REVIEW
OF
LITERATURE

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Physico-chemical parameters of estuary:

In the marine hydrosphere estuarine ecosystems are more fragile, highly dynamic and complex than any other coastal ecosystem (Gogate, 1960; Lonkar, 1972).

In natural water interesting spatial and temporal thermal changes are brought about by solar energy and atmospheric temperature. Discharge of heated effluents also brings about thermal change in natural waters (Thermal pollution). Temperature is an important factor for its effects on chemical and biological reactions in water. Temperature increase accelerates chemical reaction, reduces solubility of gases, elevates metabolic activities of organisms, and amplifies taste and odour (Saxena, 1998).

Many biological processes that occur in marine and estuarine waters are sensitive to temperature changes. Water salinity and temperature are the most important physical factors affecting the activities of marine and estuarine organisms, marine and estuarine waters. They affect every physical property of seawater (Cox, 1965; Houston, 1982). The processes of the chemical, petrochemical, and pulp and paper industries; municipal sewage and thermal generating stations change these physical factors (Swiss, 1984). Other disturbances of aquatic temperatures may be related to the physical alteration of a water body. Such as river diversions, water withdrawals from coastal areas, retaining walls in estuaries, large jetties, breakwaters, and causeways in coastal areas may significantly alter water temperatures (Greenberg, 1984).

Temperature affects many chemical and biological processes. Chemical equilibrium constants, solubilities and the rates of chemical reactions are temperature-dependent (Whitehouse, 1984). Most marine and estuarine organisms are poikilotherms. As a result, biological processes, such as photosynthetic and respiration rates, spawning, uptake of toxic substances and behavioural patterns of organisms, are all responsive to changes in temperature (Strickland, 1965; Houston, 1982; Aiken and Waddy, 1990).

A comprehensive review of the effects of water temperature on marine and brackish water animals was conducted by Kinne in1963. The results of this review indicated that the biological processes may be greatly affected by water temperature fluctuations. Most of the marine and estuarine species, or populations within species, have characteristic tolerable temperature ranges (Canadian Environmental Quality Guidelines Canadian Council of Ministers of the Environment, 1999).

The extremely low water temperatures affect integration of nervous and metabolic processes, rates of energy liberation, changes in water and mineral balance, increase in osmoconcentration in aquatic organisms (Kinne, 1963). Many species of marine fish have body fluids with lower osmotic pressure than seawater, causing such species to freeze at temperatures above the freezing point of seawater. Most species of marine fish avoid freezing by either avoiding ice-laden seawater and/or by increasing the osmotic concentration of their blood (DeVries, 1971).

Extremely high temperatures affect supply of oxygen, failures in process integration, desiccation among the inter tidal organisms, enzyme inactivation, and change in lipid state, increase in protoplasmic viscosity, and increase in cell membrane permeability, protein denaturation, and release of toxic substances from damaged cells. Death can result from exposure to either extremely high or extremely low water temperatures. Water temperature changes that can not be lethal but can produce a wide variety of significant sub lethal effects. The temperature changes can significantly affect the rate of respiration, susceptibility to disease, osmoregulation, uptake of pollutants, susceptibility to the toxic effects of pollutants; various behavioural patterns, including physical activity, reproduction, feeding, growth, distribution, intra and inter specific competition, predator - prey relationships, community composition and parasite host relationships; and many other biological processes (Kinne ,1963).

Temperature of water plays a limiting role on the feeding and recruitment success of fish and crustacean larvae. Paul and Nunes (1983) reported that northern pink shrimp larvae (*Pandalus borealis*) that hatched during a warm year (at 6 °C) required 63 % more calories to meet their metabolic requirements.

Increasing water temperature generally causes increases in the respiratory rates of aquatic animals. Beyond a given temperature, thermal stress is induced (Kinne, 1963; Paul and Nunes, 1983; Paul, 1986; Paul, *et. al.*, 1988). As respiratory rates increases, the organisms' metabolic energy need also increases. This relationship has important implications for the overall productivity and survival of marine and estuarine organisms that thrive in habitats with varying temperatures (Paul, 1986; Paul, *et. al.*, 1988, 1990).

Water temperature may also affect the reproductive capacity of marine organisms (Tanasichuk and Ware, 1987). All the relationships demonstrate that even very small change in the extent and timing of temperature in coastal waters can significantly alter the biological processes.

The sensitivity of aquatic organisms to toxic substances increases with increasing water temperature (Cairns, *et. al.,* 1975). The interactions between temperature and toxicity are very complex. According to Voyer and Modica, (1990) insufficient data exist to permit the development of relationships between water temperature or salinity and the toxicity of heavy metals in marine water. The available evidence indicates that the survival, bioconcentration and sub lethal effects of pollutants on estuarine organisms are often related to ambient salinity and temperature conditions, as water temperature increases, the rate of metabolic processes increases, resulting in enhanced uptake and toxicity of metals to marine and estuarine organisms (Phillips, 1976; Waldichuk, 1985; McLusky, *et. al.,* 1986; Voyer and Modica, 1990).

Materials hotness is measured as temperature. Water temperature measurement is essential because it affects the chemistry and biochemical reactions in the organisms. It is important in determination of pH, saturation level of gases of water and conductivity (Trivedy, *et. al.,* 1987). Cairn, *et. al.,* (1975) Studied temperature influence on chemical toxicity to aquatic organism. Sarkar (1991) reported the effects of temperature on eggs, fry and fingerlings of *Labio rohita* exposed to urea. Chauhan and Saxena, (1992) observed that toxicity of heavy metals increases as temperature increases.

Mortality patterns of aquatic organisms induced by pollutants are measurably related to various physico-chemical parameters such as temperature, pH, salinity, dissolved oxygen and hardness of medium but temperature has marked effect as toxicity of substances (Doudoroff and Katz, 1953).

The pH is the negative logarithm (base 10) of the chemical activity of the hydrogen ion in solution. The pH scale indicates a neutral solution at pH 7.0, an acidic solution below 7.0, and an alkaline (basic) solution above 7.0. A unit change in pH corresponds to a tenfold change in the hydrogen ion concentration; so the small change in pH can significantly change the chemistry of marine and estuarine waters. Exposure of air, temperature and disposal of industrial wastes etc brings changes in pH (Saxena, 1998). High pH is unfavorable to aquatic organisms (Robbert, *et. al.*, 1940; Das, 1978). Paterson, *et. al.*, (1980); Kane, *et. al.*, (1987) observed that low pH has adversely effect on early life stages of fish.

The pH of marine waters is usually quite stable between 7.5 and 8.5 worldwide and is similar to the estuarine waters because of the buffering capacity provided by the abundance of strong basic cations as sodium, potassium, and calcium and weak acid anions as carbonates and borates.

pH values of the surface waters remain elevated because of solar radiation. The solar radiation affects pH in two ways as it promotes photosynthesis and increases surface temperature. Because of which there is decrease in the amount of free carbonic acid and consequently rise in the pH (Skirrow, 1965). The fluctuations in pH due to anthropogenic influences in aquatic environments are largely due to the effects of industrial activities. NOX and SO_2 emissions from industries and vehicles cause acid precipitation, which depresses the pH of surface waters (Knutzen, 1981; Kaufmann, *et al.*, 1992). Direct inputs of acid, through acid mine drainage and some industrial waste leachates can change the pH of aquatic environments (McNeely, *et. al.*, 1979).

The marine and estuarine organisms have adversely affected by pH fluctuations, some of these effects are physiological. Decrease in pH was correlated with a reduction in carapace weight, increased magnesium content and a slight decrease in strontium content of the marine prawn *Penaeus monodon* (Wickins, 1984). In the marine octopus *Octopus dofleini*, a pH below 7.2 was correlated with a decrease in maximum oxygen saturation of hemocyanin, a blood oxygen transporter (Miller and Magnum, 1988). The reduced growth, weight loss, reduced shell size, shell dissolution, and suppressed feeding occurred in four species of bivalves at pH below 7 (Bamber, 1987). Cell aggregation of sea urchin embryos (*Hemicentrotus pulcherrimus*) was found to be affected by low pH and was obliterated at a pH lower than 4.0 (Tonegawa, *et. al.*, 1990).

In the marine environment, changes in pH can also significantly affect the chemical forms and toxicity of other substances (Miller, *et. al.*, 1990). The speciation of metals and the solubility of some organic chemicals are also strongly influenced by pH (Bengtsson, 1978; Kaiser and Valdmanis, 1982; Drever, 1988).

Dissolved oxygen is an important factor for estuarine environment. It largely fluctuates with the temperature, salinity and rainfall. Oxygen solubility decreases with rise in temperature. Organic pollution, which produces a biological oxygen demand intensified increase in temperature. Therefore temperature can intensify chemical toxicity. When dissolved oxygen in water is reduced, many substances are more toxic (Cairns, *et. al.,* 1975).

Dissolved oxygen is a very important parameter of water quality. It is often referred as oxygen dissolved in the water. There are two main sources of dissolved oxygen in water- i) Diffusion from air, and ii) Photosynthetic activity within the water.

Diffusion of oxygen from air to water is a physical phenomenon which depends upon solubility of oxygen which in turn is influenced by factors like water movements, temperature, salinity etc. Photosynthetic activity is a biological phenomenon carried out mainly by phytoplankton in water and depends upon population of autotrophs, light and available gases etc. Non polluted surface waters are normally saturated with dissolved oxygen. Organic waste, which is oxygen demanding pollutants, causes rapid depletion of dissolved oxygen from water.

Inorganic substances like hydrogen sulfide, ammonia, nitrites, ferrous iron etc also responsible in depletion of dissolved oxygen. Low dissolved oxygen may prove lethal for many organisms. Saxena, (1998); Mishra and Saksena, (1989) Studied the physicochemical parameters of the industrial effluents from textile mill in Birla nagar, Gwalior. They found that range of dissolved oxygen was 1.0 - 4.0 which is low. Similar observation was recorded by Subramaniam, *et. al.,* (1988); Pandey, *et. al.,* (1993); Chauhan, *et. al.,* (1993); Jesudas and Akila, (1995); Thorat and Wagh, (1998).

Dissolved oxygen (DO) is the amount of oxygen, usually measured in milligrams or milliliters, dissolved in one liter of water. The solubility of oxygen in water is inversely correlated with temperature and salinity (APHA, 1992). Oxygen is essential for the respiration of almost all life, including most marine and estuarine organisms. The amount of oxygen available for aquatic life depends on a number of factors that affect the solubility of oxygen in seawater. These factors include salinity, temperature, atmospheric exchange, barometric pressure, currents, upwelling, tides, ice cover, and biological processes. Oxygen-consuming chemical processes may also be important factors in certain areas (Colinvaux, 1973; Davis, 1975).

The aquatic body has greatest DO depletion, where there is restricted circulation and abundant supply of organic matter from the natural sources, sewage, food related industries, agricultural runoff, pulp mills, or other human activities (Topping, 1976; Colodey, *et. al.*, 1990).

Reduced oxygen levels have been shown to cause lethal and sub lethal effects on the physiology and behaviour of variety of organisms, mainly in fish. Hughes and Ballintijn, (1968) observed an increase in the ventilatory muscle activity of dragonets in oxygen-depleted marine water. Sockeye salmon *(Oncorhynchus nerka)* showed signs of elevated blood and buccal pressure and an increased breathing rate (Randall and Smith, 1967). Physiological studies indicate that reduced DO levels restrict the ability of fish to maximize metabolic processes (Birtwell, 1989). Consequently, the growth rates of fish are affected by reduced DO levels, reductions in the growth rate of salmon (USEPA, 1986).

Oxygen dependency of invertebrate is determined by numerous factors, including the existence of a circulatory system, diffusion distances, temperature, degree of locomotor activity, ability to regulate external respiration, and the existence of respiratory pigments (Davis, 1975b). A study on the fluctuations of the benthic community of Petpeswick Inlet, Nova Scotia, illustrates the impact of anthropogenically or naturally altered DO regimes. Dissolved oxygen has been observed to modify the susceptibility of aquatic organisms to environmental stresses such as the presence of toxic substances (Alabaster and Lloyd, 1980; Hutcheson, *et. al.*, 1985; McLeay and Associates Ltd., 1987). The toxicity of kraft pulp mill effluents and hypoxia may act synergistically on aquatic organisms (Birtwell, 1989).

Free carbon-dioxide in the water accumulates due to microbial activity and respiration of organisms. This imparts the acidity of the water because of formation of carbonic acid. Free carbon-dioxide is determined by titrating sample using a strong alkali of pH 8.3 (Trivedy and Goel, 1987).

Many aquatic invertebrate animals are intolerant of even relatively modest increase in the ambient concentration of carbon dioxide (CO_2). Carbon dioxide intolerance is based on the chemical reaction of CO_2 with water to form Carbonic Acid (H_2CO_3). In aquatic animals carbon dioxide from aerobic metabolism is diffused as dissolved free CO_2 or HCO_3^- down concentration gradients across the gills and/or general body epitheliums into the surrounding medium.

Mishra and Saksena (1989) studied free carbon-dioxide from textile mill effluent. Chauhan, *et. al.,* (1993) studied free carbon-dioxide from tannery effluent. Similarly Pandey, *et. al.,* (1993); Jesudas and Akila, (1995) studied free carbon-dioxide from sugar factory effluent and industrially polluted lake respectively.

The salinity of water is an expression of the concentration of total dissolved solids. The salinity of the oceans of the world is ranges from 32 – 38 % with an average of 35 ‰ (Kalle, 1971). Salinity levels in coastal waters show variations because of river inputs, influx of groundwater, freshwater runoff with rainfall, variable evaporation rates, and tidal and ocean currents (Harrison, *et. al.,* 1983). Many semi-enclosed areas and estuaries show extreme salinity variability in time and space (Krauel, 1975; Thomson, 1981; Dickie and Trites, 1983; Prinsenberg, 1986; Smith, 1989; Hopky, *et. al.,* 1990). Saline water is a composite of many different solutes and as such, its density is variable and greater than that of freshwater (Moore, 1966).

The salinity of coastal water affects several important physical and chemical properties of water, such as the freezing point, specific gravity and osmotic pressure; this can have biological significance. The relationship appears to exist, in some cases, between adverse effects on marine and estuarine organisms and the length of exposure to extreme salinities (Voyer and Modica, 1990; Voyer and McGovern, 1991).

In the distribution of aquatic organisms salinity is a limiting factor. The freshwater diversions, large volume of industrial and municipal effluent discharges and barriers to existing flow patterns by anthropogenic contribution can change the salinity of estuaries and other coastal water bodies and affect the biota (Thomson, 1981; Dickie and Trites, 1983). In many coastal areas, salinity fluctuations limit the distribution of organisms that require stable salinities. However, natural diurnal changes in salinity, which occur in many coastal waters, are vital to the biota (Davenport, *et. al.,* 1975; Rijstenbil, *et. al.,* 1989; Voyer, *et. al.,* 1989).The salinity of coastal waters is also vital because of the physical and chemical interactions that may occur with stress factors and toxicants. The solubility, uptake, and bioavailability of certain compounds in aqueous media, these are the physical interactions can be affected by salinity (Whitehouse, 1984).The chemical interactions such as the modification of the chemical speciation of trace metals are also affected by salinity (Hong and Kester, 1985). In addition, salinity affects many responses of organisms such as survival, reproduction, behaviour, and other sub lethal effects to a variety of substances (Sprague, 1985; De Lisle and Roberts, 1988).

In water phosphorus occurs in both the forms as organic and inorganic state. Over 85 percent of total phosphorus is usually present in organic form, as bound in the organic matter. Inorganic phosphorous as orthophosphate plays a dynamic role in aquatic ecosystem. Under oxidizing conditions it get precipitated and lost to the sediments which result in depletion of phosphorous in water. Under reducing condition some phosphorous returns to water in soluble form. Even though it is present in low concentration, it is one of the most important nutrients limiting growth of autotrophs and so biological productivity of the system. Increased algal growth as a bloom occurs because of high phosphorous content. The enrichment of phosphorous leads to the process of eutrophication. Industrial effluents, Domestic sewage, Detergents are main sources of phosphorous in water.

Mishra and Saksena, (1988) reported that phosphate content of the textile mill effluent was within limit of standards. Pandey, *et. al.,* (1993) studied that phosphate content of Hussain Sagar, industrially polluted lake in Hyderabad. Jesudas and Akila, (1995) reported that the difference in phosphate content of raw and treated effluent of the sugar factory.

Only Nitrate (NO_3^-) is one of several forms of nitrogen occurring in surface waters. Nitrate tends to be the dominant form of nitrogen in waters. With the help of bacteria, it can be converted naturally in soils and waters from other forms of nitrogen or it can be released directly to surface waters from residential, industrial, and agricultural sources. Extensively nitrate is used in the production of fertilizers. It is also used in a variety of industrial applications, such as oxidizing agents in explosives, matches and firework. Nitrate is also used in glass making, photography, textile dyes, food preservatives, and as a raw material for manufacturing nitric acid.

Nitrogen undergoes a complex cycle through the terrestrial and aquatic environments. Several factors, such as pH, amount of available oxygen, and the types of biological communities present in the water are the determinant factors of form of nitrogen in surface water. Different forms of nitrogen released to surface waters may be transformed by bacteria to nitrate. Nitrate can be directly added to surface waters through surface water runoff, atmospheric deposition, or through the seepage of groundwater to streams and lakes. More nitrogen can be released to surface waters from diffused sources like atmospheric deposition and agricultural and domestic runoff and through the point sources like municipal wastewater effluents or industrial discharges.

Nitrate is highly mobile in the environment, it is an essential plant nutrient, and high levels of nitrate in lakes and coastal areas therefore contribute for the excessive plant and algal growth. This may result in indirect toxic effects to other aquatic organisms. The algal blooms can reduce oxygen levels in the water, causing stress on aquatic animals, and some algae can produce toxins that may be hazardous to other aquatic organisms and people. Excess level of nitrate is also directly harmful to aquatic animals. Aquatic invertebrates and fish exposed to nitrate may be smaller in size, slower maturity period, or have lesser reproductive success. Extremely high exposure levels of nitrate can kill the aquatic invertebrates and fishes. The early life stages of aquatic animals are more sensitive to nitrate. Larval stages of amphibians appear to be more sensitive to nitrate exposure (Canadian Water Quality Guideline, 1984).

Highest oxidized form of nitrogen is nitrate. In water its most important source is biological oxidation of organic matters. Agricultural runoff and domestic sewage are the main sources of nitrogenous organic matters. Metabolic waste of aquatic community and dead organisms add nitrogenous organic matter. Nitrifying bacteria as aminifying bacteria, nitrosomonas, and nitrobactor plays significant role in oxidation of such organic matter. Azobactor, a nitrogen fixing bacteria and blue green algae like nostoc, anabaena has capacity to fix molecular nitrogen in nitrates. Higher concentration of nitrate indicates pollution of water. This is an important plant nutrient, when present in excess causes more growth of algae often present in blooms. High nitrate content (740 mg NO_3^- N/ lit) may cause blue baby disease.

Janiyani, *et. al.,* (1993) recorded low nitrate content in oil sludge from various sources of refinery. Nitrate content of the Sugar Factory effluent showed marked difference in treated and untreated effluents (Jesudas, *et. al.,* 1995).

Estuaries are areas of constant change caused by varying meteorological and hydrographic factors which regulate the physico-chemical properties of estuarine water and sediments. Especially those estuaries, exposed to strong tidal action, shows fluctuations in various parameters such as, pH, salinity, oxygen concentration and temperature. These are important abiotic factors dictating the chemical 'species' and all together the aqueous solubility of the compound, affinity for sediments, organic carbon content and sediment mineral constituents with the sorption behaviour and in turn the bioavailability of many pollutants (Chapman and Wang, 2001). In addition, represents some of the physical means of releasing pollutants from sediments and their re-introduction into the sediment-water interface by storms, tides and anthropogenic activity (Watanabe, *et. al.,* 1997).

The sediment water interface is an important habitat for demersel or benthic fish, in search for food or shelter. The constant input of particle-associated pollutants, due to sedimentation, brings about potentially high concentrations of contaminants in sediments. Toxic metals and organic compounds can affect living organisms at various levels of organization, inducing a biomarker response. It is therefore necessary to study the toxic effects of pollutants at each level and to incorporate additional parameters, such as season, the developmental stage and sex of the test organism, in order to determine the quantitative relationship between pollutant toxicity, the organism, population sustainability and ecosystem effects. The concise selection of some examples of various benthic fish species used as sentinels for a range of toxic pollutants commonly found in estuarine sediments and their respective biomarkers (Calmano and Forstner, 1996; Luoma and Ho, 1998).

Whole sediment toxicity assays are widely used for ecological impact assessments of anthropogenic activity on fish. The test organism can be observed in its natural habitat, exposed to field-collected sediments in the laboratory or transplanted from a "clean" environment to a polluted one in caging experiments. Apart from a few examples, the biomarker response mainly indicates the presence of a substance class, rather than a specific chemical. Polluted sediments regularly contain complex chemical mixtures, giving rise to the potential problem of synergistic and/or antagonistic effects (Eggens, *et. al.*, 1995). This makes establishing causality in the field particularly difficult and therefore most evidence of biomarker responses recorded in whole sediment toxicity assays are of a correlative nature. Whole sediment toxicity tests without specific biomarkers can highlight the route of exposure and, more importantly, the bioavailability of sediment associated pollutants to a given organism (Crane, *et. al.*, 2000).

Metals exist in the environment as various inorganic species and/or as organometallic compounds. The speciation of most metals affect sorption behaviour and in turn dictates bioavailability to benthic organisms (Bryan, 1985). Hylland, *et. al.*, (1996) found sediment-associated metals to be largely unavailable to caged flounders, whereas bivalves are known to concentrate large amounts of metals in their tissues (Goldberg, 1975).The chemical analysis provides baseline information on the occurrence of potentially toxic pollutants in the environment, but it fails to predict synergistic, additive or antagonistic effects, that may give an important measure of potential biological effects. Biomarkers on the other hand can detect direct and indirect effects of sub lethal concentrations of toxic pollutants and offer additional biologically and ecologically relevant information as a valuable tool for the establishment of guide lines for effective environmental management (Long, *et. al.*, 2000).

However all benthic organisms are not suitable as sentinels for sediment toxicity. Some may be more susceptible to certain pollutants than others (Driscoll and McElroy, 1996). Furthermore, certain pollutants may not cause a measurable response at every level of organization in a specific organism, has negligible impact on one level of organization but might lead to serious knock-out effects on others (Kurelec, 1993). Biomarkers should, therefore, ideally be employed as a part of an integrated programme of pollution monitoring, involving general measurements of biological damage and animal health in a variety of taxa as well as analysis of chemical contaminants in the biota and environment.

The analysis of lake and river sediments is a useful method of studying environmental pollution with heavy metals. Potentially, heavy metals may be detected and monitored by analyzing the sediment, water, or biota. However, sediments record a history of heavy metal accumulation. In addition, bottom sediments are not only a sink of heavy metals but also a source of resuspended sediment. Under changing environmental conditions, sediment bound heavy metals may be remobilized and enter the water or food chain (Dickinson, 1996).

Metal concentrations in sediments have been analyzed in many countries. Industrialization and human activities during the previous century has increased the emission of heavy metals and nutrients like nitrogen and carbon into the environment. The metals enter terrestrial and aquatic environments through several pathways, like atmospheric deposition and point as well as non point source release to surface water (Bohlin, *et. al.,* 2005).

Bioassay can be taken as a tool for identifying the effects or tolerance of chemical by organism. The bioassay can be conducted by using any type of organism but fish bioassay is very common and useful. Thus bioassay could find a very important role in studying synergetic effect of chemical assessment of efficiency of a waste treatment method etc. In the bioassay test the organisms are exposed to different concentrations of toxicants for a definitive period and mortality, behavioral change or other signals of distress are noted periodically. The aim of bioassay is to find out either lethal concentration or effective concentration. Also, they are used to determine safe concentration of chemical or maximum acceptable toxicant concentration.

In static bioassay the organisms are exposed to the same toxicant solution for the whole experimental period.

Renewal bioassay:

The test solution may change many times during the experimental period for renewal bioassay. Metals find extensive use in human civilization because of their high density, high melting and boiling points, high electrical conductivity, ductility and other useful properties.

Toxicity study:

Prof. Truhaut, toxicologist in the late 1960s, introduced "ecotoxicology" term, when it was considered as a sub-discipline of medical toxicology. Since then, ecotoxicology has developed as a scientific discipline in its own right, describing not only effects of exposure to chemicals and radiation but also the environmental fate of contaminants. Affected organism, often cause characteristic responses to the toxic pollutants and are commonly known as 'toxicological 'biomarkers' or endpoints' (Bright, *et. al.,* 2002). A biomarker, as defined by Depledge, *et. al.*, (1993), is "...a biochemical, (genetic) cellular, physiological or behavioural variations that can be measured in tissue or body fluid samples or at the level of the whole organism (either individuals or populations), that provides evidence of exposure and/or effects of one or more chemical pollutants (and/or radiation)". To detect the impact of exposure to sub lethal concentrations of a complex chemical mixtures or given substance, enabling the evaluation of less obvious effects on organisms biomarkers are the powerful tools.

The biomarkers have traditionally been applied to the exposure of sentinel organisms or in vitro test systems to pollutants in aqueous solutions or suspensions in aquatic ecotoxicology. These approaches have been instrumental in providing guidelines for legislative measures aimed at reducing the impact of anthropogenic activity on marine and freshwater environments.

Man invented many chemicals, heavy metals and tested their effects into day's present fast developing world. Up to 1940s or 1950s possible adverse effects of materials on organisms was not recognized. In a developing country like India due to lack of required facilities for effective collection and treatment of effluents, the effluents are either released directly, to water bodies or disposed off on land. This causes gross pollution of aquatic bodies and results in biomagnification of various pollutants. Many indicator organisms have stressed by various scientists which could be employed to monitor environmental contamination by heavy metals (Preston, *et. al.,* 1972; Hung, 1982; Goldberg, 1975).

In the aquatic environmental studies one of the most commonly used biological study methods is toxicity bioassay. Sprague (1973) defined bioassay as "tests" in which the quantity or strength of pollutant material is determined by the reaction of living organisms to it. In the year 1983 Chapman and Long defined the bioassay tests as "Estimation of amount of biologically active substances by the level of their effects on test organisms." Laboratory determination of toxicity may divide into short-term acute toxicity and long term chronic toxicity tests. These are bioassay based responses on short and long term application of times. The former measures the more or less immediate biochemical disruption of life processes, the later permits the day to day mechanism to proceed until they become affected by the disruption of same slower metabolic processes or blockage of metabolic processes.

Acute toxicity tests are easy to conduct and are economical. They are widely used in aquatic tests to provide basic criteria for establishing water quality standards and also help to set dosage levels for long terms or chronic exposure. A diverse bioassay tests have been reported with choice of test type, time under consideration, test organism and sophistication of technique (Sprague, 1973). For the last three decades acute toxicity bioassay is generally useful for measuring the toxicity of different pollutants to aquatic organisms. Since then many workers directed the studies towards the toxicity evaluation (Eisler and Hannekey, 1977; Mohan, *et. al.,* 1986).

To assess the effect of pollutants on growth, physiology, biochemistry, reproduction and behaviors of animal chronic toxicity tests are also important. Presently sub lethal toxicity study has an important status in toxicological investigation. It provides a sensitive tool for evaluation of impact of pollutants on aquatic life. The LC_{50} values rarely fall in the ranges observed in contaminated ecosystem. Therefore to find out effect of metals on animals in real concentration, a large number of studies related to sub lethal effects on metals have been conducted.

The chronic toxicity study involves prolonged exposure of experimental animals to toxic environment can indicate and provide the sole concentration level, which is more safety point of view.

Death of aquatic organisms due to the heavy metals depends on the concentration of heavy metals, type of metal and organism. Generally mercury, silver and copper are most toxic metals followed by cadmium, zinc and lead (Brayan, 1971).This is not rigid toxicity order and it is different for different species. Doudoroff, 1953 has reported that more stable copper-cyanide complex is less toxic to fish than copper ions. Toxicity also depends on certain environmental parameters. In estuarine organisms changes in the salinity with tidal cycles would be the important factor in relation to the metal toxicities. The toxicity of various pollutants and sensitivities also related with variation in temperature in estuarine organisms. Generally, temperature increase reduces the period of survival of fish in lethal concentrations of toxic substances (Bodrova and Kraiukhin, 1956).

Bioaccumulation:

The importance of metals in sediments lies in the fact that under appropriate conditions these metals could leach out of the sediments for many years after pollution discharge had been stopped and thus continues to pollute the water medium. Many workers have estimated concentration of heavy metals in the sea water and sediments (Sengupta, *et. al.,* 1979). In marine environment sedimentation plays a vital role. There are several documents on the effect of released organic pollutants and heavy metal ions from sediments and its effects on coastal area, human beings and other organisms (Copeland and Ayres, 1972). Small amount of toxic materials in the aquatic environment causes changes in internal dynamics of aquatic organisms at sub-lethal level.

The living organisms are affected by negative feed back of environmental pollution. Increasing industrialization and discharge of effluents, heavy metals are becoming important pollutants in aquatic ecosystem (Joshi, *et. al.,* 2002). Heavy metals accumulate directly or indirectly in organism might affect an organism and transformed to next trophic level in food chain (Shah and Altindag, 2005). The heavy metal uptake, distribution and persistence in tissues are well documented (Karuppasamy, 1999). Heavy metals accumulate in the tissues of aquatic animals and may become toxic when accumulated at substantially high level (Kalay and Canali, 2000). Bioaccumulation of heavy metals in fish occurs, either directly from the surrounding water or by ingestion of food (Patrick and Loutit, 1978; Kumar and Mathur, 1991). Fish exposed to a high concentration of trace metals in water may take up substantial quantities of these meals (Sultana and Rao, 1998).

The heavy metals have a great ecological significance due to their toxicity and accumulation nature. World have a great concern to aquatic environment due to release of heavy metals. Water pollution resulted from various sources like accidental spillage of chemical wastes, discharge of industrial or sewage effluents, domestic waste water and agriculture drainage led to affect fish with toxic metals (Handy, 1994; Jent *et. al.*, 1998).

Heavy metals accumulate in the tissues of aquatic animals and affect them due to toxicity at substantial level of accumulation. Levels of accumulation vary with metals and species (Heath, 1987).

The presence and accumulation of heavy metals in aquatic environment have been investigated during recent years (Chen, 2002; Canli, and Ath, 2003; Yimaz, 2003; Murugan, *et. al.*, (2008).

Biochemical composition:

Protein, carbohydrate and lipids are the major components of body, plays an important role in the energy metabolism and body construction. The biochemical composition determines the quality of flesh. Biochemical composition of fish is dependent on species, size, age, sex, environmental factors, fishing ground etc.

Energy for various activities is derived from the metabolic breakdown of carbohydrate by most of the living organisms. In the tissues, the chief carbohydrate reserve is glycogen, which release glucose, an utilizable sugar by glycogenolysis, according to the physiological demands of the organisms. Carbohydrate metabolism is affected due to various environmental pollutants.

For human diet the fish provides an excellent source of protein. Fish protein has relatively high digestibility, biological and growth promoting value for human consumption. Fish protein rank in the same class as chicken protein and are superior to milk, beef protein and egg albumen.

The fat content of fish flesh determines the quality and price of fish. Different parameters and even its variations, these include species difference, diet of fish, selective mobilization, and distribution of lipids in fish body, salinity and temperature of water.

Small amount of toxic materials in the aquatic environment causes changes in the aquatic environment causes changes in internal dynamics of aquatic organisms at sub-lethal level. In aquatic toxicology some literature is available on effects of various pollutants on the biochemical composition of tissues of different types of fishes.

Source of energy for all living organisms is food. Energy obtained from the food is used for building up body tissue. The proximate composition study helps us to find out the nutritional quality of food as a balanced diet is necessary for proper functioning of body (George and Mathew, 1996).

Different biochemical processes are useful to determine the mechanism of toxicity in different toxicants; it is also useful in unfolding the adaptive protective mechanism of the body to combat the toxic effect of the pollutants. Some of the biochemical alterations occurring in the body give the first indication of stress in the organisms.

Amudha and Mahalingam (1999) estimated the protein, carbohydrate and lipid from *Cyprinus carpio* exposed to dairy effluents. Maruthi and Subba Rao (2000) reported the effect of distillery effluent on glycogen, protein and lipid content in liver and muscle of fish *Channa punctatus*. Emad H., *et. al., (*2005) reported toxicity of Cd and Cu and their effect on some biochemical parameters of marine fish *Mugil seheli*. Muley, *et. al.,(* 2007) reported the decrease in total protein and lipid content in tissues like gill, liver, kidney and muscle of fresh water fish *Labio rohita* exposed to tannery, electroplating and textile mill effluents. Sobha, *et. al.,* (2007) reported the biochemical changes in the fresh water fish, *Catla catla* (Ham.) exposed to the heavy metal toxicant Cadmium chloride. Metwally and Fouad, (2008), reported biochemical changes induced by heavy metal pollution in marine fishes and khomse coast, Libya.