a single day's test. There were, therefore, three replicates in each day's test. The larvae were exposed for 24 hours after which time the baskets were removed for examination. The baskets in these tests were used to recover the larvae which were difficult to see against the dark background provided by the mud.

After a test pan was set up as described, the medium was tested daily (usually) until it was no longer toxic to mosquito larvae. The pan was then set up with a different dosage of granules and the test run again. Three dosages in all were used. Controls were run simultaneously in 800 ml. beakers using similar mixtures of mud and water but without insecticides. The pH of the water in these tests ranged from 7.1 to 7.5.

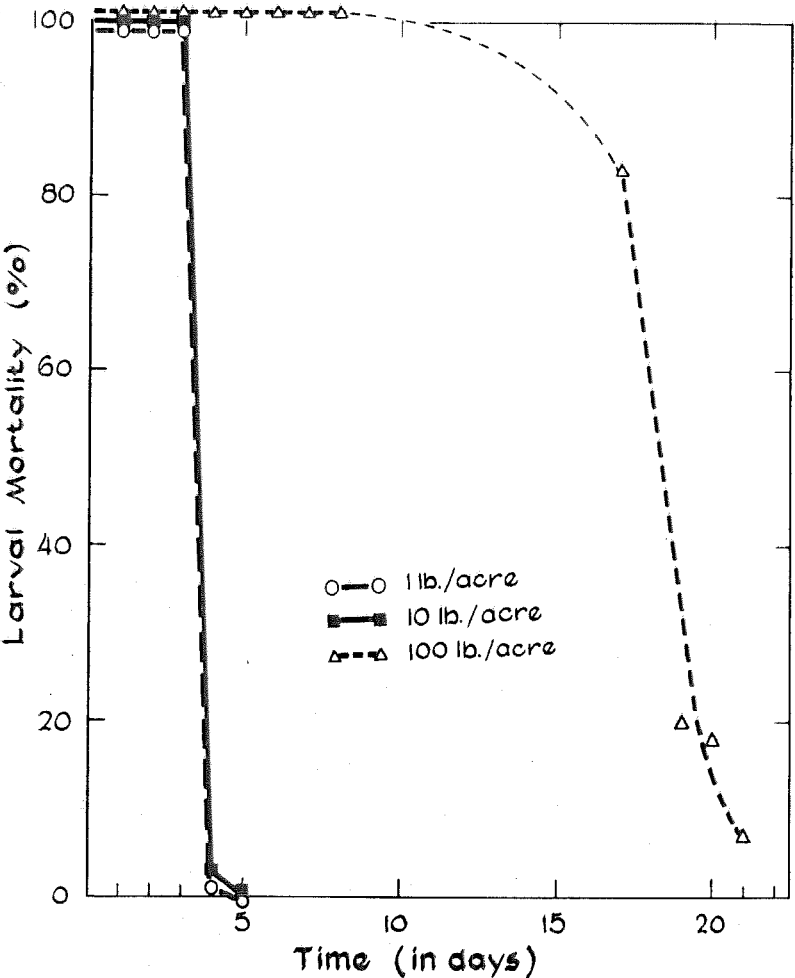


FIG. 1.—Residual effects of malathion granules applied at different dosage levels to 4th instar larvae of *Culex pipiens quinquefasciatus* Say.

Mud samples were tested for toxicity by adding 350 gm. to 400 ml. of pasture or tap water in an 800 ml. beaker, mixing, and allowing to settle. Larvae were exposed in wire baskets as above.

The granules used in these tests were Attaclay—AALUM, 24/48 mesh containing 10 percent by weight of malathion.<sup>3</sup> All dosages mentioned are in terms of weight of toxicant used, not weight of granules.

**RESULTS.** Collections of water and mud from the pasture studied by Gjullin and Lewallen were made on July 9, 1959 to determine whether they still had a toxic effect on mosquito larvae. Four samples of water were tested by the paper cup technique with two control samples in tap water. No larvae of the experimental or control series died.

Two samples of mud were tested, one combined with pasture water and the other with tap water. All of the larvae exposed in these tests also lived.

The results of tests on the residual effectiveness of malathion granules are shown in Figure 1. Malathion applied at 1 or 10 lbs./acre killed larvae for three days after which its effectiveness disappeared almost completely. When malathion at 100 lbs./acre was used, all larvae exposed during the first week died. The mortality rate dropped to 83 percent by

the 17th day after the application, and to 20 percent by the 19th day. Practically all larvae survived which were exposed three weeks after the application.

**DISCUSSION.** The above tests indicate that the presumed residual effect noted by Gjullin and Lewallen in 1956 and 1957 was no longer present in 1959. These authors described what was apparently a residual action of malathion granules lasting for several months in the field. There is some doubt, however, as to the type and quantity of granules used. It seems likely that the dosage of malathion used by these authors did not exceed 100 lbs./acre as was used in the present tests. It also seems likely that the type of granules used by these authors did not differ markedly from the type used in the present tests. It, therefore, is unlikely that the toxic effects noted by Gjullin and Lewallen in 1957 was due to the granules applied in 1956.

**SUMMARY.** The residual toxicity of malathion granules to *C. pipiens quinquefasciatus* Say larvae in a mud-water complex was studied under laboratory conditions. Malathion applied at 1 and 10 lbs./acre was effective over a three-day period; at 100 lbs./acre it was toxic for more than 17 days.

#### Literature Cited

GJULLIN, C. M., and LEWALLEN, LAWRENCE L. 1958. Report on cooperative mosquito research in California. Calif. Mosquito Control Assn., Proc. and Pap. Ann. Conf. 26:28-31.

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