Review of Literature
Chapter II

REVIEW OF LITERATURE

2.1. LEAD IN MARINE SEDIMENTS

The carry metal in solution, adsorbed on inorganic solids as cat-ions, on organic solids and in detrital crystalline materials (Gibbs, 1973). The role of rivers in transporting detrital material from continents to oceans is paramount, being 10 times that of glaciers and 100 times that of wind (Goldberg, 1976). All heavy metals become toxic at some concentration (Bryan, 1971).

Ndiokwerre (1984) studies heavy metal pollution and its effect on soil, vegetation and crops. Estuaries and coastal area exhibit a wide array of human impacts that can compromise their ecological integrity, because of rapid population growth and uncontrolled development in many coastal regions worldwide (Kulikova et al., 1985). Wallace and Cooper (1986) have compiled a list of 120 occupations for example auto-mechanic, painting, printing, welding and etc., that may involve exposure to lead. Several studies indicated that plants have the ability to concentrate lead (Sridhar, 1988). Lead and root contain more lead than stem, and the contents of lead in different plant organs were positively correlated to the lead content in soils. Commonly the Pb does not concentrate in the edible fruited part of the plant. Osibanjo and Ajayi (1989) reported that the highest level of Pb occurs in Aviation gas (915 μg/ml) and super grade gasoline, 600-800 μg/ml (with a mean of 700 μg/ml).
The comparable maximum levels in United States and Britain (UK) being 200 µg/ml and 500 µg/ml, respectively (Osibanjo and Ajaiyi, 1989). The authors however reported that trace amounts of Pb (2-7 µg/ml) are present in diesel oil, kerosene and lubricating oil. The variation of heavy metal in salt water was low but the mean concentration was high and exceeded most of the levels recorded from a variety of inland surface waters in Africa (Okoye et al., 1994). The concentration range is higher than that detected along the continental shelf of Pakistan (Tariq et al., 1994).

Studies have shown that the body lead levels of modern humans are about 500 times higher than those of pre-industrial times. Organic lead compounds (tetraethyl lead and tetramethyl lead) are extensively used as additives in petrol. It has been pointed out that Africa's contribution to global lead pollution has increased from just 5% in 1980s to 20% in 1996 (Anon, 1996).

The heavy metals that are emitted in the atmosphere in the form of aerosols, as by product are taken away by wet or dry deposition and cause major problems to the surface waters and the organisms (Finlayson-Pitts and Pitts, 2000; Quiterio et al., 2004a; Quiterio et al., 2004b).

In contrast, the total values in this study are based on the whole sediment with only very coarse (> 2 mm) detritus removed. It is generally accepted that contaminants are likely to be almost entirely associated with
particle sizes of <63 μm (i.e., silt and clay-sized particles, collectively defined as "mud" by Loring and Rantala (1992) and that sand-sized particles act to dilute metal contamination in sediments. In the three profiles examined in the present study, the <63-μm size fraction generally comprises <30% of each profile. If the total Cu, Pb and Zn concentrations were corrected for the abundance of <63-μm particles, then a relatively large proportion of depth intervals do indeed exceed the background values. This supports the assertion that sediment texture should be considered when assessing metal enrichment in sediments (Loring and Rantala, 1992). Cadmium (Cd), copper (Cu), lead (Pb) and zinc (Zn) are the most present trace heavy metals in the wastewater of the Gulf (Abdennour et al., 2000, 2004).

Okoye (1994) in a survey of Pb and other metal contents of dried fish from Nigerian markets remarks that the Pb content is high. He observes that the high Pb content in fish from Nigerian markets indicates serious Pb contamination in Nigeria. He attributes this mainly to heavy automobile traffic and the high lead content of the local automobile fuels.

Exposure to Pb can occur through multiple pathways, including inhalation of air and ingestion of Pb in food, water, soil or dust. Excessive Pb exposure can cause seizures, mental retardation and behavioural disorders. The danger of Pb is aggravated by low environmental mobility even under high precipitations (Mench et al., 1994).
Though some of the metals such as Cu, Fe, Mn, Ni and Zn are essential as micronutrients for life processes in plants and microorganisms, while many other metals like Cd, Cr and Pb have no known physiological activity, but they are proved detrimental beyond a certain limit, which is very much narrow for some elements like Cd (0.01 mg/L), Pb (0.10 mg/L) (ISI, 1982) and Cu (0.050 mg/L) (Bruins et al., 2000).

Lead concentrations in clean coastal sediments are around 25 μg g\(^{-1}\) or less (United Nations Environment Programme (UNEP), 1985; Bryan and Langston, 1992) and the average Pb levels in Indian River sediments is about 14 mg kg\(^{-1}\) (Decov et al., 1999). However the present values are below the USEPA (1996).

Heavy metals studies in aquatic organisms, along with bioconcentration have been extensively studies in various places around the world (Vijayram and Geraldine, 1996; Teodorovic et al., 2000; Wu and Chen, 2004; Amaranemi, 2006; Dural, 2007; Sivaperumal et al., 2007; Yilmaz et al., 2007; Hamilton, 2008; Sharif et al., 2008).

The indiscriminate use of heavy metal containing fertilizers and pesticides in agriculture resulted in deterioration of water quality rendering serious environmental problems posing threat on human beings (Lantzy and Mackenzie, 1979; Nriagu, 1979; Ross, 1994) and sustaining aquatic biodiversity (Ghosh and Vass, 1997; Das et al., 1997).
It is important to note that the background Cu, Pb and Zn concentrations quoted above reflect Cu, Pb and Zn concentrations in the <63-μm size fraction of Queensland estuarine sediments (Moss and Costanzo, 1998).

Lead, one of the oldest known metals, is also one of the most widespread toxicants, and lead poisoning remains a health threat (Hernberg, 2000). It is well established that organic matter contents are important controlling factors in the abundance of trace metals (Rubio et al., 2000).

Effluents are indiscriminately discharged into natural waters, thereby contaminating aquatic ecosystem (Fafioye et al., 2002). Frequently, anthropogenic emissions cause the levels of metal in suspended particles to be above natural background levels (Finlayson-Pitts and Pitts, 2000; Quiterio et al., 2004a and 2004b).

Industrial discharges containing toxic and hazardous substances, including heavy metals contribute tremendously to the pollution of aquatic ecosystems (Gbem et al., 2001; Woodling et al., 2001).

Metals enter the environment and oceans by two means: natural processes (including erosion of ore-bearing rocks, wind-blown dust, volcanic activity and forest fires); and processes derived from human activities by means of atmospheric deposition, rivers and direct discharges of dumping (Clark, 2001).
The highest lead concentrations were found in sediments collected in October and December. Very high lead concentrations (> 10 lg g⁻¹) were observed in mussels from those stations where the sediments also contained high concentrations. Mussels had their highest lead concentrations in January and April (Unsal, 2001).

Thus, copper, zinc, cadmium and lead were selected to develop toxicity bioassays, bearing in mind that they have been recognized as a potential risk for this environment (Ferrer, 2001).

According to Hashmi et al. (2002) continental sources (river runoff and atmospheric transport), oceanic sources (upwelling) and diagenetic exchanges at water-sediment interface have been identified as the factors that influence the heavy metals in aquaculture organisms. Moreover, anthropogenic atmospheric inputs, sewage sludge and fertilizers are often inferred to be significant because of important these metals input.

Anthropogenic activities like mining, ultimate disposal of treated and untreated waste effluents containing toxic metals as well as metal chelates from different industries, e.g. tanner, steel plants, battery industries, thermal power plants etc. (Amman et al., 2002).

In recent years, much attention has been directed to the concentrations of some inorganic elements in marine fish and other aquatic organisms (Mansour and Sidky, 2002; Farkas et al., 2003). With the advent of agricultural and industrial revolution, most of the water
sources are becoming contaminated (Khare and Singh, 2002). In sediment phase, during monsoon and pre-monsoon periods heavy metal concentrations were observed minimum and maximum respectively. Apart from acute toxicity and aforesaid metals in water has proved dangerous and harmful due to their bioaccumulation and their impact on tissue degeneration, thus influencing growth, survival and reproductive potential of animals with special reference to fishes (Gupta et al., 2002).

Significant variations in Cd, Cu, Hg, Pb and Zn concentrations occur in surficial marine sediments collected in the Bay of Chaleur, an estuary located between northern New Brunswick and Quebec’s Gaspe Peninsula. The bay receives metals from a variety of sources including a lead smelter, who thermal generating stations, a mercury-cell chlor-alkali plant, and numerous mined and unmined base-metal deposits. The purpose of this study is to assess the fluxes and dispersal patterns of metals released to the bay from both natural and anthropogenic sources, and to characterize the processes that collect and redistribute metals in the marine environment (Parsons, 2002).

The present experiment reported made on the studied area concerning the trace heavy metals (Abdennour et al., 2000, 2004) or on D. trunculus (Abbes et al., 2003; Bouzeraa et al., 2004). Shrimp farmers in Nigeria do not have access to an adequate supply of healthy, domesticated seed to stock their ponds because post larvae typically are
produced from wild-caught brood stock. Wild brood stocks are often polluted with anthropogenic wastes in the lagoon (Moss, 2004).

The estuary has become vulnerable to chemical pollutants such as heavy metals, organochlorine pesticides, petroleum-derived hydrocarbons, chlorinated hydrocarbons etc., which may have changed the estuary’s geochemistry and affected the quality of the local coastal environmental (Sarkar et al., 2002, 2003, 2004, Saha et al., 2006).

Metal accumulation was found to be highest during the period just after the summer monsoon due to the flushing of previously deposited sediments. As the season progresses, they are further impoverished in metals (normalized) through size segregation of marine sediments. The correlations between various sedimentary parameters are elucidated to interpret the coherence between metallic and carrier phases during each season (Balachandran et al., 2003).

This salinity effect was strongest for cadmium and lead and could be attributed to complexation with chloride ions. The toxicity of nickel, copper and zinc was affected to a smaller extent by salinity (Verslycke et al., 2003). Lead levels in soils vary depending on the location and nearness to lead based activities and vehicular density (Chirenje et al., 2004). All metals, except Cu and Cd, resulted accumulated in the finest fraction, which constituted up to 95 per cent of most of the considered sediments. A good correlation has been found for Ni, Cr, Fe, Zn, Cu in the
entire area, indicating a common origin for these metals in the analyzed sediments. Though in some selected areas such as the Drin and the Skumbin Bay, an anthropogenic input of Cr, Fe, Ni and Cu can be observed, as the result of discharging of mines and smelter activities (Rivaro et al., 2004).

One of the greatest concerns for human health is caused by lead (Pb) contamination. Lead (Pb) occurs naturally in all soils, in concentrations ranging from 1 to 200 mg/kg, with a mean of 15 mg/kg (Chirenje et al., 2004). Metal enrichment is higher in the bay than in the adjacent coastal zone, however an important increase especially in Pb content was detected in an area previously considered as a pristine one. Considering both metal content and benthic environment characteristics, the study area can be clearly divided in at least two well-defined regions. One is the inner region of the bay near the streams and the outermost stations of the bay and the adjacent coastal zone form the other. The first one can be considered highly polluted and the other moderately polluted (Muniz et al., 2004).

The role coastal sediments play in biogeochemical cycles of many trace metals is poorly understood. For metals such as Silver, Copper and Lead, this is especially important, as elevated concentrations are often found in coastal sediments due to anthropogenic activities. Sediments are large sources of these heavy metals (American Geophysical Union/Ocean
Estuaries and coastal area received significant anthropogenic inputs from both point and nonpoint upstream sources and from metropolitan areas, tourism and industries located along the estuarine edges (Caeiro, 2005). The association of Cu, Pb and Zn with amorphous oxides, crystalline oxides and organic matter was linearly dependent on the abundance of each respective phase (Burton et al., 2005).

The concentration ranges of exchangeable metals decrease in the order Fe > Cu > Cd > Cr > Ba > Ni > Pb > Co in the vicinity of a refinery and Fe > Cu > Ba > Pb > Cd > Ni > Co > Cr. In descending order of predominance, the overall mean ranges of the exchangeable metal concentrations in the sediments are Fe (0.32-5.82), Cu (0.02-3.00), Ba (0-2.01), Pb (0-1.67), Ni (0.03-1.27), Cd (0.81) and Cr (0.02-1.27), Co (0-0.34). (Adeeyinwoo et al., 2005).

Dispersion of smelter effluents and atmospheric emissions by wind and/or nearshore currents has results in an area of elevated as Cd, Cu, Hg, Pb and Zn concentrations in surficial sediments within 10-20 km of the smelter. The concentrations of most metals decrease sharply with increasing distance from the smelter; however, Pb concentrations exceed background levels in surface sediments throughout the bay. Lead isotope ratios suggest that the surface enrichment of Pb throughout the bay is
mainly derived from smelter emissions and historical leaded gasoline combustion (Parsons and Cranston, 2006). Heavy metals are present in the atmosphere in ever increasing levels as a result of anthropogenic and natural emission (Suzuki, 2006).

Fossil fuel and wood combustion as well as waste incineration and industrial processes are the main anthropogenic sources of metals to the atmosphere. In urban areas and due to road traffics contribute, become the most important manmade source for such pollution (Birmili et al., 2006; Preciado and Li, 2006; Sternbeck et al., 2002; Lin et al., 2005; Harrison et al., 2003), while vehicle ageing and wearing actions release off some heavy metals such as Zn from tires, Cu from brake linings, Mn from moving metals parts and gasoline additives (Preciado and Li, 2006, Swaine, 2000) and Pt, Pd and Rh from catalytic converters of automobiles (Lesniewska et al., 2006).

The overall variation in concentration can be attributed to differential discharge of untreated effluents originating from industrial, agricultural and aquacultural sources as well as from domestic sewage along with the fishing and boating activities. The resulting compositional dataset was tested by principal component analyses and cluster analyses. Pollution load index (PLI) and index of Geoaccumulation (Igeo) revealed overall low values but the enrichment factors (EFs) for Pb were typically high for all the stations (Chatterjee et al., 2006). However, geochemical
studies have focused mainly on surface sediments rather than assessing historical trends. A recent geochemical study of the Guadiana estuary (Caetano et al., 2006) reported results of one core collected at the coastal area. Dural (2007) stated heavy metal pollution in estuaries and coastal area has been recognized as a serious environmental concern.

The pesticides include insecticides, herbicides, fungicides, molluscides and nematicides and heavy metals like copper, zinc, arsenic, lead, cadmium, mercury etc. These pesticides are non-biodegradable and accumulate in the food chain. Mostly they are prone to affect the nervous system causing tumours in living organisms. They are not only neurotoxic but also affect other systems and have shown a high degree of impact on metabolism by altering the proteins, carbohydrate and lipids (Senthilkumar et al., 2007). Accumulation of heavy metals was observed in the order of Sediments > Fish > Water. In water, the order was found to be Mn > Fe > Zn > Cu > Ni > Cd > Co > Pb. Pr recorded a minimum of 0.006 μ.L. In sediments Pb recorded a minimum of below detectable levels. In fish, a minimum of below detectable levels were found in Pb (Martin Deva Prasath and Hidayathulla Khan, 2007). Accumulation of trace metals occur in upper sediment in aquatic environment by biological and geochemical mechanisms and become toxic to sediment-dwelling organisms and fish, resulting in death, reduced growth, or in impaired reproduction and lower species diversity (Praveena et al., 2007).

A total of 96 surface water samples collected from river Ganga in West Bengal during 2004-05 was analyzed for pH, EC, Fe, Mn, Zn, Cu,
Cd, Cr, Pb and Ni. The highest mean concentrations (mg/L) of Fe (1.485), Zn (0.085) and Cu (0.006) were observed at Palta, those for Mn (0.420) and Ni (0.054) at Berhampore, whereas the maximum of Pb (0.024 mg/L) and Cr (0.018 mg/L) was obtained at the downstream station, Uluberia. All in all, the dominance of various heavy metals in the surface water of the river Ganga followed the sequence: Fe > Mn > Ni > Cr > Pb > Zn > Cu > Cd (Kar et al., 2008).

Monitoring of the contamination of soil and sediment with heavy metals is of interest due to their influence on ground water and surface water and also on plants, animals and humans (Suciu et al., 2008). Lead is relatively unavailable to plants when the soil pH is above 6.5 while availability of zinc decreases with increasing soil pH due to increased adsorptive capacity (Nweke et al., 2008).

Elemental concentrations suggest a heavy metal (Cu, Zn and Pb) enrichment at the sections of all the cores. Cu, Zn and Pb are heavy metals associated with mining exploitation along the Iberian Pyrite Belt, one of the most important mining area of southwestern Europe, with massive orebodies of these metals. The combination of enrichment factors down core profiles with sedimentation rate values signifies the beginning of heavy metal pollution in shelf sediments (Corredeira et al., 2008).

Ololade et al. (2008) examined the distribution of heavy metals in stream bed sediment from an oil-producing region in Nigeria in the two
seasons (dry and wet) of the year. Comparison of sediment with guidelines values indicated anthropogenic enrichment and it was considered that only Cu and possibly Pb posed potential threats to the ecology of the area. Pb was also found to higher in locations that were located near industrial areas (Praveena et al., 2008). For this reason, determination of chemical quality of aquatic organisms, particularly the content of heavy metals is extremely important human health (Mokhtar et al., 2009). Pore water content of the samples was extracted in order to determine the concentrations of heavy metals such as lead (Pb), Zinc (Zn), Cadmium (Cd), iron (Fe), manganese (Mn) and Chromium (Cr). In this study, surfer software was used to plot the contouring of heavy metals concentration. These results play an important role in order to determine and visualize the locations, which are affected with the leachate plume (Samuding et al., 2009).

Metal concentrations in the downstream indicate an increase in the pollution load due to movement of fertilizers, agricultural ashes, industrial effluents and anthropogenic wastes. An immediate attention from the concerned authorities is required in order to protect the river from further pollution (Abida Begam et al., 2009).

Ten heavy metals (Fe, Cu, Mn, Cr, Zn, Hg, Pb, Cd, N1 and V) were analyzed in sediment samples in the dry and wet seasons of year 2008 using AAS. Mercury was not detected in all the samples. The mean concentrations of the heavy metals in sediment (0.38 ± 0.03 ppm to 6.619
± 290 ppm – dry season and 0.24 ± 0.05 ppm to 8.144 ± 129 ppm – wet season) were lower than the values recommended in Consensus-Based Sediment Quality Guidelines of Wisconsin (Olubunmi and Olorunsola, 2010).

2.2. HAEMATOLOGICAL PARAMETERS

Lead contamination is worldwide and it is mainly due to human activities. Lead toxicity results in loss of appetite, neurological and haematological problems in young animals including man (Pagalia et al., 1975).

In recent years, much attention has been directed to the concentrations of some inorganic elements in marine fish and other aquatic organisms (Mansour and Sidky, 2002; Farkas et al., 2003). Anthropogenic activities like mining, ultimate disposal of treated and untreated waste effluents containing toxic metals as well as metal chelates from different industries, e.g. tanner, steel plants, battery industries, thermal power plants etc. (Amman et al., 2002). With the advent of agricultural and industrial revolution, most of the water sources are becoming contaminated (Khare and Singh, 2002).

LC₅₀ decreased with increase of time period small size group were more sensitive to mercury at low salinities while larger size groups were more tolerant at high salinities. Penaeus monodon was more sensitive to mercury than Penaeus indicus (Snehalatha Das and Sahu, 2002). The
activity of the haem biosynthetic enzymes o-aminolaevulenic acid dehydratase, coproporphyrinogen oxidase and ferrochelatase were decreased by increasing lead pretreatment. The activity of the haem catabolic enzyme, haem oxygenase were increased by lead pretreatment (Al-Ayed, 2002). Blood samples were collected to obtain serum for biochemical studies and heparinized blood for hematological investigations. RBCs, Hb, Hct and MCHC showed significant elevations, the serum GPT and GOT were increased significantly. LDH, glucose and cortisol were elevated, while serum cholesterol concentration was reduced significantly in *Oreochromis niloticus* during lead pollution (Zaki et al., 2008).

The effect of sublethal concentration of Portland cement powder in solution on some haematological parameters in Nile tilapia (*Oreochromis nilotica* (L.)) mean weight 8.20 ± 0.25 g was investigated using static bioassay system for 70 days. The sublethal concentrations used were 19.60, 9.80, 4.90, 2.45, 1.23 and 0.00 (control) mg/l, there were significant differences (P < 0.05) in the water quality parameters monitored. However, temperature did not show any significant variation (P < 0.05) in both test tanks and the control. Haematological parameters examined include Pack cell volume (PCV), Haemoglobin (Hgb), Total erythrocyte count (TEC), total leucocyte count (TLC) and Erythrocyte Sedimentation Ratio (ESR) which all decreased significant (P < 0.05), the decrease being proportional to the increase in Portland cement powder in solution (Mohamed and Sambo, 2008).
2.3. BIOCHEMICAL COMPOSITIONS

The PL were exposed to a sublethal concentration of lead (1.44 ppm) for 30 days. The major biochemical constituents, including total carbohydrates, proteins, lipids and ninhydrin-positive substances (TNPS) were estimated using standard methods. Lead exposure resulted in retardation of growth with a significant decrease in length and weight occurring at day 10 and onwards. Of all the biochemical constituents, total protein showed the maximum decrease (79.3%) followed by total lipids (68.1%) and then by total carbohydrates (51.4%) in lead-exposed PL. The data suggest lead exposure causes reduced growth and the depletion of biochemical constituents. This may be due to metal interactions and inhibition of metabolic pathways responsible for synthesis of biochemical constituents or to greater utilization of these constituents under metal stress conditions (Chinni and Yallapragada, 2000).

The fish, as a bioindicator species, plays an increasingly important role in the monitoring of water pollution because it responds with great sensitivity to changes in the aquatic environment. The sudden death of fish indicates heavy pollution; the effects of exposure to sublethal levels of pollutants can be measured in terms of biochemical, physiological or histological responses of the fish organism (Mondon et al., 2001).

The biochemical components such as protein, lipid and carbohydrate of the liver of two important penaeid prawns, were significantly reduced, following six days of exposure to 0.005 ppm and
0.01 ppm of mercuric chloride during various reproductive stages i.e., preparatory, prespawning, spawning and postspawning. Liver protein recorded highest in contrast to lipid and carbohydrate irrespective of the species, sex and medium depletion was at 0.01 ppm Hg medium. The effect of mercury was more in *Penaeus indicus* than that of *Penaeus monodon*, the female species and prespawning stage. Liver-lipid deleteriously affected the female *Penaeus indicus* during spawning while carbohydrate affected it prominently during preparatory stage. Hg concentration of 0.01 ppm had much damaging effect on liver. The change caused due to test solutions in the biochemical constituents of the liver of the prawns indicate that female had more affected than male. The protein content indicated decline with the increase of time period, the depletion of percentage also raised with the increase of time exposure for carbohydrates and lipids (Snehalata Das *et al*., 2001).

Heavy metal contamination has been reported in aquatic organisms (Adham *et al*., 2002; Olojo *et al*., 2005). These pollutants build up in the food chain and are responsible for adverse effects and death in the aquatic organisms (Farkas *et al*., 2002).

Cadmium (Cd), one of the twenty three heavy metal toxicants, is widely used in Ni-Cd batteries manufacture, metal and mining industry, dentistry etc. because of its non-corrosive nature. Cd is released in considerable amounts through industrial effluents into soil, surface and ground water systems. These excess amounts in addition to naturally
occurring levels gradually build up to toxic levels causing damage to the biota of the aquatic ecosystem. It shows biomagnifications and has greater half-life periods. Cd was found to interfere with processes. The present study evaluates toxicity of Cd and its impact on biochemical constituents like glucose, glycogen, total proteins, lipid and free amino acids in the freshwater edible crab *Catla catla* as Cd bioaccumulation can affect humans through biomagnifications. Short term tests of acute toxicity were performed over a period of 96 hours using cadmium chloride. The renewal technique was followed by exposing the fish to test solutions of different concentrations in the range of 1 mg/l to 8 mg/l. Preliminary experiments were conducted to choose concentrations that resulted in the mortality of the fish in the range of 10-90%. The toxicity experiments were then conducted using the chosen concentrations of CdCl$_2$ on the fingerlings (wt 6 ± 1 g) in triplicate and the LC$_{50}$ was determined using simple graphic (% mortality vs. log concentration), probit graphic (probit value vs. log concentration) and unweighted regression analysis methods. The calculated average 96 hr LC$_{50}$ is 4.533 mg/l and the equation for the dose-mortality regression line was found to be $Y = 2.65 X + 3.368$. Levels of the five biochemical constituents viz., glucose, glycogen, total proteins, lipids and free amino acids were determined by standard biochemical procedures in the five tissues i.e., muscle, gill, liver, heart and kidney of unexposed (control) healthy fish and fish exposed to 96 hr LC$_{50}$ (lethal) and sublethal concentrations (1/10$^{th}$ of the lethal dose for 7 days of exposure) of cadmium chloride. Results showed significant fall in all the biochemical constituents in all the tissues except glucose prompting to
suggest that the fish cultured in the aquatic systems closer to the industrial locations would not have the expected nutritive value. The elevated levels of glucose are apparently indicative of the organisms response to the toxicant stress. Also, such fish when consumed as food lead to the deposition of the heavy metal in the soft tissues of the human body leading to exposure health effects (Shobha et al., 2007).

Evaluation of the toxic effect of copper on the experimental crab for the LC$_{50}$ value was carried out. Effect of copper on the quantitative study of nutritive value viz., protein, carbohydrate and lipid in ovary, spermatheca, hepatopancreas, muscle, gills, haemolymph, brain, thoracic ganglia and eyestalk was observed similarly (Senthilkumar and Samyappan, 2007).

In nature chromium occurs in divalent, trivalent and hexavalent forms. Hexavalent chromium predominates over the trivalent form in natural waters. Knowledge of acute toxicity of a xenobiotic often can be very helpful in predicting and preventing acute damage to aquatic life in receiving waters as well as in regulating toxic waste discharges. The 96 h LC$_{50}$ tests can be used to obtain toxicity data as rapidly and inexpensively as possible. In the present study, results showed a significant decrease in total glycogen, total lipids and total protein of liver, muscles and gills after 24 and 96 h of exposure to 96 h LC$_{50}$ of hexavalent chromium (43.7 mg/l). Cytotoxicity of metals is important because some metals are potential mutagens able to induce tumours in humans and experimental animals, the
treatment of (Cr VI) at 43.7 mg/l was for 24 and 96 h respectively. DNA was extracted after treatment from brain and liver of the tested fish. Our results showed appearance of polymorphic bands at the long treatment interval (96 h) of hexavalent chromium. However, these bands were not appeared when the fish were exposed to the (Cr.VI) for the short treatment interval (24 h). Histopathological changes were seen in liver, muscle and gills sections of chromium-exposed fishes. The obtained results were discussed in the study research (Abbas et al., 2007).

The present study has been undertaken to investigate, the effect of lethal concentrations 0.33, 0.26, 0.17 and 0.09 ppm of tributyltin chloride on glycogen content in ovary, hepatopancreas, gill and muscle of a freshwater prawn, *Macrobrachium kistnensis* for 24, 48, 72 and 96 hrs respectively. The disturbance in the glycogen profile is one of the outstanding biochemical lesions due to the action of TBTCI. There is significant decrease in glycogen profiles in ovary, hepatopancreas, gill and muscle after exposed to lethal concentration of TBTCI under stress condition. This might be due to increase in glycogenolysis by increase in phosphorylase enzyme activity and elevation of succinate and pyruvate dehydrogenase leading to anaerobic metabolism during anoxic stress condition by toxicant (Kharat et al., 2009).

Endosulfan, a broad-spectrum non-systemic organochlorine (OC) pesticide is extensively used to control a wide variety of pests in agriculture, horticulture and public health programmes. Biochemical
changes occurring in the metabolically active tissues of gills (GL), hepatopancreas (HP) and muscle (MU) of the penaeid shrimp, *Metapenaeus monoceros* (Fabricius) on exposure to two sublethal doses (40 and 60 ng l\(^{-1}\)) of endosulfan were studied for 23 days of exposure (DoE). The results of the study revealed that sublethal doses of endosulfan significantly alters the proximate composition of major tissues, particularly the TP levels in the MU tissues thereby reducing the nutritive value of this economically important penaeid shrimp. Since *M. monoceros* exhibits significant biochemical changes on exposure to endosulfan, this species could possibly be used as biosensor of coastal marine and estuarine pollution (Suryavanish *et al.*, 2009).

Test results indicated that the brackish water juvenile shrimp, *Palaemonetes africanus* were sensitive to the cadmium solution especially at concentration above 4.0 mg/l (Joel and Amajuoyi, 2009).

Evaluation of toxic effect of lead on the edible lobster, *Thenus orientalis* for the LC\(_{50}\) value and effect of heavy metal lead on the nutritional status viz., protein, carbohydrate and lipid in ovary, spermatheca, hepatopancreas, muscle and haemolymph was made. The results assume greater interest as most water bodies are increasingly subjected to environmental pressure due to pollution (Kalyanaraman and Senthilkumar, 2009).

The experimental teleost fish *Heteropneustes fossilis* divided in to four equal groups. Tissue brain, liver, kidney and gills removed and
processed for the estimation of protein contents. It has been observed that lead nitrate cause deleterious effects in all the tissue with reference to protein contents. When chabazite added with lead nitrate, protein contents improved towards normal (Meeta Mishra and Subodh Kumar Jain, 2009).

2.4. BIOACCUMULATION

Liver is the most important target organ for lead and cadmium toxicity (Holcombe et al., 1976 and Dallinger et al., 1987). Exposure of fish to lead via gills effects adversely the body weight and digestive enzymes (Shafi and Choundhary, 1979; Sastry and Gupta, 1978; WHO, 1980 and Jain et al., 1996).

Accumulation of these metals in aquatic organisms such as tiger prawn and tilapia fish was influence by amount of industrialization and urbanization effluent as well as the usage of phosphate fertilizers in agricultural practices received by the Langat estuary (Warren, 1981).

In some countries like Sweden, Japan, Switzerland and Germany, there are maximum permissible levels of metals allowed in fish (about 1 mg/kg body weight). Italy allows 0.7 mg/kg bodyweight while the standard for the United States and Canada allow 0.5 mg/kg body weight (Gerlach, 1981).

However, aquaculture is facing an increasing threat from water pollution. Consequently, small fish and prawns, in polluted aquaculture
will become enriched with the accumulated substances. Predatory fish again, generally displays higher levels than their prey. Eventually human, consuming the fishes and prawns, inevitably suffers from the results of an enrichment taken place at each topic level, where is extracted than ingested (Forstner and Wittmann, 1983).

Many Nigerian farmers have observed poor growth and survival of both *P. monodon* and *M. rosenbergii* stocks, resulting in decreased production and profitability. The reason for this is mainly on the condition of shrimp seed being used for stocking their ponds. Although Asejire lake has not been reportedly polluted, but the estuaries around Agbara and Lekki industrial areas of Lagos contain heavy metals in their surficial sediments (Okoye *et al.*, 1984; Otitoloju, 2000).

A Primary consideration is the bioaccumulation of the metals such as Lead, Cadmium, Zinc and Copper etc. in to the tissues of plants and animals which results intoxic effects Landrigan (1990). Heavy metals can enter food chain and as a result of bioaccumulation, cause serious health problems to human’s. These metals have long biological half-life in bone tissue. Lead continuously accumulates in the bone of humans over the course of exposure which subsequently provides a pool of lead for its slow release over an extended period of years (Long *et al.*, 1990).

Lead is among the most toxic heavy element in the atmosphere. Fergusson (1990) reported that aerosol lead enters the human blood
stream by way of the respiratory tract and indirectly by surface deposition in the elementary tract followed by absorption. Some of the lead in the ambient air around urban centers is in the form of sub-micron sized particulate. The main source of adult human exposure is food, which is believed to account for over 60% of blood levels; air inhalation accounts for approximately 30% and water of 10% (John et al., 1991). All humans have lead in their bodies primarily as a result of exposure to man-made sources (ATSDR, 1992).

Lead is a potentially toxic chemical that may be directly ingested by man or indirectly through aquatic animals like fish and shellfish. The effects of lead on man include mental retardation, learning dysfunction and loss of coordination (Goodman and Gilman, 1992).

A study done by Mokhtar et al. (1994) in Ko-Nelayan tiger prawn aquaculture ponds, in Sabah, Malaysia (Borneo) showed that light industries and workshops nearby aquaculture ponds could influence the water quality of the aquaculture ponds. The water in aquaculture ponds was contaminated by heavy metals such as Pb and Co.

Tiger prawn and fish farming has been one of the major aquaculture products in some Asian countries, such as Indonesia, Philippines, Taiwan, Thailand, Vietnam and including Malaysia since 1987. The major producers of *Penaeus monodon* and *Oreochromis* spp.
include Thailand, Vietnam, Indonesia, India, the Philippines, Malaysia and Myanmar (FAO, 1995).

Although, expansion in aquaculture production of *Penaeus monodon* and *Oreochromis* spp. have been as great as was originally expected, assessments of bioavailability of heavy metals for bioaccumulation are very scarce. This gives an indication that studies on the fate and effect of metals in tiger prawn and fish as major aquaculture products have unfortunately received less attention for both species (Ismail, 1993; FAO, 1995).

This output of the study will play a role as a benchmark and good reference in heavy metal bioaccumulation studies. Thus, the conducted study on heavy metals concentration in *Penaeus monodon* and *Oreochromis* spp. will be a good reference for the future studies in Malaysia and also from other countries in Asia viz. Thailand, Bangladesh, Sri Lanka which involve intensively as major producers and exporter of *Penaeus monodon* and *Oreochromis* spp. Furthermore, these species also have been the popular food source in each country.

Moreover, Shazili et al. (2006) stated that environmental parameters such as salinity, water hardness, pH, temperature as well as analytical methods (dry and wet digestion methods) limit the investigations of farmed aquaculture products such as fishes and crustaceans.
The effects of lead on fish observed during this experiment showed that caution should be exercised in allowing lead into the aquatic environment. These effects included loss of balance, skin bleaching and weakness. A thick layer of mucus on the skin covered the dead fish and there were air bubbles on the water. (James et al., 1996) who observed that fish exposed to sub-lethal levels of lead in the liver and muscle of *Oreochromis mossambicus*.

The percentage mortality of these snails increased with increasing concentration and exposure time. In the bioaccumulation experiment, the snails were exposed to 19.17 mg/l (10% 96 hour LC50) of lead nitrate for 42 days exposure time and 30 days recovery time. During the 42 day exposure period, lead uptake occurred in different organs with the greatest uptake in the intestine, and less in the prostate gland, digestive gland, ovary and albumen gland, testis, stomach and cerebral ganglia. After exposure, lead concentration in all organs decreased during the 30 day recovery period (Jantataeme et al., 1996).

Acute lead toxicity studies were carried out in the snails, Filopaludina (Siamopaludina) martensi martensi (Frauenfoldt). The 96 hr static bioassay was conducted in order to estimate the median lethal concentration (LC50). The snails were exposed to lead nitrate [Pb(NO3)2]. The LC50 for 24, 48, 72 and 96 hrs were 319.47, 271.03, 235.35 and 191.69 mg/l respectively. The percentage mortality of these snails increased with increasing concentration and exposure time. In the
bioaccumulation experiment, the snails were exposed to 19.17 mg/l (10% 96 hr LC₅₀) of lead nitrate for 42 days exposure time and 30 days recovery time. During the 42 day exposure period, lead uptake occurred in different organs with the greatest uptake in the intestine and less in the prostate gland, digestive gland, ovary and albumen gland, testis, stomach and cerebral ganglia. After exposure, lead concentration in all organs decreased during the 30 day recovery period (Jantataeme et al., 1996).

Acute toxicity tests were performed in *Echinogammarus olivii* (Amphipoda), *Sphaeroma serratum* (Isopoda) and *Palaemon elegans* (Decapoda) from the Sinop Peninsula in the Black Sea. 96 h LC₅₀ values were estimated for copper, zinc and lead in these species using the static bioassay method. The results indicated that Cu was more toxic to the species followed by Pb and Zn. *E. olivii* was more sensitive to the metals than *S. serratum* and *P. elegans* (Bat et al., 1999).

Marine organisms, in general, accumulate contaminants from the environment and therefore have been extensively used in marine pollution monitoring programmes (Linde et al., 1998; de Mora et al., 2004).

Length and weight, physical and chemical status of water (Al-Yousuf et al., 1999) can play a role in the tissue accumulation of metals. The high level accumulation of metals in some fish species could be due to heavy rainfall during monsoon season which increase the metal content of water by washing down the agricultural waste.
Bioaccumulation is species-dependent and therefore feeding habits and life style can be strongly related to the sediment exposure (Chen and Chen, 1999). Other factors, such as sex and size may also influence metal bioaccumulation (Al-Yousuf et al., 1999; Canli and Atli, 2003).

These metals could find their way and remain for a very long time in seafoods, animals and plants and then passed on through food chain to man. Pollution by heavy metals occur largely from industrial processes of electroplating, metallurgy, mining, petroleum processing, large scale use of coal, batteries, fertilizers, dyes and pigments (Admoroti, 1996).

Fish also have been popular targets of heavy metal monitoring programs in marine environments because sampling, sample preparation and chemical analysis are usually simpler, more rapid and less expensive than alternative choices such as water and sediment (Rayment and Barry, 2000). According to Teodorovic et al. (2000) and Abdullah et al. (2008) heavy metals studies in aquatic biota give an idea that heavy metals in aquatic organisms could be more reliable water quality indicator than chemical analysis of water column and sediment. Heavy metals content in aquatic organisms has also been successfully used in evaluation in heavy metals input into European and American rivers. Accumulation of heavy metals, including lead in goat meat has been recorded 0.080 mg/kg in liver (Abou-Arab, 2001). Heavy metals including lead are found in various tissues of fish and shrimps (Vazquez et al., 2001). The high accumulation of lead in the liver which Gbem et al. (2001) also noted in their findings,
is related to the fact that liver plays a key role in accumulation and detoxification.

In general, studies on heavy metals can be important in two main aspects, public health point of view and the aquatic environment view point. Heavy metals are present in the aquatic environment where it can accumulate along the food chain. Moreover, small amounts of absorbed heavy metals are either stored in a metabolically available form for essential biochemical processes or detoxified into metabolically inert forms and held in the body either temporarily or permanently (Hashmi et al., 2002).

Besides, the high production of seafood through aquaculture provides a good source of high-quality protein. As 56 per cent of the world’s population obtains at least 20 per cent of their animal protein, the increasing trend of aquaculture production is a welcome sign considering to the global burden of diseases (Ezzati, 2002). Tiger prawn and tilapia fish until now have been the most important aquaculture, tolerance with the salinity ranges and meets the market needs. Aquaculture projects are often located on or near estuaries and coastal area because these water often provide ideal conditions for salt water aquaculture (Hashmi et al., 2002). Toxicity occurs when the rate of metal uptake into the body exceeds the combined rate of excretion and detoxification of metabolically available metal (Rainbow, 2002). A decrease in oxygen consumption and
ammonia-N excretion is observed in *Penaeus indicus* post larvae with increasing concentration in lead (Chinni *et al.*, 2002).

Heavy metals are non-biodegradable and can be accumulated by organisms to a level that affects their physiological states. Heavy metals absorbed to (or accumulated by) algae pass to zooplankton via ingestion, causing reduced survival and reproduction of the latter (Baratta *et al.*, 2002).

Heavy metal contamination has been reported in aquatic organisms (Adham *et al.*, 2002; Olojo *et al.*, 2005). These pollutants build up in the food chain and are responsible for adverse effects and death in the aquatic organisms (Farkas *et al.*, 2002).

The effects of lead on fish observed during this experiment showed that caution should be exercised in allowing lead into the aquatic environment. These effects included loss of balance, skin bleaching and weakness. A thick layer of mucus on the skin covered the dead fish and there were air bubbles on the water. There was reduced activity evidenced by vertical positioning and less mobility of the fish in solutions with less concentration of lead (3.2 and 5.6 mg/l), while no survivors were recorded for the highest concentration (10.0 mg/l) (Olaifa, 2003). The concentration of both Mn and Co in gonads, Cr in bronchial hearts and Zn in both gills and mantle increased linearly with animal weight (Nessim Ramzy and Rafik, 2003).
Overall, Pb and Cd concentrations were low in the water column of Makupa and Tudor creeks, with a few incidents of elevated levels in sediments and some fish species, especially during the rainy season. Makupa creek had the higher levels overall. The levels of Pb and Cd in most of the fish species analysed were generally, within acceptable limits by FAO standards (Benjamin M. Mwashote, 2003).

Shrimp farmers in Nigeria do not have access to an adequate supply of healthy, domesticated seed to stock their ponds because post larvae typically are produced from wild-caught brood stock. Wild brood stocks are often polluted with anthropogenic wastes in the lagoon (Moss, 2004). This accounts for decrease in availability and quality of wild brood stock.

Lead exposure and accumulation in children are especially serious in that lead is incorporated into the matrix of rapidly growing bone (Committee on the Environmental Health, 2005). Metal such as Zn and Cu are essential metals since they play an important role in biological systems, whereas Pb is non-essential metals, as they are toxic, even in traces. The essential metals can also produce toxic effects when the metal intake is excessively elevated (Turkmen et al., 2005).

The commercial and edible species have been investigated in order to check for those hazardous to human health. Metals can be taken up by fish from water, food, sediment and suspended particulate material (Agusa
et al., 2005). This study shows that the lowest concentrations of Cd and Pb were in the crayfish muscles in comparison to the exoskeleton, gills and digestive gland. Also, these results indicate that the analysis of Cd and Pb in different organs of *P. clarkia* might be a useful as a bioindicator for trace metals pollution in the freshwater system, due to their ability to rapidly accumulate and retain them in their tissues for long periods of time (El-Shaikh et al., 2005).

However, the presence heavy metal at high concentrations in water or sediment does not involved direct toxicological risk to fish, especially in the absence of significant bioaccumulation. It is known that bioaccumulation is to a large extent mediated by abiotic and biotic factors that influence metal uptake. Due to the deleterious effects of metals on aquatic ecosystems, it is necessary to monitor their bioaccumulation in key species, because this will give an indication of the temporal and spatial extent of the process, as well as an assessment of the potential impact on organism health (Fernandes et al., 2006).

Seasonal variations in the concentrations of four trace heavy metals (cadmium (Cd), copper (Cu), lead (Pb) and zinc (Zn)) were determined in *Donax trunculus* (Mollusca, Bivalvia) at two contaminated sites in the gulf of Annaba (East of Algeria): El Battah and Sidi Salem. The average concentrations of the metals exhibited the following order: Zn > Cu > Pb > Cd for the two sites. The statistical analysis revealed a significant effect
of seasons for all metals measured, the highest values being recorded in winter for Zn and in summer for the other metals (Beldi et al., 2006).

Lead accumulation was dose and tissue-dependent, with highest uptake by the gills. Gill concentrations of aquarium D fish averaged about 4-fold higher than in skeleton or skin and muscle. In vitro, lead (2.5 – 25 ppm) caused dose-dependent reductions in the ratio of reduced glutathione/oxidized glutathione (GSH-GSSG) in gills incubated in physiological buffer. These findings demonstrate that fathead minnow gills bind and accumulate waterborne lead rapidly and preferentially and raise the possibility that gill lipid peroxidation contributes to lead toxicity at low water hardness (Spokas et al., 2006).

Regardless of the tissue type, accumulation was maximum for Zn and minimum for Cd. A distinct metal accumulation capacity for each species was noteworthy. The low variability in metal content in the gills indicated its inertness towards bioaccumulation compared with muscle or liver. The high positive correlation between Zn and Pb in tissues is indicative of the anthropogenic impact in the estuary. Conclusions are that, in a weakly polluted area, the organ of choice for estimating bioaccumulation potential is the liver, whereas in a heavily polluted area, the tissue of choice is the gill (Maheswari Nair et al., 2006).

Comparative acute toxicity tests were carried out with three heavy metals viz., cadmium, copper and lead on giant prawn (Macrobrachium
rosenbergi) and tiger prawn (Penaeus monodon) post larvae. The metals showed toxicities in order Cu > Cd > Pb. The LC50 values recorded for M. rosenbergii were higher than the values recorded for P. monodon, hence the specific metals were more toxic to P. monodon than M. rosenbergii (Fafioye and Ogunsanwo, 2007).

During this investigation as the concentrations of test medium increased the accumulation pattern of lead also increased significantly among the three fish species viz., C. catla, L. rohita and C. mrigala. L. rohita showed significantly higher lead accumulation, followed by that of C. catla and C. mrigala (Javidi et al., 2007).

Aquaculture continues to grow more rapidly than all other animal food-producing sectors. Aquaculture sector has grown at an average rate of 8.8 per cent per year since 1970, compared with only 1.2 per cent for capture fisheries and 2.8 per cent for terrestrial farmed meat production over the same period worldwide. Furthermore, the growth of aquaculture production of fish, crustaceans and mollusks within developing countries has exceeded the corresponding growth of aquaculture production of fish, crustaceans and mollusks within developing countries has exceeded the corresponding growth in developed countries. Aquaculture production has been increasing at an average rate of 3.9 per cent within developed countries while at an average of 8.2 per cent in developing countries (Engle and Quagrainie, 2006; FAO, 2007).
The accumulation of Cu and Pb in *M. lanchesterii* was observed to be rapid and bioaccumulation increased with increasing metal concentration in the water and with exposure time. In the elimination study, Cu and Pb were found to be eliminate rapidly from *M. lanchesteri*. Metal accumulation and elimination patterns in this organisms are discussed (Shuhaimi-Othman *et al.*, 2006). High level of trace metals is found in liver, kidney, and muscles of Antarctic penguin *Pygoscelis adeliae* (Smichoowski *et al.*, 2006). Higher levels of metal was observed in stomach followed by gills and muscle. Several studies shown that various factors such as season (Dural *et al.*, 2006),

Seasonal variations in the concentrations of four trace heavy metals (cadmium (Cd), copper (Cu), lead (Pb) and zinc (Zn)) were determined in *Donax trunculus* (Mollusca, Bivalvia) at two contaminated sites in the gulf of Annaba (East of Algeria: El-Battah and Sidi Salem). The average concentrations of the metals exhibited the following order Zn > Cu > Pb > Cd for the two sites. The statistical analysis revealed a significant effect of seasons for all metals measured, the highest values being recorded in winter for Zn and in summer for the other metals (Beldi *et al.*, 2006).

However, non-significant differences for 96 h LC₅₀ tolerance limits towards zinc and lead were found. Among the three fish age groups, 30 day fish were more sensitive to metals toxicity, followed by that of 60 and 90 day respectively. The responses of three fish age groups and five metals were statistically significant. Among the three age groups, 90 day
fish showed significantly higher tolerance against all metals than that of 60 and 30 day fish (Sajid Abdullah and Muhammad Javed, 2006).

Aquaculture industry plays a vital role in the economy, market and nutritional requirement (Amaranemi, 2006; Hashmi et al., 2002). In global terms, some 99.8 per cent of cultured aquatic plants, 97.5 per cent of cyprinids, 87.4 per cent of penaeids and 93.4 per cent of oysters come from Asia and the Pacific. In the Asia and the Pacific region, aquaculture production from China, South Asia and most of Southeast Asia consists primarily of cyprinids, while production from the rest of East Asia consists of high-value marine fish. Top ten aquaculture producers in terms of growth in the worlds from 2002 to 2004 are Myanmar (45.1%), Vietnam (30.6%), Turkey (24.0%), Netherlands (20.4%), Republic of Korea (16.9%), Iran (16.5%), Egypt (11.9%), Chile (11.2%), Thailand (10.8%) and United States of America (10.4%), respectively (FAO, 2007).

The concentrations of lead (Pb) and zinc (Zn) were quantitatively determined in surface and sub-surface soils in Enyigba lead is relatively unavailable to plants when the soil pH is above 6.5, while availability of zinc decreases with increasing soil pH due to increased adsorptive capacity (Nweke et al., 2008).

Cray fish exposed to lead nitrate (500 μgL⁻¹) 3 days, the bioaccumulation shows higher lead concentration on gills and lowest in muscles in a following descending order gill > exoskeleton.
hepatopancreas > digestive tract.green gland>testis and ovary>muscles (Naghshbandi et al., 2007).

In many countries, industrial wastes, geochemical structure and mining of metals create a potential source of heavy metals pollution in the aquatic environment, due to their toxicity and accumulation behaviour. Under certain environmental conditions, heavy metals might accumulate up to a toxic concentration and cause ecological damage (Sivaperumal et al., 2007).

For freshwater field crab, Spiralothelphusa hydrodroma effect of copper on the quantitative study of nutritive value viz., protein, carbohydrate and lipid in ovary, spermatheca, hepatopancreas, muscle, gills, haemolymph, brain, thoracic ganglia and eyestalk was observed (Senthilkumar et al., 2001).

The 96 h LC50 and lethal concentration of lead varied significantly among three fish species. C. catla were found more sensitive to lead concentration followed by that of L. rohita and C. mrigala. It was concluded that from the major carps. C. catla is more susceptible to lead toxicity, while the L. rohita has the higher tendency to accumulate metallic ions (Javidi et al., 2007).

The toxicity of lead at high levels of exposure is well known but of a major concern is the possibility that continual exposure to relatively low
levels of these heavy metals through the consumption of these fauna and flora may entail adverse health effects (Gbaruko and Friday, 2007). As the metal concentration of test medium increased, the accumulation pattern in fish body also increased significantly (Javidi et al., 2007).

Bioaccumulation factor (BAF) was in the sequence of Cd > Fe > Cu > Zn. For fish exposed for 60 d, bioaccumulation exhibited increasing metal levels in liver followed by muscle tissues and gills. These results will support ecologist to use *P. waltoni* as a bioindicator to metal pollution in the future (Bu-Olayan and Thomas, 2008).

Of all the metals, the maximum decrease of oxygen consumption of fish was recorded under lead nitrate toxicity. The magnitude of toxicity of metals to the fish, *Tilapia mossambica* appears to be in the order of lead nitrate > Zinc Sulphate > Cadmium Carbonate > Cobalt Carbonate > Copper Sulphate (Shereena and Logaswamy, 2008). The study was conducted to determined the concentration of heavy metals (copper, lead, zinc, cadmium and mercury) in seawater as well as in three different types of fishes (*Pagellus acarne, Sarpa salpa* and *Liza saliens*) with emphasis on their relation with some biomarkers. Among studied fish species, Pb in muscles and liver tissues were obviously higher in *Pagellus acarne* than in the two other fish species (Metwally and Foud, 2008).

The effect of both chemicals on fish chromosomes and mitotic indices was investigated and the results revealed that gill cells of the
treated fish by copper sulfate and lead acetate displayed lower mitotic activity than that of the control group, both pollutants were found to be positive inducer of macro-DNA damage which represented by different types of aberrations, for example chromatid deletions, chromatid breaks, gaps, fragments, stickness, translocations, ring chromosomes and centromeric attenuation. We also observed that chromatid deletion, stickness and fragments were more frequent than other chromosomal aberrations (Mohamed et al., 2008).

Heavy metal concentration in the tissues tend to vary significantly among season and monsoon period showed particularly high metal concentration compared to pre-monsoon and post-monsoon. Muscle tissues and gill showed higher concentration of zinc, chromium, copper, cadmium and lead than gonads and skin (P < 0.05). Highest concentration of zinc, chromium, copper, cadmium and lead were detected in gill tissues (P < 0.05). Lowest concentration observed in gonads of fish sampled from upper course of the River Ganga. Further, metal accumulation showed high degree of species specificity, where the order of accumulation of heavy metals was zinc > copper > chromium > cadmium > lead (Bhattacharya et al., 2008).

Concentrations of Cd, Cu and Zn were found to be higher in tiger prawns (Penaeus monodon) in Bandar; whilst only Fe, Mn and Ni in tiger prawns (Penaeus monodon) was found to be higher in Jugra. Concentrations of Cu, Zn, Cr, Fe, Mn and Ni were found to be higher in Jugra whereas those of Pb and Cd were higher in Bandar for tilapia fish (Oreochromis
Concentrations of heavy metals studied were found to be lower than the recommended maximum level allowed in from Bandar and Jugra aquaculture pond were safe for human consumption (Mokhtar, 2009).

The accumulation of the heavy metals in the shrimps will reach the human beings and will result in ‘biomagnifications’ (Soundarapandian et al., 2010). The life history parameters, such as time (hour) the rotifer released neonates and life span, were evaluated. These results showed that the population growth of *B. plicatilis* decreased with increasing concentrations of cadmium chloride (Chinnasamy Arulvasu et al., 2010). The increased concentrations of Pb(II), Cu(II) and Zn(II) in the mangrove environment can significantly affect growth and impair normal reproduction of *Halophytophthora* species (Leano and Pang, 2009).

Among the various organs, bioaccumulation was higher in hepatopancreas and lower in gills. The higher accumulation of the heavy metal, mercury in the hepatopancreas may be due to uptake of nutrients and it is the storage organ of inorganic reserves. Large amount of accumulation of heavy metal in the tissues of shrimp in the industrially polluted area clearly revealed the effect of heavy metal, mercury which impacts the inhabitants, especially shrimps (Soundarapandian et al., 2010).

Lead was not detected in prawn. The mean concentration of copper, cadmium and zinc in prawn was observed to be within the range NAFDAC standard for water and aquatic foods (Olowu et al., 2010).
Among the various organs, bioaccumulation was higher in hepatopancreas and lower in gills of *Penaeus monodon*. The higher accumulation of the heavy metal, mercury in the hepatopancreas may be due to uptake of nutrients and it is the storage organ of inorganic reserves. Large amount of accumulation of heavy metal in the tissues of shrimp in the industrially polluted area clearly revealed the effect of heavy metal, mercury which impacts the inhabitants, especially shrimps, in the Uppanar estuary. The accumulation of the heavy metals in the shrimps will reach the human beings and will result in biomagnifications. This hazardous situation may be prevented by treating the effluents properly before being let off into the Uppanar estuary (Soundarapandian et al., 2010).

2.5. DEPURATION

The organisms tend to accumulate elements / metals whether they are essential or non-essential, until reaching a threshold concentration / level after which they have to eliminate or face the damages and the consequent difficulties; this has necessitated the concurrent development of a mechanism for the elimination of these elements by each of the various groups of organisms (Bryan, 1968; Cearley and Coleman, 1974).

Approximately 75% of inorganic lead absorbed into the body is excreted in urine and less than 25% is excreted in feces. Lead is also excreted in breast milk and therefore, available for intake by infants (Jensen, 1983; EPA, 1989a).
Lead that is not retained by the body is excreted unchanged in urine (65-75%) and in bile (25-30%). The urinary lead excretion rate depends on renal blood flow and glomerular filtration rate. Factors that affect either of these 2 functions affect blood lead concentrations. Small amounts of lead may be found in sweat and milk (Rania Habal, 2008). Approximately 75% of inorganic lead absorbed into the body is excreted in urine and less than 25% is excreted in feces. Lead is also excreted in breast milk and therefore, available for intake by infants (Jensen, 1983; EPA, 1989a).

Some Decapod crustaceans possess an innate ability to regulate the internal concentration of essential but potentially toxic metals within a constant limits presumably to meet their metabolic demands (Rainbow, 1985). Further, White and Rainbow (1985) have also opined that the marine prawns are believed to regulate tissue Zn concentrations within levels which approximate the theoretically estimated Zn requirements for enzymes and possibly stabilizing the respiratory pigment, haemocyanin molecule. On the contrary, tissue levels of non essential metals such as Cd, Pb and Ni are apparently not regulated by marine decapods (Rainbow, 1985; Depledge and Rainbow, 1990).

In vertebrates, elimination can proceed by several routes, including transport across the integuments or respiratory surfaces, secretion in gall bladder and excretion from kidney in urine. The control of metal kinetics in eukaryotic cells involves the integration of several complex mechanisms.
In mammalian systems, membrane transport, induction of intracellular metal binding ligands and lysosomal compartmentalization contribute to the intracellular regulation of both essential (Zn and Cu) and toxic (Cd) metals. Research in this field focused on the role of a family of inducible cystein rich metal binding proteins known as metallothioneins (MTs) in the kinetic processes. The occurrence of MTs like protein has been demonstrated in many species including fishes of both teleost (Kito et al., 1982) and elasmobranch (Hildago et al., 1985) which are believed to perform protective role against the toxic effects of these metals by sequestering and reducing the amount of free metal in tissues, thereby reducing the potential toxicity (Friberg et al., 1974; Bouquegneau et al., 1975; Cherian and Goyer, 1978).

Tissue distribution of MTs in fish primarily includes the gill, liver, kidney and intestine and small amounts occur in blood (Noel-Lambot et al., 1978; Reichert et al., 1979; Kito et al., 1982b). MTs have been found in the hepatopancreas of the crustacean, Cancer magister and Acetes sibogae, in the digestive gland of chiton, Cryptochiton stelleri (Olafson et al., 1979) and in the soft parts of the mussel, Mytilus edulis (Frankenne et al., 1980).

Yet another possible detoxification mechanism is the sequestering of metals in vesicles, lysosomes and other membrane bound structures, as granules and intracellular insoluble metal precipitates and therefore
isolated from chemical activity in the cells (Mason and Nott, 1981; Brown, 1982; George, 1982).

Surprisingly, very few attempts have been made to distinguish between the various routes of elimination in aquatic organisms, perhaps due to experimental difficulties. At least a few chemicals are known to proceed by each of the principal routes in certain species (Rand and Petrocelli, 1985). With regard to metals, Nakatani (1966) has shown that rainbow trout, *Salmo gairdner* removes the ingested $^{65}$Zn by the way of gills; Bryan (1967), has observed quicker Zn-turnover in the gills of freshwater crayfish, *Austropotamobias pallipes pallipes* and in the subsequent year (1968), he has also recorded the excretion of metals like Hg, Cu, Cr and Mn in the urine of some crustaceans; Narayanan (1989) has traced the elimination of Zn via gills in *Scylla serrata*; Mathiessen and Brafield (1993), have observed the quick Zn-turnover in sticklebacks by the secretary activity of chloride cells of gills; Bertine and Goldberg (1973) and Ranfro *et al.* (1975) have opined that larger proportions of the body burden of trace metals such as Zn can be lost as moulting in crustaceans; Varanasi and Markey (1978) have found the excretion of Cd and Pb via body mucus in *Coho salmon (Oncorhynchus kisutch)*.

Calamari *et al.* (1982) and Davis and White (1985) have also reported a marked drop in metal concentration from the tissues after the exposure has been terminated. These observations are in line with the findings of Devi (1990) and Rahim (1992).
The concentrations of lead in these tissues declined on transfer of fish to lead-free water. The recovery of fish was faster for those placed in lower concentrations of lead than those that had been placed in higher concentrations. The effects of heavy metals such as lead on the environment is usually highlighted and addressed in respect to their effects on man (Olaifa, 2003).

Lead that is not retained by the body is excreted unchanged in urine (65-75%) and in bile (25-30%). The urinary lead excretion rate depends on renal blood flow and glomerular filtration rate. Factors that affect either of these 2 functions affect blood lead concentrations. Small amounts of lead may be found in sweat and milk (Rania Habai, 2008).

Lead was not detected in prawn. The mean concentration of copper, cadmium and zinc in prawn was observed to be within the range NAFDAC standard for water and aquatic foods while crabs have higher mean concentration of heavy metals with the exception of zinc and copper are within the limit (Olowu et al., 2010).

2.6. HISTOPATHOLOGY

Although major advances have been made in recent years, the histology and histopathology of fish and aquatic invertebrates are still infant sciences compared to their counterparts in mammals. Examination of tissues from fish and other aquatic organisms after death may serve to identify the cause of death and possibly the causative agent. This
information along with physiological and biochemical data may provide a more complete and accurate description of the activity of a chemical agent. Skidmore (1970) opines that the respiratory handicap has been imposed by lifting up of the epithelium (as welling) must out weight any protective effect against pollutant uptake in the later stages of acute poisoning, especially when asphyxia is the immediate cause of death. The metals have been reported to cause damage to fish organs (Skidmore and Towell, 1972, Wong et al., 1977; Noel-Lambot, et al., 1978; Kumar and Pant, 1981).

The respiratory system of fish provides the most extensive interface with the aquatic environment which in turn is covered only by a thin epithelium (Ellen, 1975). Thus fish gills are constantly being exposed to various external factors. A wide variety of structural changes in fish gills have been reported as a consequence of an exposure of fish to pollutants (Mallett, 1985; Evans, 1987). Hence the gills are invariably subjected to histological study in any work related to the assessment of the effect of pollutants on aquatic organisms gill epithelium.

Narayanan et al. (1987) have demonstrated that the sulphur oins of the pollutant are eliminated in the form of sulphated mucopolysaccharides in the mucus which has been secreted by the gill of Lepidocephalichthyes thermalis. Further, Othuman (1994) has observed the secretion of neutral and acid as well as sulphated mucopolysaccharides in mucus of
O. mossambicus when exposed to distillery effluent. Thus it can be safely assumed that the chemicals entering into the fish are eliminated by mucus.

The vertebrate liver has a remarkable role to play in the physiology and the well being of the organism. Apart from partaking in digestion, it serves as a storage house for glycogen while performing the most other anabolic functions and detoxification centre for most of the pollutants / toxicants entering the systems. Expectedly, liver often shows conspicuous damage by toxicants. Hence, the liver has to be studied for histological changes while biomonitoring the environment (Crandall and Goodnight, 1962; Mount and Stephen, 1967; Burton et al., 1970; Jackim, 1973; Wong et al., 1977; Dalela et al. 1978; Zaba and Horis, 1978; Natarajan, 1979; Kumar and Pant, 1981; Singh and Sivalingam, 1982; Wani and Latey, 1983; Kothari and Suneeta, 1990).

Several lines of evidence implicate high-level lead exposure as a cause of many of pathological conditions such as renal insufficiency, gout and hypertension (Wedeen et al., 1979; Batuman, 1993; Ding et al., 2001). Oxidative stress has also been implicated in specific organs with lead-associated injury, including liver, kidney and brain tissue (Kostial et al., 1999; Patra et al., 2001). Although different considerations were raised to explain the pathogenesis of lead toxicity, several studies suggested the primary involvement of the increased production of reactive oxygen species (ROS) observed in lead-exposed animals (Ding et al., 2001). The observed toxicity induced by lead acetate in rats was similar to
those previously reported by Kostial et al. (1999); Patra et al. (2001) and Shalan et al. (2005).

The deficiency of Coenzyme Q has been reported in many diseases such as diabetes (Kucharska et al., 2000), heart failure, angina and hypertension (Mongthuong et al., 2001). Therefore, our study has been conducted to evaluate the toxic effect of lead injection on the coenzyme Q level in rat tissues. In addition, lead-induced oxidative stress has been identified as the primary contributory agent in the pathogenesis of lead poisoning (Ding et al., 2001; Hsu and Guo, 2002).

Following exposure the gills exhibited rapid alterations that include detachment and lifting of the epithelial linings from the surface of the gill filament (primary, PL) and respiratory (secondary, SL) lamellae. This led to extensive haemorrhage from the gills. Thus the quantity of blood flowing across the gills decreased substantially. Simultaneously, uncontrolled regeneration of the PL and SL occurred, leading to extensive hyperplasia of the epithelial cells lining the PL and SL. Consequently, the gill filaments appeared as a cylindrical solid mass of cells with very little or almost no free surface left on the SL for gaseous exchange. The goblet mucous cells also exhibited periodic fluctuations in their density and staining behaviour. The chloride cells showed periodic fluctuation in their number at different stages of exposure. The density of the chloride cells is inversely proportional to the thickness of the epithelial lining of the PL and SL. Due to prolonged exposure, the neighbouring SL fused together
and the entire gills appeared as solid mass of undifferentiated cells. Subsequently, the ladder-like arrangement of the pillar cells-blooc capillaries of the gills also collapsed, causing asphyxiation and the death of the fish (Parashar and Banerjee, 2002).

According to Ramirez-Perez et al. (2004), when the mercury concentration in the medium was enhanced from 0 to 0.005 mg/l, there was a considerable reduction in both average lifespan and life expectancy of B. calyciflorus. The sublethal concentrations (0.006 and 0.008 mg/l) of lead for a period of three weeks maintained. The liver and gill of fish were removed every 9 days for histological examination. The results showed that the one hundred and sixty (160) fingerlings of Clarias gariepinus were exposed to continuous exposure degree of distortion of the gills and liver was proportional to the exposure periods and concentration of the metals was found to be dose and time dependent (Olojo et al., 2005).

It was found that the metals were accumulated in different tissues of both fish by various levels, where, the non-edible parts accumulated more metals than the edible muscles. Zn, Cu, Pb and Cd concentrations in the fish muscles were below the maximum permissible limit, however, Fe in the muscles exceeded the permissible limit. Several histopathological alterations, including vacuolar degeneration with focal areas of necrosis in liver, proliferation in the epithelium of gill filaments and fusion of secondary lamellae, severe degenerative and necrotic changes in the intestinal mucosa and seminiferous tubules, degeneration and atrophy in
cardiac muscle fibers and degeneration in muscle bundles were observed in the studied tissues of both fish as a result of the accumulated metals (Mohamed, 2008).

The ultrastructural alterations are suggestive of the operation of compensatory mechanisms within the test prawns to enable it to tolerate Hg toxicity. However, these alterations would have an impact on the cellular integrity of the gills and hepatopancreas and such alterations can be taken as 'biomarkers' for assessing Hg pollution in the aquatic environment (Yamuna et al., 2009).

Histological alterations were observed in the muscles of both fish, including degeneration in muscles bundles with focal areas of necrosis, atrophy of muscle bundles and edema between muscle bundles. The liver showed vacuolar degeneration in the hepatocytes, focal areas of necrosis and fibrosis, aggregations of inflammatory cells between the hepatocytes, dilation and congestion in blood sinusoids and thrombosis formation in the central veins. In the gills, the pathological alterations included proliferative, degenerative and necrotic changes in the epithelium of gill filaments and secondary lamellae, edema in secondary lamellae, dilation and congestion in blood vessels of gills filaments and mucous cells proliferation. The kidney showed vacuolar degeneration in the epithelium of renal tubules, focal areas of necrosis, haemorrhage and haemosiderin between the renal tubules and edema in Bowman’s capsules with atrophy in the glomeruli (Mohamed, 2009).
Lead is a persistent and common environmental contaminant. Like other commonly found persistent toxic metals e.g. mercury, lead damages cellular material and alters cellular genetics (Abdullah, 2008). Concentration of Zn, Cu and Pb were determined in eight commercially valuable fish species, *Selaroides leptolepis, Euhynus affinis, Parastromateus niger, Lutjanius malabaricus, Epinephelus sexfasciatus, Rastrelliger kanagurta, Nemipterus japonicas* and *Megalaspis cordyla* from Pahang coastal water. The estimated values of all metals in muscles of fish in the study were below the established values. Therefore, it can be concluded that the fish from Pahang coastal water are comparatively clean and do not constitute a risk for human health (Kamaruzzaman et al., 2010).