Branchial and plasma $\text{Na}^+$, $\text{K}^+$ - ATPase activity in a freshwater teleost *Cyprinus carpio* var. *communis* under methomyl toxicity

S. KRISHNAVENI, A. CHEZHIAN*, M. RAMESH AND R. MANAVALARAMANUJAM

Department of Zoology, Bharathiar University, Coimbatore — 641 046, India

ABSTRACT

Methomyl, a carbamate pesticide, proved to be toxic to *Cyprinus carpio* var. *communis* even at sublethal level (2.014 ppm). $\text{Na}^+$, $\text{K}^+$ - ATPase activity from both gills and plasma showed significant reduction throughout the experimental period. The per cent change in the enzyme activity was more drastic in the case of plasma. The results are discussed in relation to the significance of the above enzymes as non-specific biomarkers against environmental stress.

The carbamate insecticides are widely used as a replacement for its more persistent organochlorine counterparts. This chemical is highly valued for its rapid insecticidal action and its relatively short environmental persistence (Kuhr and Dorough, 1976). Pesticides used in pest control programmes seem to produce many physiological and biochemical changes in freshwater organisms by influencing the activities of several enzymes. In recent years, many investigators have studied the effect of pesticides on the enzyme activities in fish. Enzymes such as acid and alkaline phosphatases have been examined (Bulusu and Chakravarthy, 1987) with reference to the influence of pesticides on their activities.

$\text{Na}^+$, $\text{K}^+$ - ATPase is an enzyme responsible for the transport and maintenance of $\text{Na}^+$ and $\text{K}^+$ gradients across the membranes and resting membrane potential for neuronal excitability (Siegel et al, 1981). $\text{Na}^+$, $\text{K}^+$ - ATPase has been reviewed and assessed as a potentially useful indicator of pollution stress in aquatic animals. Hay and Waiwood (1983) reported that database on $\text{Na}^+$, $\text{K}^+$ - ATPase activity of aquatic fauna is inadequate and more research should be done so that they can be used for hazard assessment of pollutants. Hence, an attempt was made to study the toxicity of methomyl, a carbamate pesticide on $\text{Na}^+$, $\text{K}^+$ - ATPase of a freshwater fish *Cyprinus carpio* var. *communis*.

Specimens of *Cyprinus carpio* var. *communis* were collected from the fish farm of Tamil Nadu Fisheries Develop-
ment Corporation Limited, Aliyar and acclimatised to the laboratory conditions for fifteen days. Water was changed daily and fish were fed *ad libitum* with rice bran and groundnut oil cake twice a day. The physicochemical parameters of the water was estimated (APHA, 1976) as: dissolved oxygen: 6.25 ± 0.02 mg/l, pH: 7.2 ± 0.2, temperature: 25.0 ± 2.0°C, salinity: 0.2 ± 0.07 ppm, total hardness: 13 ± 2.0 mg/l, calcium: 5.0 ± 1.0 mg/l, magnesium: 8.0 ± 2.0 mg/l and total alkalinity: 20.0 ± 10.0 mg/l.

The median tolerance limit of methomyl to fish was found out following probit analysis method (Finney, 1978) and it was 20.14 ppm. For sublethal study, a large glass tank of 60 l capacity with 50 l of water and 50 fish were taken and 1/10 of the LC₅₀ concentration of methomyl (2.014 ppm) was added. A common control was maintained. Fish were fed with *ad libitum*. Water was changed daily and toxicant was renewed. At the end of every seventh day, fish from control and experimental tanks were taken and blood was drawn from the heart region by cardiac puncture with heparin as an anticoagulant and centrifuged at 10,000 rpm for 20 min and supernatant was collected. Then 100 mg of gill were taken and homogenised with 2.5 ml of 0.25M sucrose solution in ice-cold condition and centrifuged for 10 min, at 6,000 rpm and clear supernatant was taken for enzyme assay. Both plasma and gill Na⁺, K⁺ were estimated following the method of Shiosaka *et al*. (1971). The significance between the sample mean of control and experimental fish was tested using students `t` test at 5 % level.

Table 1 shows the changes in Na⁺, K⁺ - ATPase activity in gills and plasma of *Cyprinus carpio var. communis* exposed to sublethal concentration of methomyl for 28 days. In gills, the enzyme activity decreased in the exposed fish showing a per cent decrease of 25.97 at the end of 7th day treatment. After the 7th day, enzyme activity recovered showing a per cent increase of 24.91 at the end of 14th day treatment. Again after 14th day, enzyme activity decreased showing a per cent decrease of 24.86 and 11.68 at the end of 21st and 28th days respectively.

In plasma, enzyme activity decreased throughout the experimental period showing a minimum per cent decrease of 7.69 at the end of 14th day. A maximum per cent decrease of 44.83 at the end of 28th day indicates that the changes in enzyme activity in the

<table>
<thead>
<tr>
<th>Organ/Plasma</th>
<th>Exposure period (in days)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>7</td>
</tr>
</tbody>
</table>

**Gills**
- Control: 42.48 ± 0.107, 31.720 ± 0.447, 31.720 ± 0.057, 39.040 ± 0.0286
- Experiment: 31.440* + 0.179, 39.620* ± 0.009, 26.840* ± 0.012, 34.040* ± 0.036
  - (-25.97), (+24.91), (-24.86), (-11.68)

**Plasma**
- Control: 0.029 ± 0.0005, 0.026 ± 0.0005, 0.026 ± 0.0005, 0.029 ± 0.0005
- Experiment: 0.026*+ ± 0.0005, 0.024* + 0.0005, 0.017* + 0.0005, 0.016* + 0.0005
  - (-10.35), (-7.69), (-34.62), (-44.83)

Values are means ± S.E of five individuals. (-) % decrease over control, (+) % increase over control.
* p<0.05.
pesticide treated fish was significant at 5% level.

The activity of Na\(^+\), K\(^+\) - ATPase in fish gill and possibly in the other animal tissues may be a useful non-specific biomarker as it is easily quantified and affected by a variety of toxicants (Mayer et al., 1992). Watson and Beamish (1980) reported that in vitro exposure of Na\(^+\), K\(^+\) - ATPase to a variety of organochloride compounds and metals inhibits this enzyme. ATPase systems were adversely affected in chronic in vivo studies with fathead minnows, *Pimephales promelas*, exposed to archolar 1242 (Kinter et al. 1972). Davis et al. (1972) reported a significant decrease of Na\(^+\), K\(^+\)-ATPase activity in the microsomal fraction of gill homogenate of rainbow trout exposed to aldrin, chlordane, DDD, DDE, DDT, dieldrin, heptachlor, lentane, methoxychlor, perthane, strobane, toxaphene 2, 4, 5-T and PCBs (Archolar 1254).

Solomonson et al. (1976) noted that changes in the membrane lipid content or physical properties of the membrane influenced Na\(^+\), K\(^+\) - ATPase activity. Rangaraj and Kalant (1981) are of the opinion that membrane disordering effect may be the reason for enzyme inhibition. The loss of ion-specific ATPase could be attributed to the the loss of sodium and potassium ions due to cellular leakage into the body fluids. Nonavailability of substrates like ATP molecules may also result in the inhibition of these ion - specific ATPases (Kunhert, 1976). In the present study, the significant increase in Na\(^+\), K\(^+\)-ATPase in gills and plasma may be due to changes in plasma Na\(^+\), K\(^+\) and chloride (Cl) levels. The inhibition of Na\(^+\), K\(^+\)-ATPase activity from gills and plasma of *Cyprinus carpio var. communis* during sublethal treatment may be due to a changes in the physical properties of the membrane or disruption of oxidative phosphorylation processes within the cells.

**Acknowledgment**

M. Ramesh thanks CSIR, New Delhi, for the award of a Research Associateship.

**References**


