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# Level of Lactate Dehydrogenase (LDH) in Resistant and Susceptible Strains of Culicine Mosquitoes of the Karachi Region after Treatment with DDT, Malathion and Cyfluthrin

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Research Article

# Level of Lactate Dehydrogenase (LDH) in Resistant and Susceptible Strains of Culicine Mosquitoes of the Karachi Region after Treatment with DDT, Malathion and Cyfluthrin

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**Abstract:** The effects of Malathion, Cyfluthrin, and DDT were observed on the activity of lactate dehydrogenase (LDH) in susceptible and resistant strains of culicine mosquitoes.

Biochemical estimation (colorimetry) revealed a remarkable increase in the enzyme activity in *Culex fatigans* (L.Y. strain). In contrast, in *Culex fatigans* (G.I. strain) and *Culex fatigans* (K.U strain), inhibition of enzyme activity was found after treatment with pesticides.

**Key Words:** Malathion, Cyfluthrin, DDT, LDH, Mosquito larvae

## DDT, Malathion ile Cyfluthrin Uygulamasından Sonra Dayanıklı ve Dayanıksız *Culex* Sivrisinek Soylarında Lactate Dehydrogenase (LDH) Düzeyleri

**Özet:** Malathion, Cyfluthrin ile DDT'nin, gerek dirençli gerekse duyarlı *Culex* sivrisinek soylarındaki laktat dehidrogenaz (LDH) aktivitesi üzerine etkileri gözlenmiştir.

Biyokimyasal hesaplamada (kolorimetri) *Culex fatigans*'ta (L.Y. soyu) enzim aktivitesinde kayda değer bir artma gözlenmemiştir. Buna karşın *Culex fatigans* G.I. ile *Culex fatigans* K.U. soylarında böcek zehiri uygulamasından sonra enzim aktivitesinin engellendiği görülmüştür.

**Anahtar Sözcükler:** Malathion, Cyfluthrin, DDT, LDH, Sivrisinek larvası

## Introduction

Mosquitoes have a great impact on human welfare because the females of approximately 300 species of mosquitoes are blood sucking, many of which bite man and serve as vectors of several significant human diseases. Mosquitoes are more harmful, in fact, than all species of rats and lice combined and their complete control is yet not in sight. Investigations on different aspects of organochlorine, organophosphate and pyrethroids have been carried out by various workers. Most of these studies deal with sclerotization, fecundity, toxicity, morphological abnormalities and enzyme levels (1-9). On enzyme estimation in the resistant, treated and non-treated insects, very little work has been carried out (10-13). In Pakistan, only, a few investigations have been reported on resistance in mosquitoes (14-18), and no systematic studies on the determination of the level of resistance in the culicine mosquito population in Karachi have been conducted. To fill this gap the present studies were undertaken with three insecticides (DDT (organochlorine), Cyfluthrin (Pyrethroid), and Malathion (Organophosphate)) used against *Culex fatigans* to determine the level of resistance by determining the enzyme level through the colorimetric method.

## Materials and Methods

For the rearing of all strains, the technique of Ashrafi et al. (19) was followed and in the experimints only late 3$^{rd}$ instar larvae were used. The three strains of *C. fatigans* reared were the K.U. strain, collected form campus, the L.Y. strain, collected from Lyari, and the G.I. strain, collected from Gulshan-e-Iqbal in Karachi. The rearing was caried out at the insectory of the Department of Zoology, University of Karachi. During rearing period the relative humidity and the temperature were

maintained at 65±5% and 28±3C respcetively. The late 3rd instar larvae of each strain were treated with LC50 concentrations of insecticide following the WHO standard method. After 24 h, larvae were homogenized. The homogenates were centrifiuged at 1500 rpm and then supernatants were separated and subjected to biochemical (colorimetry) tests. For the determination of lactate dehydrogenase (LDH) activity, Boehringer Mannheim Gmbh dignostic Kit no. 124885 was used, Readings were taken after exactly 1 minute at 340 nm.

**Results**

Biochemical estimation (colorimetry) after one minute incubation revealed that Malathion decreased LDH activity 30% in the K.U. strain and 58.33% in the G.I. strain, and increased activity 22.22% in the L.Y. Strain (Tables 1-3); Cyfluthrin decreased activity 33.33% in the K.U. strain and 50% in the G.I. strain, and increased activity 7.20% in the L.Y. strain (Tables 4-6); and DDT decreased activity 50% in the K.U. strain and 66.66% in the G.I. strain, and increased activity 41.66% in the L.Y. strain (Tables 7-9).

| Compound | Mean of enzyme unit (U/L) | S.D. | S.E. | 95% Confidence limit | % Inhibition |
|---|---|---|---|---|---|
| Control | 30 | 14.2 | 8.25 | 14.15 - 46.5 | 0.0 |
| Malathion | 21 | 13.74 | 7.93 | 5.44 - 36.55 | 30.0 |

Table 1. Effect of Malathion treatment (0.225 ppm for 24 h) on LDH activity in the larvae of the K.U. strain. Interval time (A) .....1 min.

| Compound | Mean of enzyme unit (U/L) | S.D. | S.E. | 95% Confidence limit | % Inhibition |
|---|---|---|---|---|---|
| Control | 36 | 24.37 | 14.0 | 9.07 - 64.25 | 0.0 |
| Malathion | 15 | 5.19 | 3.0 | 9.12 - 20.88 | 58.33 |

Table 2. Effect of Malathion treatment (0.44 ppm for 24 h) on LDH activity in the larvae of the G.I. strain. Interval time (A) .....1 min.

| Compound | Mean of enzyme unit (U/L) | S.D. | S.E. | 95% Confidence limit | % Inhibition |
|---|---|---|---|---|---|
| Control | 21 | 13.74 | 7.93 | 5.44 - 36.55 | 0.0 |
| Malathion | 27 | 9.0 | 5.19 | 16.81 - 37.18 | 22.22 |

Table 3. Effect of Malathion treatment (0.5 ppm for 24 h) on LDH activity in the larvae of the L.Y. strain. Interval time (A) .....1 min.

| Compound | Mean of enzyme unit (U/L) | S.D. | S.E. | 95% Confidence limit | % Inhibition |
|---|---|---|---|---|---|
| Control | 36 | 5.19 | 3.0 | 18.12 - 29.88 | 0.0 |
| Cyfluthrin | 24 | 18.5 | 10.68 | 15.39 - 57.27 | 33.33 |

Table 4. Effect of Cyfluthrin treatment (0.000165 ppm for 24 h) on LDH activity in the larvae of the K.U. strain. Interval time (A) .....1 min.

| Compound | Mean of enzyme unit (U/L) | S.D. | S.E. | 95% Confidence limit | % Inhibition |
|---|---|---|---|---|---|
| Control | 42 | 14.29 | 8.25 | 26.49 - 58.84 | 0.0 |
| Cyfluthrin | 21 | 10.30 | 6.0 | 9.24 - 32.76 | 50.0 |

Table 5. Effect of Cyfluthrin treatment (0.00018 ppm for 24 h) on LDH activity in the larvae of the G.I. strain. Interval time (A) .....1 min.

| Compound | Mean of enzyme unit (U/L) | S.D. | S.E. | 95% Confidence limit | % Inhibition |
|---|---|---|---|---|---|
| Control | 38 | 19.21 | 11.09 | 16.91 - 60.41 | 0.0 |
| Cyfluthrin | 41 | 25.40 | 14.66 | 12.92 - 70.41 | 7.20 |

Table 6. Effect of Cyfluthrin treatment (0.00045 ppm for 24 h) on LDH activity in the larvae of the L.Y. strain. Interval time (A) .....1 min.

Table 7. Effect of DDT treatment (0.175 ppm for 24 h) on LDH activity in the larvae of the K.U. strain. Interval time (A) .....1 min.

| Compound | Mean of enzyme unit (U/L) | S.D. | S.E. | 95% Confidence limit | % Inhibition |
|---|---|---|---|---|---|
| Control | 30 | 21.36 | 12.33 | 6.16 - 54.5 | 0.0 |
| DDT | 15 | 5.19 | 3.0 | 9.12 - 20.88 | 50.0 |

Table 8. Effect of DDT treatment (0.325 ppm for 24 h) on LDH activity in the larvae of the G.I. strain. Interval time (A) .....1 min.

| Compound | Mean of enzyme unit (U/L) | S.D. | S.E. | 95% Confidence limit | % Inhibition |
|---|---|---|---|---|---|
| Control | 36 | 18.5 | 10.68 | 15.39 - 57.27 | 0.0 |
| DDT | 12 | 5.19 | 3.0 | 6.12 - 17.88 | 66.66 |

Table 9. Effect of DDT treatment (0.425 ppm for 24 h) on LDH activity in the larvae of the L.Y. strain. Interval time (A) .....1 min.

| Compound | Mean of enzyme unit (U/L) | S.D. | S.E. | 95% Confidence limit | % Inhibition |
|---|---|---|---|---|---|
| Control | 21 | 7.93 | 13.74 | 5.44 - 36.55 | 0.0 |
| DDT | 36 | 18.5 | 10.68 | 15.39 - 57.27 | 41.55 |

## Discussion

Insecticides generally affect the enzymatic activities of insects. In the present investigation the effects of organochlorine (DDT), organophosphate (Malathion) and pyrethroids (Cyfluthrin) were observed on the lactate dehydrogenase (LDH) activity in the larvae of *C. fatigans.* Some workers in the past also studied these effects; for instance, Ashrafi et al. (20) found that the acetone (check) inhibits the activity of phosphatase about 30% and this inhibiton was restored in Malathion, Parathion and Petkolin (OC) (in acetone solvent) treated insects. This confirms the LDH inhibiton by DDT and Malathion in the present suspecptible strain. In the case of the L.Y. strain, which is resistant to some extent, there is an increase in LDH activity.

Hayaoka and Dauterman (21) reported that chlorinated hydrocarbons were more effective inducers of glutathione S-tansferase activity than the other pesticides evaluated. Likewise, Verma and Rahman (22) reported the effect of certain insecticides on transaminases in *C. fatigans* whereas Meany and Pocker (23) observed inactivation of LDH by organochlorine. Scott and Matsumura (24) reported that cypermethrin and deltamethrin did not cause apparent nerve excitation but had toxic effects. Change and Jordan (25) reported reduced esterase activity by synthetic pyrethroid treatment. Azmi et al. (17-18) reported the activity of Phosphomonoesterases in susceptible and resistant strains of *C. fatigans*. Saleem and Shakoori (26), reporting resistance in *Tribolium castaneum* (Pak strain) against Malathion, reported the absence of cross resistance in this strain against permethrin and cypermethrin. In the CTC -12 strain they found resistance against Pyethroid. They reported increased carboxylesterase, cholinesterase, protease, and lactate dehydrogenase activities, but reduced acid phosphatase activity. In the present findings LDH activity was higher in the treaated insects than in those in the control batch. Thus the present findings confirm their results. The differences may be due either to strain difference or to some unknown factors.

Form the above discussion, it is probable that the levels of some enzymes increase in resistant strains, as reported by a number of the above werkers. It is hoped that the present study will contribute to a better understanding of the resistance of mosquitoes of Karachi region and would help prevent indiscriminate use of such pesticides.

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