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Biological Monitoring of Rivers : Problems and Prospects

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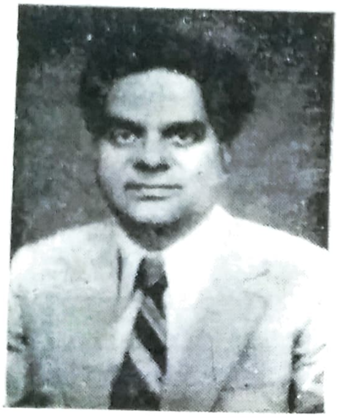
Population pressure, urban and industrial growth and land development have contributed significantly to pollution and toxicity of aquatic ecosystems including the waters of rivers and streams which are undergoing fast qualitative degradation. Pollutants bring about a change not only in physico-chemical quality of water but also trigger a series of changes in biotic components of the ecosystem resulting in depletion of some of the valuable species. This leads to destabilization of the entire system. Biological monitoring of the environment is immensely helpful in assessing the health of ecosystems. Biological monitoring is essentially diagnostic in nature and is based on the simple principle of sensitivity or tolerance of the organisms/communities to environmental changes. Biomonitoring is less expensive and is fairly reliable. The importance of utilizing biological information to supplement chemical findings for better evaluation of pollutional status has attained importance only in the recent years. The use of "Bioindicators" has two distinct advantages (Ravera 1975) (i) the organism does not react to a single factor, but to the entire environmental situation in its totality (ii) the response of the organism is the cumulative effect of its reaction to the preceding as well as the present environmental conditions as against the chemical analyses which gives information only about the "Current affairs" of the system. The physico-chemical and biological qualities of water get changed at all such points where tributaries, industrial effluents and municipal wastes etc. are added to the river. Consequently the primary and secondary productivity of the rivers get affected leading to (i) change in pattern of available energy (ii) change in habitat quality which affects growth, reproduction, breeding and spawning pattern of biota, (iii) changes in flow regime due to silting and floods finally leading to change in biotic interactions, their population dynamics and food chain. Due to these changes, the set pattern of species dynamics gets sharply disturbed. The tolerant species remain almost unaffected but the sensitive ones develop various types of abnormalities/deformities or may even disappear due to addition of "foreign materials." It is the sensitive species that are preferred for biomonitoring exercises.



We got interested in biomonitoring while studying the different ecological aspects of the river Ganga. By using suitable indicators it is not difficult to recognize the pollutional load in different segments of the river. The indicators may also be helpful in determining the nutrient status of the flowing waters. The speed with which an organism reacts to the changing aquatic environment depends upon a large number of variables. Before undertaking any biomonitoring programme in specified ecosystems, it is essential to distinguish the eco-regions, that is the regions of greater homogeneity within an ecosystem.

Biological monitoring of aquatic bodies specially the rivers is a Herculian task because of their ever changing nature both region and season-wise due to variations in climate, topography, soil, surface geology, vegetation and land use pattern of the catchment areas. Moreover a wide variety of toxic materials which enter the riverine systems through different routes greatly affect the water quality. Potential toxicants are usually present as a mixture. A number of physico-chemical characters of water like hardness, pH, temperature and dissolved oxygen affect the effectiveness of the toxicants and vice-versa. The Ganga basin has been distinguished in 6 ecoregions i.e., Himalayas, northern plains, central high land, central plateau, eastern plateau and eastern plains. The physico-chemical and biological quality of water differs in different eco-regions. Habitat heterogeneity has important bearings on the functioning of the ecosystem. Vannote *et al* (1980) however, feel that heterogenicity is unimportant in rivers and they have proposed "River Continuum Concept" (RCC). According to this, the rivers exhibit a longitudinal constancy or structural uniformity despite different environmental histories. However, the organisms for biomonitoring work should be selected according to the objective and essential features of eco-region. While it may be possible to select an organism that is sensitive to several toxicants in a particular type of industrial waste effluent, it is unlikely that any single organism could respond at a proper level to the wide range of chemicals in drinking water that might be harmful to man. Indicator organism can be selected practically from every group of river biota i.e., algae, fungi, bacteria, macrophytes, fishes, insects, reptiles, molluscs etc. Selection of proper indictor species for a particular zone or stretch of river is one of the most challenging aspects of biomonitoring work. We must ensure that the selected organisms are such which are able to indicate the health of the system through their perceptible and discernible functional snags. Some species may show loss of sensitivity to the toxicants following long-term exposure to very low levels of toxic materials. It is because the polluted water, as an environment, selects those individual of a single population which are resistant to pollutants. Bioindicators in this context are themselves endowed with extreme plasticity and they can be put in various categories of primary, secondary and tertiary detectors, depending upon the past history of the polluted water body they live in.

Algae Algae are one of the most rapid detectors of environmental pollution. This is because of their quick response to toxicants and other chemicals. Being the principal primary producers in aquatic ecosystems, algae are uniquely significant because those algae which accumulate toxic materials may intoxicate the entire food chain. Pollution stress reduces the number of algal species with a concurrent increase in the number of their individuals. In this manner the complex organization of the community gets actually simplified under pollution pressure (Patrick 1949). A marked change in algal community severely affects the species diversity. It is observed that in heavily polluted habitats, bacteria replace algae as a result of this, the photosynthetic biomass decreases. The lowering of species diversity leads to many detrimental effects on structural and functional aspects of ecosystem. Highly diverse ecosystems have longer as well as many alternative food chains thus such systems are more resistant to perturbations. Pollution of aquatic ecosystems often results in substantial increase of blue-green algae which are more tolerant but replacement of green algae and diatoms by such algae is not desirable because many organisms do not prefer the blue-greens as food. Plentiful algal species were recorded from organically polluted sites of river Ganga (Bilgrami *et al*. 1985). A number of indices including Nygaard's algal indices (1949), Shannon and Weaver's (1949) species diversity index, Cairn's *et al*. (1968) and Palmer's algal pollution indices (1969) have been used to sort out such algal species which can be employed as pollution indicators. Margalef (1958) recommended the use of information

theory by Shannon and Weaver for studying the structure of algal communities. This theory relates diversity with information. According to it, more information is contained in a community with high diversity and vice-versa. Palmer (1969) reviewed the work of 165 investigators and found 8 algal genera to be the most dominant in organically polluted waters. They are : *Euglena, Chlamydomonas, Oscillatoria, Scenedesmus, Chlorella, Nitzschia, Navicula* **and** *Stigeoclonium.* **Bilgrami** *et al* **(1985) recorded** *Euglena acus, E. viridis, Oscillatoria chlorina, O. limosa, O. tenuis, Microcystis aeruginosa, Chlorella vulgaris, Closterium acerosum, Ankistrodesmus falcatus, Scenedesmus quadricauda, Stigeoclonium tenue* **and** *Synedra ulna* **from the sewage disposal sites in river Ganga between Patna to Farakka.**

The ability of algae to take up heavy metals from water is well established. Due to greater accumulation of heavy metals the damage to the metabolic processes may be quite distinct and extensive. We have recorded a large variety of metal accumulating algae in river Ganga. These may be planktonic, periphytic or benthic in nature. Periphytic and benthic algae are more reliable for monitoring work. Collection, segregation and identification of benthic algae, however, pose many problems. Patrick ***et al.*** **(1954) suggested the use of plexiglas slides for determining the colonization of algal communities. This method is open to criticism because of the use of artificial substrates; nevertheless, it is widely used. A direct scrap of periphytic communities also does not give correct estimates since an important fraction of the algal community withers away during sampling. Several other artificial substrates have been tried but none of them is satisfactory. Some of the important algae which can be used as indicators of industrial waste are:**

Algal indicators of industrial wastes

Material	Indicator algae
Copper	*Calothrix braunii*
	Scenedesmus obliquus
	Stigeoclonium tenue
	Navicula viridula
	Cymbella ventricosa
	Achnanthes affinis
	Nitzschia palea
Iron	*Pinnularia subcapitata*
	Suriella delicatissima
	Trachelomonas hispida
Chromium	*Stigeoclonium tenue*
	Closterium acerosum
	Euglena acus
	E. viridis
	Navicula cuspidata
	Nitzschia linearis
Oil waste	*Amphora ovalis*
	Diatoma vulgare
	Melosira varians
	Navicula radiosa
	Synedra acus
Paper Mill Waste	*Ulothrix zonata*
	Pandorina morum
	Pediastrum simplex
	Scenedesmus bijuga
	Cymbella ventricosa
	Navicula cryptocephala
	Synedra pulchella
	Surirella ovata
Distillary waste	*Chlamydobotrys sp.*
	Chlorogonium gracillima
Phenolic waste	*Achnanthes affinis*
	Cocconeis placentula
	Cyclotella kuetzingii
	Cymbella naviculiformis

	Fragilaria virescens
	Pinnularia borealis
Hydrogen sulphide waste	*Cymbella ventricosa*
	Navicula minima
	Nitzschia ignorata
Highly acidic material	*Euglena mutabilis*
	E. stellata
	Lepocinclis ovum
	Xanthidium antilopaeum
	Cryptomonas erosa

As a general rule, Cyanophyceae, Euglenophyceae, Centric diatoms and members of Chlorococcales are characteristics of polluted waters while pennate diatoms and desmids are usually found in oligotrophic waters as they do not tolerate high nutrient levels. Variation in pollution tolerance of algae is attributed to genetical and physiological adaptations. To a great extent the degree of sensitivity of algae to metals and other pollutants depends upon the sensitivity of the membrane system.

The genus *Oscillatoria* is perhaps the most frequently encountered algae in polluted waters. Species of *Oscillatoria* and *Phormidium* have been used as oil detectors in aquatic bodies.

Species Diversity At the beginning of this century Kolkwitz and Marsson (1908) proposed that some species tend to occur under a certain kind of pollution and their presence is indicative of the quality of water. The concept of species diversity is based on (i) *Richness,* also called as species density indicating the total number ofspecies present. (ii) *Evenness,* based on relative abundance and degree of its dominance. *Relativeabundance* is the mean of the number of individuals of any species per sampling unit.

The maintenance of *genotypic, heterozygosity, polymorphism* and other genetic variability is an adaptive necessity of natural populations. Many ecologists are concerned about reduction in species and genetic diversity resulting from various anthropogenic activities which is jeopardizing the future adaptability of natural ecosystem. For a species to survive and reproduce in an environment, it must have a behavioural flexibility which is almost inversely proportional to the predictability of its environment (Goodman 1975.

Diversity is principally a mechanism which generates community stability; while dominance is principally a mechanism which generates community productivity. In fact, greater diversity provides a number of alternative pathways to the ecosystem whereas single species crops are least stable.

A number of indices to indicate dominance, similarity/dissimilarity and species diversity have been proposed by ecologists from time to time which I do not consider them necessary to recall.

Bacteria Bacteria exist in very diverse habitats in the aquatic environment. They are associated with all the types of available surfaces in water including plants, rocks, animals, sediments, floating materials and planktons. They are found in environments showing extreme physical and chemical variations. Bacteria are a great health risk but are also very good indicators of organic pollution.

Due to their characteristic physiological and genetical features bacteria serve as very good indicators of organically polluted environment. The main activity of bacteria is concerned with the transformation of organically bound carbon, nitrogen, phosphorous, magnesium, sulphur and other complex materials into unbound oxidized states i.e., their role in mineralization. If mineralization and nutrient recycling get disturbed, the functioning of the ecosystem is also affected because the two are intimately linked.

Effect of toxicants on bacterial growth is relatively easy to measure in laboratory. It can be studied in natural mixed populations. Growth pattern sharply responds to specific physical and chemical characteristics of aquatic environment.

Indicator coliform organisms have been used extensively by many investigators to gauge the quality of aquatic environment. On the basis of occurrence and distribution of human pathogen in aquatic environments, the coliform indicator organisms such as faecal coliform and other species like *Escherichia coli*, *Pseudomonas aeruginosa*, *Streptococcus faecalis* or *Clostridium perfringens* have been used. Contradictory results have been published on the behaviour of the most commonly used indicator *E. coli* when counted by standardized methods. *E. coli* is the first to disappear in space and time while *Salmonella* spp. would be recovered from the same niche. The validity of such results, however, is questionable. Bacteria suit for monitoring exercises due to their rapid life cycle.

Many compounds enter the rivers as industrial effluents and agricultural run off causing alteration in bacterial communities. Thus bacteria have the advantage of being used as indicators of physical and chemical pollution. Specialized bacterial species have been used for monitoring hydrocarbons, heavy metals and polychlorinated biphenols. Oil spillage in rivers leads to an enrichment of hydrocarbon bacteria. Monitoring of population density of *Salmonella* can be successfully exploited as alternative approach because mere determination of coliform MPN may not give the correct picture about the organic load of water. MPN of coliform bacteria is invariably very high in all such waters which have domination of algae belonging to Cyanophyceae and Euglenophyceae. A complex automated monitoring system developed by U.S. Department of National Aeronautics involves the utilization of three bacterial biomass devices along with conventional physico-chemical censors. Total bacteria (living and dead) counts are determined by measuring light from chemiluminescent reaction catalysed by porphyrins from lysed bacterial cells. An estimate of living bacterial biomass is found by assaying ATP from lysed cells. Total coliform and faecal coliform levels are estimated by measuring hydrogen gas evolved by these organisms in presence of lactose at certain incubation temperatures. Kogure (1985), however, feels that ATP is not a suitable material because bacteria cannot be completely separated from other microbes. Moreover, there is no chemical which is specific only for bacteria. Lipopolysaccharide is considered reliable material for bacteria, although gram-positive bacteria lack it. It is, however, difficult to apply only one parameter to any environment.

Macrophytes Aquatic vascular plants have been used as pollution indicators due to their capacity to accumulate metals and other pollutants from water. *Lemna*, *Eichhornia*, *Utricularia*, *Myriyophyllum*, *Nuphar* ,*Potamogeton* and several other macrophytes have been extensively employed as indices of water quality in culture and field conditions. Since many pollutants occur in surface water, plant community functions are likely to be affected by them. The change in metabolism, growth rate, altered photosynthetic and respiration rate disrupts the chloroplast structure and chlorophyll content indicating that vascular plants are quite sensitive to pollutants. Duck weeds offer a promising material for such studies. Stunting of growth of *Lemna paucicostata* by heavy metals as well as a reduction in the frond count of *Spirodella polyrrhiza* was recorded. *Elodea densa* plants exposed to low levels of methylmercury result in evere damage to the surface membrane and internal organization of the leaf chloroplast.

Another important aspect of uptake and toxicity of pollutants to vascular plants is altered the plant component levels. It is known that metaisomers of phenols were toxic to common wild growing species of *Vallisnaria*, *Nitella* and *Elodea* where unspecified cell protein, structural protein of the organelles and also the cytoplasmic protein, involving cell mortality and cytoplasmic streaming were much less than those of the control cells. Since these processes are energy consuming the implication of the study could go as far as the ATP generation system of the cells being affected by these phenolic compounds. Some of the macrophytes which are quite extensive in fresh water ecosystems can be used for biomonitoring work in view of their rapid multiplication rate, easy propagation, genetic uniformity and high sensitivity pollutants.

Zooplankton Like algae and macrophytes, the zooplankton also constitute a very broad base of the food chain and the second trophic level in aquatic ecosystems. They form the most important link between the primary producers and higher aquatic biota. Among the zooplankton, the rotifers and crustaceans like the cladocera and the copepods have been often used as indicators of aquatic environment. Some biologists have suggested that instead of simple presence/absence criterion of individual species, their actual and relative abundance can provide more dependable data for biomonitoring works. *Brachionus plicatus* shows extreme tolerance to brackish water and it is a good indicator of estuarine environment. Bick (1972) published a list of 84 ciliates which could be used as indicators of fresh water quality. Some cladocerans like *Daphnia magna* are excellent materials to test aquatic toxicity. Because of their relatively large size, short generation time, genetic uniformity, easy handling and maintenance, high fecundity rate, low experimental cost and high sensitivity to pollutants this species ideally suited for monitoring the aquatic environment. Cladocerans are among the most important organisms which are nutritionally very valuable for higher animals like fishes. Some biologists are of the opinion that *Ceriodaphnia reticulata* is easier to culture and more convenient for toxicity test than D. *magna*. D. *carinata* is more sensitive to parathion, malathion, DDT and mercuric chloride than the fingerlings of silver carps. The possibility of monitoring the photolactic response of microcrustaceans like *Daphnia* and *Attemia* has been discussed by Willingham and Anderson (1967).

Fishes Fishes are very well suited for biomonitoring programmes because they can be easily identified and put in suitable tanks monitor the quality of water. Jackson and Brungs (1966) visually observed their mortality rate as well as "signs of stress" to determine the water quality. Visual monitoring of lethal effects has the obvious draw back of requiring that some one be present continually to observe the organism. Moreover, there may be considerable delay between the onset of toxicity and death. Fishes are distributed in all types of water and represent integrative picture of the watershed conditions. Moreover, from the point of view of trophic structure, they represent practically all the categories i. e. omnivores, detritivores, herbivores, insectivores, planktivores and piscivores. Their dietry range is diverse and they occupy the most important position in the aquatic food-chain. Their populations are comparatively more stable and these are convenient materials for performing toxicity tests. Due to their long range movement, they integrate relatively a larg-span of the river. Authentic record of the fish landing data at different hauling centres gives an idea about the nutritive strength of the river as well as spawning, reproductive and predatory capacity of different varieties of fishes. Unlike bacteria, algae and protozoa, the mature fishes are generally not influenced by microenvironmental factors. Moreover, the effects of toxicants or stressed conditions can be easily demonstrated as compared to bacteria and planktons. Fishes are ideal for undertaking eco-physiological experiments. Views differ regarding the indicator value of "resident" and "visitor" fishes. Both have their advantages and disadvantages. Fishes from different niches may be used for monitoring of unknown niche. Bilgrami and Datta-Munshi (1979) found "resident" fishes to be very suitable for monitoring the toxic potentials of the effluents discharged in river Ganga from a tannery and a distillery at Mokama (Bihar).

Some of the morphological structures like gills are very sensitive to pollutants and their external appearance changes and they become highly susceptible to fungal infections. Comparative respiration of two species of *Sagitta* has been used for evaluating the health of aquatic ecosystem. In most fishes the ventilatory movements change whenever the oxygen level declines. Reduced oxygen levels introduce many other morpho-physiological abnormalities in fishes.

For biomonitoring programme an understanding of physiology/biochemistry of the organism is essential. In order to undertake such studies, important factors should be considered:

a. Understanding of the function and composition of organs primarily affected by toxic chemicals,

b. Knowledge of analytical procedures for measuring appropriate constituents, cellular morphology and physiological responses,

c. Ability to interpret the significance of chemically induced biophysiological changes and relate them to the health and survival of the organism.

Variation in comparative physiology of the organisms has also been used in biomonitoring of ecosystems. The adaptation rate of different organisms varies. In some organisms the physiological changes are sharp and these may be directly proportional to the degree of pollution. A variety of biochemical methods have been used in biomonitoring studies. Some of the studies are concerned with the assay of changes in the nervous system or changes in the blood of fishes. Assessment of an ecosystem is possible by recording ventilatory and coughing frequency. Cardiac rhythm and haematological measurements in fishes are helpful in monitoring the stressed riverine ecosystems. Perhaps the most obvious physiological parameter of the respiratory system is frequency of ventilatory movement (Hughes, 1985). In most cases, frequency increases when oxygen levels are reduced (hypoxia).

Many morphological deformities have been noted in the organisms obtained from the polluted zones of rivers. Some fishes develop abnormal vertebral columns. Chironomid larvae develop abnormal mouth parts under stressed conditions.

In order to be effective, the biomonitoring programme must include measurements of toxicity and/or biostimulatory properties of effluents and the effect of effluents on the aquatic life of rivers. Some efforts have been made in the recent years to correlate them in stream biological response with laboratory toxicity tests.

However, due to a variety of complex factors, it is very difficult to predict the impact of any individual toxicant on the indicator organisms. Most of the tests are based on "whole efflument" approach. Complete simulation of riverine ecosystem is not possible under laboratory conditions.

Despite a number of studies on the effect of pollutants on physiological processes of plants in rivers, it is only with animals, specially the fish in cages, that *insitu* assays have reached the stage of practical monitoring (Cairns & Dickson 1978).

Toxicity under laboratory conditions is measured by exposing the organisms to clean or polluted water for a definite period in the static environment and observing the survival/or other symptoms during the period up to 96 h. Two toxicity tests i.e., acute and chronic tests are performed for this biomonitoring programmes. Acute toxicity test involves quick evaluation lasting for a short time. The duration of an acute aquatic toxicity is generally 4 days or less and mortality figures are counted. On the contrary, chronic tests involve a stimulus that lingers or continues for a long time, it often signifies periods from several weeks to years depending on the reproductive life-cycle of the aquatic species. Chronic aquatic toxicity test is used to study the effects of continuous, long term exposure of a chemical or other potentially toxic material. Trouts and *Daphnia magna* are reported to be sensitive to most of the toxicants. In order to select the sensitive species, it is essential that all the aquatic organisms including plankton, periphyton, macrophytes, macroinvertebrates and the fishes are sampled.

The most appropriate approach for toxicity test should begin with the selection of the sensitiv- species from different grou- and subsequently b- different dilutions tory conditions. have been perfor- and macrophytes. But no definite protocol could be developed so far. Efforts have been made to develop laboratory microcosms inoculated with limited species components (Yasuno 1985-86) but complete simulation of flowing water system has not been possible.

Some histopathological and cell biological studies are helpful in predicting and assessing the environment. Most of these investigations relate to the genetic system. Serious changes are caused to chromosomes, cell organelles including mitochondria, nuclear membrane and other cellular parts. Deming (1985) reported that chromosomes in meiotic pollen mother cells of *Tradescantia paludosa* were damaged when exposed to waste waters of rivers. The chromosome segments became micronuclei in tetrad stage and therefore, frequency of micronuclei could be used as an index of pollution. Heavy metals like chromium and zinc also increase the concentration of micronuclei. Thus this is one of the reliable parameters for biomonitoring programme.

This is the appropriate stage for developing automatic biological monitoring systems. With a large variety of sensitive organisms in fresh water bodies and recent advances in computer technology, we can hopefully look forward in designing proper early warning systems for protecting the rivers from pollutants and other ecological hazards.

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