Aminotransferases or transaminases are a group of enzymes that catalyse the process of biological transamination. Transamination allows an interplay between carbohydrate, fat and protein metabolism (Cohen and Sallach, 1961), providing a source of keto acids for Krebs' cycle and gluconeogenesis. Weber (1963) states that this mechanism provides more energy to meet the increased demand under stress. The aminotransferases found in the tissues are mainly aspartate aminotransferase (L-Aspartate: 2-oxoglutarate aminotransferase (AAT) - EC 2.6.1.1) or glutamic oxaloacetic transaminase (GOT) and Alanine aminotransferase (L - Alanine : 2 - oxoglutarate aminotransferase (ALAT) - EC 2.6.1.2) or glutamic pyruvic transaminase (GPT). Wilson (1973 a) explained that the primary enzymes concerned with the amino acid metabolism and gluconeogenesis are glutamic dehydrogenase, aspartate aminotransferase (AAT) and alanine aminotransferase (ALAT). The substrates for AAT (GOT) and ALAT (GPT) are aspartic acid and alanine which serve as the two major glucogenic amino acids which give rise to glucose precursors (Lehninger, 1979). Since the highest activity of aminotransferases in most cases was observed in heart, liver and kidney tissues, these organs play a major role in amino acid metabolism and gluconeogenesis. So it is interesting to study the effects of pollutants on the activity of these enzymes in these specific tissues. There are two approaches (1) to study the serum enzyme activity and (2) to study the specific activity of the enzymes within the tissue.

The study of serum enzyme activity got its initial impetus when Karmen (1955) showed a direct link between serum AAT (GOT) levels and acute transmural myocardial infarction and in man it was later extended to both positive and differential clinical diagnosis (Agress, 1959; Bang et al., 1959). In mice infected with hepatic virus, the increase in serum transaminases has been shown to be roughly proportional to the degree of cell necrosis (Friend et al., 1955). Later Wroblewski et al. (1956) extended their work on serum AAT or serum GOT (SGOT) and they found that the acute and chronic hepatic diseases are associated with elevation of SGOT activity which are sufficiently characteristic to permit diagnostic differentiation. Zelman et al. (1959) in
a similar study of human subjects, found an excellent relationship between the extent of necrosis of liver cells and the rise in serum transaminases.

The rationale for using serum GOT and GPT or other serum enzymes was that elevated enzyme levels should simply reflect the amount of damage which had occurred in the enzyme rich tissue and this approach is used for the diagnosis of both the site and extent of organ injury (McKim et al., 1970; Schmidt and Schmidt, 1974; Racicot et al., 1975; Goel and Garg, 1980; Wotten and Williams, 1980).

In fishes, the use of transaminases (aminotransferases) in the diagnosis of tissue damage was investigated by Mollander et al. (1955), Wroblewski and La Due (1956), Wroblewski et al. (1956) and Bell (1968). Bell (1968) discussed the practical value of GOT estimation in serum of salmon for the detection (distinction) of apparently healthy fish from those treated with hepatic poison, bromobenzene or those affected by bacterial kidney disease. Apart from these earlier workers, serum GOT and GPT were frequently used by fish pathologists to diagnose sublethal insult or injury to liver by pollution (Mehrle and Bloomfield, 1974; Malevski et al., 1974; Racicot et al., 1975). In the field of environmental toxicology, serum and tissue analysis are becoming important for the detection of chemical pollutants. It is noted that toxic agents or factors which lead to chronic impairment of animal metabolism will cause changes in the activities of some enzymes (see Bell, 1968; McKim et al., 1970; Lockhart et al., 1972). Some of these responses are likely to be of a more general nature, indicating the organisms reaction to a situation of stress brought about by a general deterioration of water quality (Oikari and Soivio, 1977).

Eventhough it is suggested that a high level of GOT in the serum of a species is associated with high values in the liver, myocardium and kidney (Zimmerman et al., 1968), there are reports of finding no such association (Gaudet et al., 1975). Apart from these, studies are few dealing with the enzyme levels in specific tissues where the toxicants are accumulated or detoxified. Similarly, can we say that the serum activity is representative of different tissues? Is there any tissue differences in the activity of transaminases in response to toxicants? In pursuit of an answer to these
questions, the present study was conducted to examine the effects of copper and mercury on the activity of GOT and GPT in the liver and kidney of Oreochromis mossambicus.

MATERIAL AND METHODS

Collection of specimens, acclimatization and experimental setup were the same as described in Chapter 2. Preparation of the enzyme sample from the selected tissues was done as described in Chapter 3.

The activity of AAT (GOT) and ALAT (GPT) in the liver and kidney was determined following the method of Reitman and Frankel (1957) with slight modification. To 0.6 ml of 0.1 M frozen phosphate buffer (pH 7.5), 0.1 ml of enzyme preparation was added and again frozen until analysis. After the completion of the samplings, the frozen buffer-enzyme mixture was placed in a water bath at 37°C. When the mixture attained 37°C, 0.5 ml of the substrate (Prepared by dissolving 30 mg 2-oxoglutarate and 1.57 g L-aspartate mono-sodium salt in 50 ml of distilled water for GOT estimation and by dissolving 30 mg 2-oxoglutarate and 1.78 g DL-alanine in 50 ml of distilled water for GPT estimation) was added and incubated for 60 minutes at 37°C. After 60 minutes, the enzyme activity was stopped by the addition of 1 ml of 1 mM 2, 4-Dinitrophenyl hydrazine (Chromogen). The mixture was shaken well and allowed to stand for 20 minutes at room temperature. The reaction was stopped by adding 10 ml of 0.4 N NaOH. After 5 minutes, the hydrazone formed was measured spectrophotometrically at 546nm.

Enzyme activity was calculated from calibration curve prepared using known concentrations of sodium pyruvate. The amount of protein in each sample was calculated by following the method of Lowry et al. (1951). From this, the specific activity of the GOT and GPT (μM/mg protein/h) was calculated. The results were analysed using student's 't' test (Zar, 1974).

RESULTS

AAT (GOT) and ALAT (GPT) activity in the liver and kidney of the experimental and control fishes are presented in Tables 9, 10, 11, 12, 13 and 14 and Figs. 9, 10, 11, 12, 13 and 14 respectively.
AAT (GOT) activity in liver:

The liver of 100 µg/l copper-dosed fishes showed significantly higher GOT activity at 72 h ($P < 0.05$), 120 h ($P < 0.05$) and 168 h ($P < 0.01$) when compared with the controls. A similar increase also was observed ($P < 0.05$) in the fishes treated with higher concentration of copper at 72, 120 and 168 h. Liver of fishes dosed with 100 µg/l mercury showed significant increase at 72 h ($P < 0.05$) and 120 h ($P < 0.05$) whereas those dosed with 150 µg/l mercury showed significant increase at 72, 120 and 168 h ($P < 0.05$).

AAT (GOT) activity in kidney:

GOT activity in the kidney of fishes dosed 100 µg/l copper and mercury showed lower values at 72, 120 and 168 h ($P < 0.01$) whereas kidney of fishes dosed with 200 µg/l copper and 150 µg/l mercury showed significantly lower values at 24, 72, 120 and 168 h ($P < 0.01$) when compared with the controls.

ALAT (GPT) activity in liver:

Liver of fishes dosed with 100 µg/l copper showed significantly lower activity at 24 h ($P < 0.05$) and 72 h ($P < 0.01$) whereas liver of fishes dosed with 200 µg/l copper showed significantly lower values at 24, 72 ($P < 0.01$) and 120 h ($P < 0.05$). Liver of fishes dosed with 100 µg/l mercury showed significantly lower values at 24 ($P < 0.05$), 72 and 120 h ($P < 0.01$) and those dosed with 150 µg/l mercury showed significantly lower values at 24 ($P < 0.05$), 72, 120 ($P < 0.01$) and 168 h ($P < 0.05$) periods.

ALAT (GPT) activity in kidney:

GPT activity in the kidney of fishes exposed to 200 µg/l copper showed a significant decrease at 72 h ($P < 0.05$). GPT activity in the kidney of fishes exposed to 100 µg/l mercury showed significant decrease at 72 ($P < 0.01$) and 120 h ($P < 0.05$) whereas those exposed to 150 µg/l mercury showed significant decrease at 24 ($P < 0.05$), 72, 120 ($P < 0.01$) and 168 h ($P < 0.05$) when compared with the controls.

When the ratio of AAT/ALAT (GOT/GPT) in liver and kidney was
### Table 9. Liver aspartate amino transferase (glutamic oxaloacetic transaminase) activity in *Oreochromis mossambicus* exposed to copper and mercury

<table>
<thead>
<tr>
<th>Concentration ( \mu g/l )</th>
<th>Specific activity ( \mu M ) pyruvate formed/mg protein/h</th>
<th>Hours of exposure</th>
<th>( \bar{x} \pm S.D ) (N = 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>24 h</td>
<td>72 h</td>
<td>120 h</td>
<td>168 h</td>
</tr>
<tr>
<td>Cu 100</td>
<td>6.15 ( \pm ) 1.22</td>
<td>8.44* ( \pm ) 1.08</td>
<td>8.51* ( \pm ) 1.03</td>
</tr>
<tr>
<td>Cu 200</td>
<td>6.85 ( \pm ) 1.32</td>
<td>8.49* ( \pm ) 1.21</td>
<td>8.60* ( \pm ) 1.14</td>
</tr>
<tr>
<td>Hg 100</td>
<td>7.12 ( \pm ) 1.41</td>
<td>8.80* ( \pm ) 1.13</td>
<td>8.72* ( \pm ) 1.23</td>
</tr>
<tr>
<td>Hg 150</td>
<td>6.74 ( \pm ) 1.15</td>
<td>8.58* ( \pm ) 1.06</td>
<td>8.62* ( \pm ) 1.31</td>
</tr>
<tr>
<td>Control</td>
<td>6.95 ( \pm ) 1.3</td>
<td>7.21 ( \pm ) 1.23</td>
<td>7.30 ( \pm ) 1.06</td>
</tr>
</tbody>
</table>

### Table 10. Kidney aspartate amino transferase (glutamic oxaloacetic transaminase) activity in *O. mossambicus* exposed to copper and mercury

<table>
<thead>
<tr>
<th>Concentration ( \mu g/l )</th>
<th>Specific activity ( \mu M ) pyruvate formed/mg protein/h</th>
<th>Hours of exposure</th>
<th>( \bar{x} \pm S.D ) (N = 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>24 h</td>
<td>72 h</td>
<td>120 h</td>
<td>168 h</td>
</tr>
<tr>
<td>Cu 100</td>
<td>10.02 ( \pm ) 1.51</td>
<td>8.05*** ( \pm ) 1.37</td>
<td>6.76** ( \pm ) 1.36</td>
</tr>
<tr>
<td>Cu 200</td>
<td>6.92** ( \pm ) 1.45</td>
<td>6.69** ( \pm ) 1.52</td>
<td>6.37** ( \pm ) 1.44</td>
</tr>
<tr>
<td>Hg 100</td>
<td>8.75 ( \pm ) 1.41</td>
<td>7.85** ( \pm ) 1.39</td>
<td>7.35** ( \pm ) 1.61</td>
</tr>
<tr>
<td>Hg 150</td>
<td>7.96** ( \pm ) 1.29</td>
<td>7.38** ( \pm ) 1.45</td>
<td>4.12** ( \pm ) 1.42</td>
</tr>
<tr>
<td>Control</td>
<td>9.76 ( \pm ) 1.34</td>
<td>9.92 ( \pm ) 1.32</td>
<td>9.61 ( \pm ) 1.43</td>
</tr>
</tbody>
</table>

* \( P < 0.05 \)  ** \( P < 0.01 \)
Table 11  Liver alanine amino transferase (glutamic pyruvic transaminase) activity in *O. mossambicus* exposed to copper and mercury

<table>
<thead>
<tr>
<th>Concentration µg/l</th>
<th>Specific activity µM pyruvate formed/mg protein/h</th>
<th>Hours of exposure 24 h</th>
<th>72 h</th>
<th>120 h</th>
<th>168 h</th>
<th>X ± S.D. (N = 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cu 100</td>
<td>9.08 ± 1.77</td>
<td>8.58** ± 2.11</td>
<td>9.86 ± 2.08</td>
<td>12.06 ± 2.82</td>
<td></td>
<td></td>
</tr>
<tr>
<td>200</td>
<td>8.72** ± 2.13</td>
<td>8.24** ± 1.88</td>
<td>9.65* ± 1.97</td>
<td>11.65 ± 2.14</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hg 100</td>
<td>9.23* ± 2.08</td>
<td>7.16** ± 2.19</td>
<td>8.42** ± 2.12</td>
<td>9.12 ± 2.06</td>
<td></td>
<td></td>
</tr>
<tr>
<td>150</td>
<td>9.14* ± 1.92</td>
<td>6.55** ± 2.27</td>
<td>7.88** ± 2.36</td>
<td>8.87* ± 2.14</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>11.37 ± 1.62</td>
<td>12.14 ± 1.31</td>
<td>11.78 ± 1.92</td>
<td>11.12 ± 2.10</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 12  Kidney alanine amino transferase (glutamic pyruvic transaminase) activity in *O. mossambicus* exposed to copper and mercury

<table>
<thead>
<tr>
<th>Concentration µg/l</th>
<th>Specific activity µM pyruvate formed/mg protein/h</th>
<th>Hours of exposure 24 h</th>
<th>72 h</th>
<th>120 h</th>
<th>168 h</th>
<th>X ± S.D. (N = 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cu 100</td>
<td>12.58 ± 2.13</td>
<td>14.16 ± 2.19</td>
<td>15.33 ± 2.32</td>
<td>15.66 ± 2.41</td>
<td></td>
<td></td>
</tr>
<tr>
<td>200</td>
<td>13.78 ± 2.52</td>
<td>11.61* ± 1.78</td>
<td>13.67 ± 2.11</td>
<td>14.32 ± 2.28</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hg 100</td>
<td>12.10 ± 1.82</td>
<td>10.96** ± 2.14</td>
<td>11.36* ± 2.07</td>
<td>13.39 ± 2.51</td>
<td></td>
<td></td>
</tr>
<tr>
<td>150</td>
<td>11.07* ± 2.06</td>
<td>10.65** ± 2.01</td>
<td>10.98** ± 1.93</td>
<td>11.82* ± 2.12</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>13.14 ± 1.84</td>
<td>13.87 ± 2.04</td>
<td>14.15 ± 2.33</td>
<td>14.73 ± 2.28</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* P < 0.05  ** P < 0.01
FIGURE 9. LIVER ASPARTATE AMINO TRANSFERASE (GLUTAMIC OXALOACETIC TRANSAMINASE) ACTIVITY IN O. MOSSAMBICUS EXPOSED TO COPPER AND MERCURY

AAT activity μM/mg protein/h

hours

Cu 100  Cu 200  Hg 100  Hg 150  Control
FIGURE 10. KIDNEY ASPARTATE AMINO TRANSFERASE (GLUTAMIC OXALOACETIC TRANSAMINASE) ACTIVITY IN O. MOSSAMBICUS EXPOSED TO COPPER AND MERCURY

AAT activity μM/mg protein/h

0 24 48 72 96 120 144 168 192

hours

Cu 100
Cu 200
Cu 100
Hg 150
Control

Hg 100
Hg 150
FIGURE 11. LIVER ALANINE AMINO TRANSFERASE (GLUTAMIC PYRUVIC TRANSAMINASE) ACTIVITY IN O. MOSSAMICUS EXPOSED TO COPPER AND MERCURY

ALAT activity μM/mg protein/h

0 24 48 72 96
12 10 8
Cu 100
Cu 200
Hg 100
Hg 150
Control
FIGURE 12. KIDNEY ALANINE AMINO TRANSFERASE (GLUTAMIC PYRUVIC TRANSAMINASE) ACTIVITY IN O. MOSSAMBICUS EXPOSED TO COPPER AND MERCURY

ALAT activity µM/mg protein/h

0 24 48 72 96 120 144 168 192

Cu 100 Cu 200 Hg 100 Hg 150 Control
calculated, it showed different pattern in the liver and kidney. In the liver the AAT/ALAT ratio of the metal-dosed fishes increased from that of controls. But in kidney the AAT/ALAT ratio of the experimentals decreased from that of controls. The decrease of the AAT/ALAT ratio in kidney was neither dose nor exposure dependent. But in liver the rise in ratio was dose dependent.

DISCUSSION

The results indicate that there is a significant increase in the GOT activity in the liver of fishes exposed to copper and mercury. There is no significant difference in enzyme activity pattern between copper-dosed and mercury-dosed fishes. Similarly there is no significant difference in the pattern of enzyme activity between concentrations of the same metal employed for dosing. Variation of GOT activity in response to toxicants have been reported earlier. Increase in GOT activity in liver, among other tissues were reported in alevins of brook trout in response to methyl mercuric chloride and cadmium (Christensen, 1975); in *Notopterus notopterus* in response to phenol (Verma et al., 1982; Gupta and Dalela, 1985); in response to phenolic compounds (Gupta et al., 1983); in *Porophrys vetulus* in response to Carbon tetra chloride (CCl₄) (Cassilas et al., 1983); in *Tilapia* in response to methyl parathion (Prasada Rao and Ramana Rao, 1984); in *Clarias batrachus* in response to lithium (Goel et al., 1985); and in *Oreochromis mossambicus* in response to napthalene, toluene and phenol (Dange, 1986 b). An increase in the GOT (AAT) activity in the blood was observed in *Poecilia latipinna* in response to dieldrin (Lane and Scura, 1970); in brook trout in response to copper (McKim et al., 1970); in carp in response to PCB (Ito and Murata, 1980); in *Channa punctatus* in response to tri amino azobenzene (Goel and Garg, 1980); in rainbow trout in response to sewage (Wieser and Hinterleitner, 1980); in rainbow trout in response to copper and formalin (Wotton and Williams, 1980); in *N. notopterus* in response to various toxicants (Verma et al., 1981 b); in *Aphanius dispar* in response to mercury (Hilmy et al., 1981); in different fishes in response to copper sulphate (Nemcsok and Boross, 1982) and ammonia and paraquat (Nemcsok et al., 1982, 1985); in *N. notopterus* in response to mercury (Verma et al., 1986); and in carp in response to cadmium (Yamawaki et al., 1986).

However, no significant change in GOT activity in liver was observed
Table 13  Ratio of AAT/ALAT (GOT/GPT) in the liver of *O. mossambicus* exposed to copper and mercury

<table>
<thead>
<tr>
<th>Concentration</th>
<th>24 h</th>
<th>72 h</th>
<th>120 h</th>
<th>168 h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cu 100 µg/l</td>
<td>0.68</td>
<td>0.98</td>
<td>0.86</td>
<td>0.71</td>
</tr>
<tr>
<td>Cu 200 µg/l</td>
<td>0.79</td>
<td>1.03</td>
<td>0.89</td>
<td>0.74</td>
</tr>
<tr>
<td>Hg 100 µg/l</td>
<td>0.77</td>
<td>1.23</td>
<td>1.04</td>
<td>0.87</td>
</tr>
<tr>
<td>Hg 150 µg/l</td>
<td>0.74</td>
<td>1.31</td>
<td>1.10</td>
<td>0.98</td>
</tr>
<tr>
<td>Control</td>
<td>0.61</td>
<td>0.59</td>
<td>0.62</td>
<td>0.64</td>
</tr>
</tbody>
</table>

Table 14  Ratio of AAT/ALAT (GOT/GPT) in the kidney of *O. mossambicus* exposed to copper and mercury

<table>
<thead>
<tr>
<th>Concentration</th>
<th>24 h</th>
<th>72 h</th>
<th>120 h</th>
<th>168 h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cu 100 µg/l</td>
<td>0.80</td>
<td>0.57</td>
<td>0.44</td>
<td>0.45</td>
</tr>
<tr>
<td>Cu 200 µg/l</td>
<td>0.50</td>
<td>0.58</td>
<td>0.47</td>
<td>0.44</td>
</tr>
<tr>
<td>Hg 100 µg/l</td>
<td>0.72</td>
<td>0.72</td>
<td>0.65</td>
<td>0.49</td>
</tr>
<tr>
<td>Hg 150 µg/l</td>
<td>0.72</td>
<td>0.69</td>
<td>0.38</td>
<td>0.35</td>
</tr>
<tr>
<td>Control</td>
<td>0.74</td>
<td>0.72</td>
<td>0.68</td>
<td>0.66</td>
</tr>
</tbody>
</table>
FIGURE 13. RATIO OF AAT/ALAT (GOT/GPT) IN THE LIVER OF O. MOSSAMBICUS EXPOSED TO COPPER AND MERCURY
FIGURE 14. RATIO OF AAT/ALAT (GOT/GPT) IN THE KIDNEY OF O. MOSSAMBICUS EXPOSED TO COPPER AND MERCURY

AAT/ALAT Ratio

0.9
0.7
0.5
0.3

hours

0 24 48 72 96 120 144 168 192

- Cu 100  - Cu 200  * Hg 100  ■ Hg 150  ○ Control
in *Anabas testudineus* in response to lindane (Bakthavathsalam and Reddy, 1982 a) and in *Nemachelius denisonii* in response to a herbicide basalin (Rashatwar and Ilyas, 1983) whereas significantly lower GOT activity in liver was observed in *Tantogolabrus adspersus* in response to cadmium (Gould and Karolus, 1974; MacInnes et al., 1977); in embryos of brook trout in response to methyl mercury (Christensen, 1975); in striped bass in response to cadmium (Dawson et al., 1977); in *Tilapia mossambica* in response to ammonium toxicity (Chetty et al., 1980); in *Cyprinus carpio* in response to cadmium (Koyama et al., 1985); in *C. batrachus* in response to aflatoxin B (Parashari and Saxena, 1986). A decrease in GOT activity in the blood was observed in brook trout after copper exposure for 337 days (McKim et al., 1970); in brook trout in response to lead (Christensen et al., 1977); in Kuwait mullet in response to lead (Helmy et al., 1979).

It should be noted that the results of the present study varied markedly from a similar study using *T. adspersus* (Gould and Karolus, 1974; MacInnes et al., 1977); *T. mossambica* (Chetty et al., 1980) and *Mugil cephalus* (Hilmy et al., 1985) when dosed with cadmium. As reported earlier they observed a significantly lower GOT activity in the liver.

In the present study, in copper and mercury - dosed fishes, the GOT showed a significant increase in activity in the liver and a significant decrease in activity in the kidney. In the kidney also there is no significant difference in enzyme activity pattern between the copper-dosed and mercury-dosed fishes. However, the kidney of fishes exposed to higher concentrations of copper and mercury showed lower GOT activity levels for longer duration than the kidney of fishes exposed to lower concentrations of copper and mercury. Studies dealing with GOT activity in kidney in response to heavy metals are few. Significant decrease in GOT activity in the kidney of *T. mossambica* in response to ammonia toxicity (Chetty et al., 1980); in *C. batrachus* in response to aflatoxin B (Parashari and Saxena, 1986) and significant increase in GOT activity in the kidney, besides other tissues in *N. notopterus* in response to phenol (Verma et al., 1982; Gupta and Dalela, 1985); in response to phenolic compounds (Gupta et al., 1983); and in *C. batrachus* in response to lithium (Goel et al., 1985) have been reported.

In response to stress due to exposure to copper and mercury GOT activity
was different in the liver and kidney. In the liver when the GOT showed
a significant increase in activity over the controls, GOT showed a significant
decrease in activity in the kidney.

When the GOT showed a significant increase in activity in the liver of
fishes exposed to copper and mercury, GPT showed a significantly lower activity.
It appears that there is difference in the GPT activity pattern between copper-
dosed and mercury-dosed fishes. Lower GPT activity lasted longer in the
liver of mercury-dosed fishes. This type of longer duration of lower GPT
activity is shown in the liver of fishes dosed with higher concentrations of
copper and mercury, when compared with the GOT activity in the liver of
fishes exposed to the respective lower concentrations copper and mercury.
Significant decrease in GPT activity in liver in response to pollutants have
been reported earlier. These include, the decreased GPT activity in the liver in
rainbow trout in response to carbon tetrachloride (Racicot et al., 1975); in
C. carpio dosed with cadmium (Koyama et al., 1985); in N. denisonii in
response to the effect of herbicide basalin (Rashatwar and Ilyas, 1983); in
M. cephalus in response to cadmium (Hilmy et al., 1985) and in C. batrachus
in response to aflatoxin B (Parashari and Saxena, 1986). However increase
of GPT activity in liver besides other tissues, was also reported as in
N. notopterus in response to phenol (Verma et al., 1982; Gupta and Dalela,
1985); in response to phenolic compounds (Gupta et al., 1983); in Tilapia exposed
to methyl parathion (Prasada Rao and Ramana Rao, 1984); in C. batrachus
in response to lithium (Goel et al., 1985); and in O. mossambicus in response
to naphthalene, toluene and phenol (Dange, 1986 b).

Based on the present study, it can be said that in the liver, the trend
of increased GOT activity persists in the later period of study, whereas the
trend of decreased GPT activity prevails in the early period of the study and
this is more pronounced in those experimental groups dosed with lower
concentrations of copper and mercury.

The results of the present study indicate that in the kidney of metal-
dosed fishes, lower GPT activity is more pronounced in the mercury-dosed
fishes. Unlike the GPT activity in the liver of the heavy metal dosed fishes,
GPT activity in the kidney does not show a definite trend, the only comparison
being the lower GPT activity in the liver and kidney of the fishes dosed with higher concentration of mercury. Studies dealing with GPT activity in the kidney in response to heavy metals are few. Reports of GPT activity in the kidney of fishes in response to toxicants include a significant decrease in GPT activity in *C. batrachus* in response to aflatoxin B (Parashari and Saxena, 1986); increase in GPT activity in *N. notopterus* in response to phenol (Verma et al., 1982; Gupta and Dalela, 1983) and in response to phenolic compounds (Gupta et al., 1983); in *C. batrachus* in response to lithium intoxication. There are also reports that various toxicants cause an increase in serum GPT (Goel and Garg, 1980; Wieser and Hinterleitner, 1980; Wotten and Williams, 1980; Hilmy et al., 1981; Dalich et al., 1982; Nemcsok and Boross, 1982; Nemcsok et al., 1982, 1985; Cassilas et al., 1983; Verma et al., 1986; Yamawaki et al., 1986).

In fishes, processes like metal detoxification and repair of cellular damage in the tissues require greater energy demands. This shows that when the fishes are subjected to sufficiently high, but sublethal pollution stress, it could lead to abnormal exhaustion of the important energy stores (see chapter 2), while not actually causing death, this type of stress could still be responsible for some reduction in the amount of metabolic energy that can be invested in the growth and development of gonads, thereby affecting the reproductive success and consequently the chances of survival of the pollution as a whole (Dange, 1986 b).

Dange and Masurekar (1981, 1982, 1984) detected an increase in glycogenolysis in fishes exposed to different pollutants. Energy produced from glycolysis is augmented with energy from amino acid metabolism for which aminotransferases are necessary. In fishes, the transamination reaction probably play a significant role at some stage in the degradation of muscle protein (Siebert and Schmitt, 1965). It is probable that the pool of amino acids present in fish tissues is utilized for transamination to produce a variety of keto acids. Increased protein breakdown is probably mediated through increased secretion of cortisol in chronically stressed fish (Dange, 1986 b). Cortisol is known to stimulate the aminotransferase activities in fish tissues (Garbus et al., 1967; Storer, 1967; Freeman and Idler, 1973). Alanine and aspartic acid serve as two major glucogenic amino acids which through the activities of GOT (AAT) and GPT (ALAT) give rise to glucose precursors (Lehninger, 1979). GOT (AAT) and GPT (ALAT) are known to act as an important link between carbohydrate...
and protein metabolism, providing a source for keto acids for Krebs' cycle and gluconeogenesis. An increase in GOT and GPT activity along with acceleration of glycogenolysis was observed in *C. batrachus* (Goel et al., 1985). Hence a stimulation of GOT (AAT) or GPT (ALAT) would alter the amino acid metabolism which in turn disturb all the metabolic processes of the body. So an increased GOT activity could supply more intermediate products for energy metabolism as the body requires more energy to meet the metal stress (Weber, 1963). In the present study, GOT (AAT) increases in the liver whereas GPT (ALAT) decreases in the liver on exposure to metals. But in the kidneys both the transaminases, decrease when dosed with both metals. The two tissues, liver and kidney show a differential response towards the metals. This could be due to their different functions.

Glutamic acid is one of the three amino acids required for the synthesis of glutathione, necessary for the detoxification of pollutants (see chapter 6). Hence, during metal stress, increased quantities of glutamic acid are required for this purpose. An increased transamination activity by GOT and GPT can synthesise glutamic acid from *α*-keto glutaric acid. However, increased GPT activity also results in the increased production of pyruvic acid and during hypoxic conditions may be converted to lactic acid which may increase acidosis. There are many reports that metals increases the lactic acid production in fishes (see chapter 2). An increase in blood pyruvate, lactate etc. was observed during ammonia toxicity (Prior and Visek, 1972). Since GPT catalyses the formation of pyruvate from alanine, excess pyruvate formed may inhibit the GPT activity due to feed back mechanism. This may be reducing the synthesis of ALAT (GPT) which will be manifested as a decreased ALAT (GPT) activity. So product inhibition and reduced synthesis of GPT may be playing a key role in decreasing the GPT activity initially. When the pyruvate formed is used up for energy production, the GPT activity reverts to normalcy. In the present study also, GPT shows such a general trend in both liver and kidney and the GPT activity returns to control values at the end of the experiment.

Another role of glutamic acid lies in its ability to remove ammonia. Ammonia is constantly produced in liver due to the deamination process and during toxicant-mediated stress, ammonia production increases. Carbon tetrachloride is known to increase the production of a blood ammonia in fish
(d'Apollonia and Anderson, 1977). Many authors (McBean et al., 1966; Janicki and Lingis, 1970; Hochachka and Somero, 1973) have pointed out that GOT (AAT) and GPT (ALAT) play an important role in the production and detoxification of ammonia. The ammonia produced is thus removed from the circulation by the liver by converting it to glutamine or to urea. Formation of glutamine is catalysed by glutamine synthetase. Glutamic acid combines with ammonia in the presence of glutamine synthetase, ATP and Mg$^{++}$ ions to form glutamine. Thus synthesis of glutamine from glutamic acid removes NH$_3$. The immediate source of glutamic acid for this purpose is $\alpha$-keto glutaric acid. This would rapidly deplete the supply of citric acid cycle intermediates unless replaced (Harper and Rodwell, 1975). GOT can increase the supply of both oxaloacetic acid which is a citric acid cycle intermediate and glutamic acid which removes ammonia. Here, there is a switching over from alanine transamination to aspartate transamination. Tissues when subjected to trauma, switches over to aspartate transamination from alanine transamination (Malhotra et al., 1986). Such a switching over, increases the GOT levels and depress the GPT activity. In the liver of metal exposed fishes, such a switching over from alanine transamination to aspartate transamination may be occurring. It may be possible that observed increase in GOT (AAT) activity and decrease in GPT (ALAT) activity in the liver is the manifestation of this switching over.

In the kidney, which is the major excretory organ for nitrogenous waste products, glutamine is converted back to glutamic acid catalysed by glutaminase releasing ammonia. The ammonia liberated is removed through urine. Increased urine flow after exposure to toxicants was postulated by Lock et al. (1981). The excretion into the urine of the ammonia produced by renal tubular cells constitute a far more significant aspect of renal ammonia metabolism. Ammonia production forms part of the renal tubular mechanism for regulation of acid-base balance as well as conservation of cations (see Harper and Rodwell, 1975). Ammonia production by the kidney is markedly increase in metabolic acidosis and is derived particularly from glutamine catalysed by glutaminase which converts glutamine to glutamic acid. The glutamic acid concentration increases in the kidney which may inhibit both the transminases, GOT and GPT in the kidney. The observed decrease of GOT and GPT activity in the kidney could be due to this phenomenon. A differential response by GOT
and GPT was found in the liver of rainbow trout dosed with CCl₄ (Racicot et al., 1975). A drop in GPT activity at the 3rd day after CCl₄ injection was observed by Racicot et al. (1975), but there was no change in the GOT activity in the liver of fish dosed with CCl₄. The GPT activity returned to control levels at the 10th day. Hilmy et al. (1985) also observed a differential response of AAT (GOT) and ALAT (GPT) in the liver of M. cephalus exposed to cadmium up to 48 h, where the AAT (GOT) increased but GPT decreased. However, the AAT (GOT) activity later decreased whereas inhibition of ALAT (GPT) activity continued throughout the experiment. There are many other reports in fishes in which the GPT activity decreased while GOT activity either increases or does not change, so that there is a rise in the GOT/GPT ratio (see Tiedge et al., 1986; Racicot et al., 1975; Verma et al., 1982; Rashatwar and Ilyas, 1983). However, these authors did not give the actual GOT/GPT ratios. Eventhough Dange (1986 b) found that both aspartate aminotransferase (AAT or GOT) and alanine aminotransferase (ALAT or GPT) increased in the liver of O. mossambicus exposed to toluene, naphthalene and phenol, if the liver AAT/ALAT (GOT/GPT) ratio was calculated in controls and fishes exposed to lower concentration of naphthalene and toluene for 10 weeks, there was an increase in the AAT/ALAT ratio in the dosed fishes from that of the controls. Since there are many reports about increase, decrease or no change in the activities of GOT or GPT, it would be better to calculate the GOT/GPT ratios in different tissues to understand the exact role played by these enzymes.

In the present study also the pattern of activity of GOT and GPT in the liver were different from that of kidney. It is pertinent to observe the findings of Malhotra et al. (1986) that tissues when subjected to trauma switches over to aspartate transamination from alanine transamination which increases the GOT levels and depress the GPT activity. According to his observation, regions of higher GOT levels tend to show lower GPT levels. The GOT and GPT activities obtained in the present study tends to support this observation. If this is true a ratio of GOT/GPT will be a better estimate of tissue reaction towards xenobiotics, as this will take into consideration both the enzyme activity pattern.

In the present study, the ratio of GOT/GPT in the liver and kidney vary slightly. The kidney show a higher GOT/GPT ratio than the liver of control
fishes. In the liver GOT/GPT ratio of the metal dosed fish shows an increase from that of controls in all days. The ratio increases to its highest level at 72 h and show a tendency to fall. In the kidney, a totally different trend was seen. The GOT/GPT ratio began to drop in the kidney of *O. mossambicus* dosed with copper and mercury. The decrease began at 72 h in the kidney of copper treated fish and a maximum decrease was observed at 168 h. But in mercury-dosed fishes, the GOT/GPT ratio fell at 120 h onwards.

However, it is to be noted that Goel and Garg (1980) reported a GOT/GPT ratio of less than 1.0 in the serum of toxicant treated fish and a ratio of more than 1.0 for the normal fish. In the present investigation, the GOT/GPT ratio of the metal treated fishes was more than that of the controls in the liver and less than that of the controls in the kidney. It is felt that GOT/GPT ratio may be a better estimate of the stress reaction in different tissues of fish exposed to toxicant. A high or low GOT/GPT ratio from the normal may indicate a stress response in this fish. However more research work is necessary before fixing arbitrary values for measuring the stress response in *O. mossambicus* exposed to heavy metals.