

all determinations (the bubble is repeatedly displaced to point X in the capillary tube).

Because the air bubble has to be expelled completely from the capillary tube, it is important that the tip of the capillary tube be made as illustrated in Fig. 1a or 1b and not as commonly constructed in conventional potometers (see Fig. 1c); in the latter case the capillary tube is first sealed off, and a lateral aperture is blown out.

The air bubble need not be completely expelled from the capillary tube before each determination; the original bubble can be displaced to the starting point several times by merely tightening a screw clamp which can be attached to the rubber teat.

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References and Notes

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Effect of DDT on Free Amino Acids of Susceptible and DDT-Resistant *Aedes aegypti* Larvae

Abstract. The exposure of mosquito larvae (*Aedes aegypti* L.) to LC_{50} concentrations of DDT for various periods of time resulted in a selective increase in the alanine level of a DDT-resistant strain after 4 to 8 hours' contact with the insecticide, whereas the concentration of this amino acid in a susceptible strain remained comparatively low.

Evidence that DDT may disturb amino acid metabolism in insects has been presented by Reiff (1) who found that this insecticide reduced the amino acid level of the hemolymph of DDT-susceptible house flies, and by Corrigan and Kearns (2) who reported that DDT

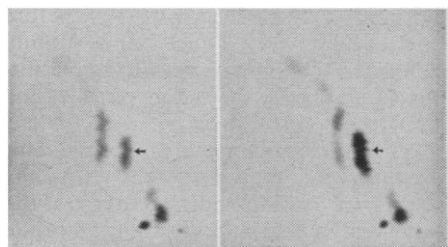


Fig. 1. Two-dimensional chromatograms of the DDT-treated Trinidad larvae (right) and the untreated controls (left) showing a marked increase in the alanine level (spot at arrow) upon 4 hours' exposure to DDT.

characteristically lowered the proline content of the hemolymph of *Periplaneta*. Previous investigations in our own laboratory have demonstrated that when larvae of a susceptible strain and a DDT-resistant strain of *Aedes aegypti* were exposed to DDT, the total free amino acid content of the resistant strain increased while that of the susceptible strain remained essentially the same (3). The present report is concerned with the continuation of this work to determine which amino acids are particularly affected by DDT.

Third and fourth-stage larvae of a susceptible strain (UTMB) and a DDT-resistant strain (Trinidad) were exposed to their respective LC_{50} concentrations of DDT (0.5 and 2.5 parts per million after 24 hours' exposure) for periods of 2, 4, 8, and 16 hours, respectively. In preparation for each experiment, approximately 1000 larvae of each strain were rinsed thoroughly in distilled water and divided equally into each of two enameled pans containing 400 ml of distilled water. The appropriate quantity of an acetone solution of DDT (p, p' isomer) was added to one pan prior to introduction of the larvae, the other pan serving as the control. At the end of the exposure period the larvae, all of which were still living, were removed from their respective containers by means of separate strainners. They were then rinsed in two changes of distilled water, weighed, and homogenized in acetone, with a tissue grinder. After centrifugation, measured amounts of the supernatants were applied to sheets of Whatman 3-mm filter paper and chromatographed by previously described techniques (4). Chromatograms of the test and control extracts were first compared visually for evidence of differences in concentration of amino acids. Densitometric measurements were then made of the various ninhydrin-positive spots appearing on two-dimensional chromatograms. The results presented here represent the combined data from five separate sets of tests conducted on different generations of larvae at each exposure period.

Virtually complete reproducibility was obtained between tests. The only striking difference in amino acid levels between the UTMB and Trinidad strains after exposure of the larvae to DDT was in the concentration of alanine, which was greater in the latter strain (Fig. 1). The maximum density values for this amino acid on chromatograms prepared after 2, 4, 8, and 16 hours of DDT exposure were 85.5, 93, 92.5, and 80, respectively, for the Trinidad strain, a peak occurring during the 4- to 8-hour period. The alanine level of the susceptible strain increased somewhat after 4 hours' exposure to DDT

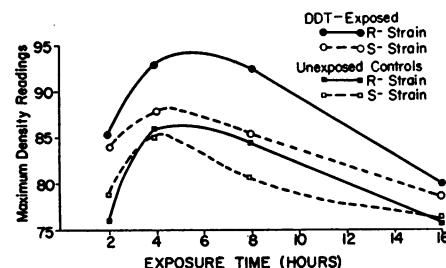


Fig. 2. Maximum density curves showing the alanine concentration in the susceptible (S) and DDT-resistant (R) strains of larvae and their corresponding controls after 2, 4, 8, and 16 hours' exposure to DDT.

and declined sharply thereafter, the corresponding density values being 84, 87.5, 85, and 78 (Fig. 2). Interestingly enough, the alanine content of the corresponding untreated and unfed controls also rose during the 2- to 4-hour period. Whereas the level of this amino acid was somewhat higher in the UTMB strain at 2 hours, the reverse was true after 8 hours. Strain differences in the concentration of several other amino acids were of extremely small magnitude, and were not consistent.

As yet, it is not known whether the increase in alanine content which follows DDT exposure is due to an increased rate of alanine synthesis or a decreased rate of metabolism. The similarity in shape of the density curves representing the DDT-treated larvae and the untreated controls suggests that the effects of DDT on alanine metabolism are of a quantitative nature. In any case, the DDT-resistant strain exhibits either greater alanine production or is less able to utilize it than is the normal strain. The finding that *Aedes aegypti* larvae are capable of synthesizing large quantities of alanine (5) is consistent with the possibility that the differences noted between the two strains used in this study may be a reflection of the comparative activity of transaminases or other enzymes.

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