

Fish Scales as Pollution Indicator in Harike Wetland

Onkar Singh Brraich¹ and Sulochana Jangu^{*}

¹Assistant Professor, Department of Zoology and Environmental Sciences,
Punjabi University, Patiala-147002, Punjab, India

^{*}Research Scholar, Department of Zoology and Environmental Sciences,
Punjabi University, Patiala-147002, Punjab, India

*E-mail: su.kj_2008@yahoo.co.in; Phone No.-8146877835

Abstract:

Wetland ecosystems are being degraded due to discharge of toxic effluents from industries. Pollutants are posing serious threats to the variety of flora and fauna existing there. Fish fauna which is important from ecological as well as economical point of view is facing threat due to the discharge of these pollutants. Fish scales are employed as pollution indicator during the course of present investigations. Altered structural and elemental composition of the cycloid scale of *Labeo rohita* (Hamilton-Buchanan) has been observed by employing scanning electron microscopy (SEM) and energy dispersive X-ray microanalysis (EDX) techniques. The structural damage was in the form of uprooting of lepidonts from the anterior side of the scale thus resulted in the loosening of hold of the scales on the body of fish. EDX technique revealed that normal scale consists of Calcium (Ca-38.26%), Phosphorus (P-31.32%), Oxygen (O-22.36%) and Magnesium (Mg-8.06%), but fish scale taken from Harike wetland (Ramsar site) consists of Oxygen (O-59.54%), Magnesium (Mg-0.49%), Aluminum (Al-0.93%), Silicon (Si-1.29%), Phosphorus (P-16.85%), Calcium (Ca-19.05%), Sulphur (S-0.10%), Lead (Pb-0.39%) and Chromium (Cr-1.36%). The changes in elemental composition of the scale have indicated that it can be used as a reliable pollution indicator in Harike wetland.

Key words: Ramsar site; Harike wetland; Pollution; Fish; Scale; SEM; EDX.

Introduction

Harike wetland is situated at the confluence of two major rivers Sutlej and Beas, which has been recognized as wetland of international significance (Ramsar site). It

provides breeding and feeding ground to many fish species. The tall grasses and other vegetation serves as a host to thousand of migratory ducks during the winter season. It also provides staging ground to large number of globally threaten migratory water fowls every year. This wetland receives pollution from various quarters. Its initial mesotrophic character, with passage of time transformed into eutropic by receiving many kinds of effluents from industry, agriculture and domestic disposal (Ladhar and Brraich, 2005).

The deteriorating water quality affects man, animals and plant life with far-reaching consequences. In India, due to tremendous urbanization and industrialization, the problem of water pollution has assumed an alarming situation, with the rise of pollution in the aquatic ecosystems (Singh and Nautiyal, 1990; Trivedi *et al.*, 1990; Kishore *et al.*, 1998). In recent years, increased industrial developments and agricultural processes resulted in increased level of toxic metals in environment. The contaminations of water with a wide range of toxic metals are a matter of serious concern because of the threat to public water supplies and damage caused to aquatic life. Harmful effects have been realized on aquatic organisms. Such common practices warrant additional monitoring of anthropogenic agents and their effects on aquatic biota. Fishes are useful organisms in the study of heavy metals contamination, because they explore freely in the different trophic levels of the aquatic ecosystem (Chopra *et al.*, 2001; Palaniappan *et al.*, 2009). The earlier studies relating to the effects of heavy metal pollutants (Johal and Mehta, 1994; Johal *et al.*, 1994; Johal and Dua, 1995; Johal and Sawhney, 1999) have indicated the alterations in the mineral profile of the different regions of the scales in laboratory conditions. However, in recent years, anthropogenic pressure on natural aquatic ecosystems has created an extensive ecological imbalance which poses a serious threat to the valuable flora and fauna existing there. River Sutlej receives toxic effluents from Budha Nallah and Kala Sanghian drains which are tributaries of this river. These drains carry toxic effluents from various industries as well as municipality sewerage of Ludhiana, Jalandhar and other cities. These toxic effluents ultimately reach at Harike wetland. Therefore, it is necessary to conserve this important wetland ecosystem. The present investigations take into account the deleterious effect of deposition of pollutants and heavy metals on fish scales, thus enabling them pollution indicator at Harike wetland.

Study Area

The Harike wetland ecosystem, rich in aquatic flora and fauna spread into the three districts i.e. Tarn Taran, Ferozpur and Kapurthala in the Indian States of Punjab. It is one of the largest aquatic wildlife sanctuaries of North India covers an area of 41 sq. km. It is located between latitudes 31°13'N and longitudes 75°12'E. The area is drained by the Sutlej and Beas rivers and their tributaries. This manmade wetland not only recharges ground water but also provides irrigation to parts of Punjab and neighbouring state of Rajasthan (Fig.1).

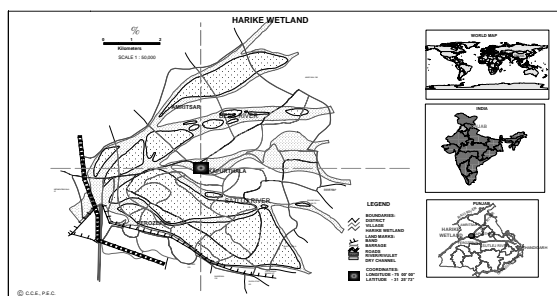


Fig. 1: Map of Harike wetland

Materials and Methods

Samples of fish species *Labeo rohita* were collected from Harike wetland. The scales were removed from the sides of body between the dorsal fin and lateral line with the help of fine tweezers. These scales were cleaned with freshwater using the fine brush. The clean and air dried scales were mounted on the metal stubs with dorsal surface upward and coated with 100Å thin layer of gold in gold coating unit. The gold coated scales were viewed under vacuum in JEOL JSM-6610 LV scanning electron microscope at an accelerating voltage of 20 kv at the low probe current. The elemental composition of scales was determined by energy dispersive X-ray microanalysis (EDX) by adjusting the scanner of INCAX-act analyzer attached to JEOL JSM-6610LV Scanning electron microscope.

Results

The normal scale of *Labeo rohita* consists of circuli which bear tooth like structures and regularly spaced lepidonts (L) placed in relatively deep sockets in anterior portion of the scale (Figs.2 & 3).

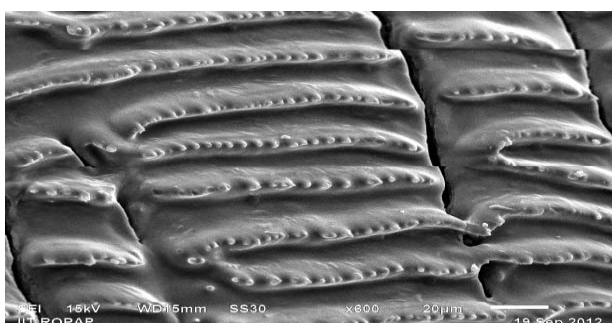


Fig.2: Scanning electron micrograph of control fish *Labeo rohita* (Hamilton-Buchanan) showing regularly spaced lepidonts on the circuli.

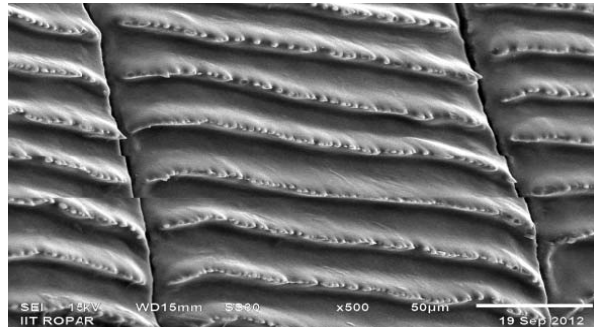


Fig.3: Closely placed lepidonts on the anterior circuli from the fish taken as control.

Various alterations were observed in the structure of scales of fish from Harike Wetland by employing Scanning Electron Microscopic technique. The polluted water and different metal pollutants present in water have adverse effects on the scale structure. The lepidonts were uprooted from their point of attachment to the scale (Figs.4&5). Circuli were broken and disorganized (Fig.6). At some places the damage was so prominent that not only individual lepidont was dislocated, but the whole row of lepidonts was sloughed off from circuli (Fig.7). All the calcareous material was seen to be disorganized. The disruption and dislocation of these lepidonts induces the loosening of the scales from the fish body. Thus, on the basis of present investigations, it is suggested that alterations in the calcareous structure of the scales can be successfully used to announce the pollution in the water of Harike wetland and scales can be used as reliable pollution indicator in aquatic ecosystems.

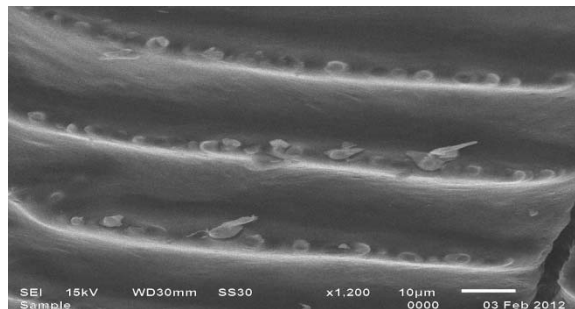


Fig.4: Scanning electron micrograph of scale of *Labeo rohita* (Hamilton-Buchanan) exposed to heavy metals showing uprooted lepidonts from the circuli.

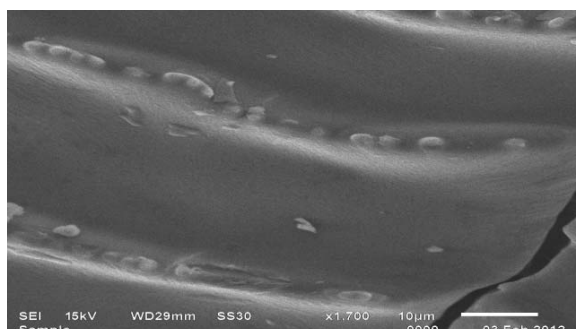


Fig.5: Scanning electron micrograph of scale showing dislocation of lepidonts from circuli.

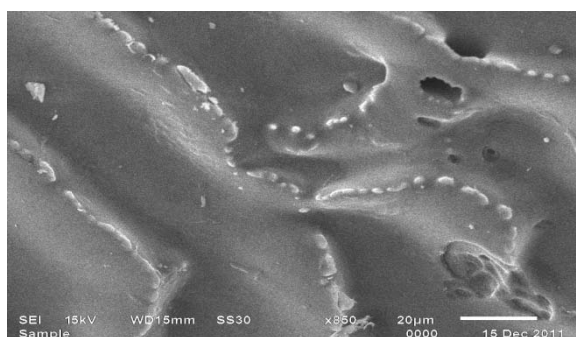


Fig.6: Broken and disorganized circuli.

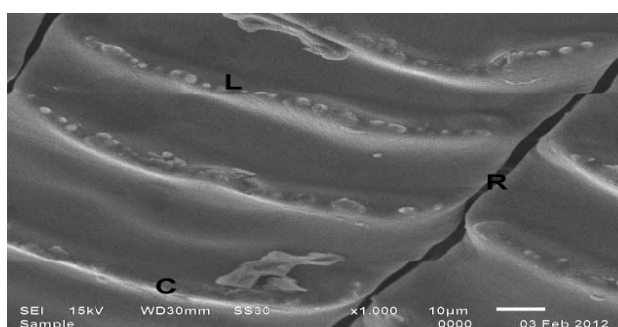


Fig.7: Scanning electron micrograph of scale showing a complete row of lepidonts is sloughed off from circuli (C).

ELEMENTAL COMPOSITION OF THE SCALES

Composition of the normal scale of *Labeo rohita* (Hamilton-Buchanan) or Control fish

EDX analysis has indicated the percentage composition of the elements which consists of four elements viz. Calcium (Ca-38.26%), Phosphorus (P-31.32%), Oxygen (O-22.36%) and Magnesium (Mg-8.06%) present in the anterior part of the control fish scale of *Labeo rohita*. Among these elements, Ca comprises the maximum

percentage while Mg forms the least of the elements recorded in the scale (Fig.8).

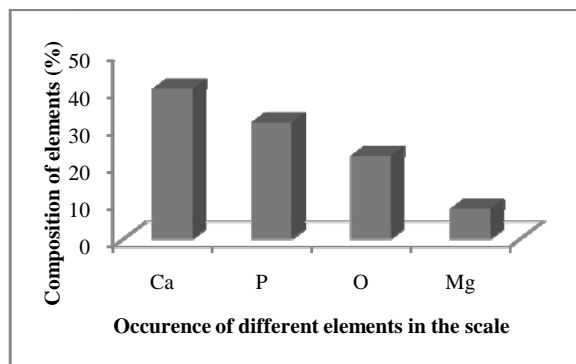


Fig. 8: Elemental composition of the scale of fish *Labeo rohita* taken as control.

Elemental composition of scale of *Labeo rohita* (Hamilton-Buchanan) exposed to Heavy Metals

As assessed by Dispersive X-ray microanalysis (EDX) the various elements present in the scale of fish includes: Calcium (Ca-19.05%); Magnesium (Mg-0.49%); Phosphorus (P-16.85%); Chromium (Cr-1.36%); Oxygen (O-59.54%); Aluminium (Al-0.93%); Silicon (Si-1.29%); Sulphur (S-0.10%) and Lead (Pb-0.39%). The percentage composition of these elements in the anterior part of the normal scale has been given in Fig. 8. Among all elements Ca, O and P constitute the major elemental part of the scale whereas S, Mg, Si, Cr, Al and Pb present in a small percentage (Fig. 9).

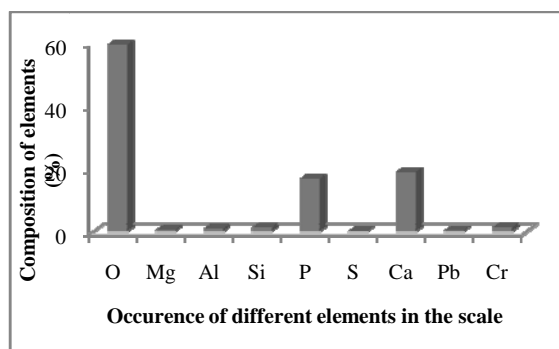


Fig. 9: Elemental composition of the scale of *Labeo rohita* exposed to heavy metals.

The elemental composition of the scale is directly related to the hydrochemistry of the aquatic environment in which the fish inhabits. Keeping above fact in mind, scales were employed for EDX analysis and found that heavy metals adsorption has altered the normal elemental composition of the scale of the fish. A normal scale of fish *Labeo rohita* has elements like Calcium (Ca-38.26%), Phosphorus (P-31.32%),

Magnesium (Mg-8.06%) and Oxygen (O-22.36%). While the EDX analysis of the scales of *Labeo rohita* from Harike wetland, has indicated that elements like Silicon (Si-1.29%), Sulphur (S-0.10%), Chromium (Cr-1.36%), Aluminium (Al-0.93%), Magnesium (Mg-0.49%), Calcium (Ca-19.05%), Oxygen (O-59.54%), Phosphorus (P-16.85%) and Lead (Pb-0.39%) are also present in the elemental composition, which means all these metals present in the water of wetland in which the fish abode. Presence of all these elements in water causes breakdown of lepidonts and ultimately shedding of scales takes place from the body of the fish. Hence, fish comes in direct contact with heavy metals which is fatal for their existence in the nature. However, the fishes comes under critically endangered category will go to extinction. The endangered species will slip into critically endangered category.

Discussion

The body of the fish is covered with scales and they come in immediate contact with the aquatic environment. The lepidonts present on the anterior dorsal surface of the scale help to anchor to the body of the fish. Scales adsorb heavy metals from the water and becomes brittle resultantly breakages in the lepidonts takes place. Cracks and lesions in the shape of circuli and changes in elemental deposition of scales on exposure to the pesticide under laboratory conditions were also reported. Ultimately scales loose their intactness and shed off. Hence, scales can be considered as bioindicator of pollution (Lanzing and Higginbothan, 1974; Tandon and Johal, 1993; Dua and Johal, 1994; Johal *et al.*, 1994; Johal and Dua, 1994, 1995; Johal and Sawhney, 1997; Johal and Grewal, 2006). Similar observations have been reported during the present investigations in the scale of the fish from Harike wetland.

Adverse effects of cadmium have been recorded as alteration in scale structure of *Cyprinus carpio* (Rishi and Jain, 1998). Similar effect of cadmium and lead on scale structure were observed in fish *Tilapia nilotica* and Atlantic cod fish (Rashed, 1999; Basu *et al.*, 2007). Effect of Mercury on the scale structure of *Channa punctatus* have been reported by Dua and Gupta (2005). Rauf *et al.* (2009) observed high concentration of Cadmium, Nickel, Iron and Chromium in the various body parts of *Catla catla*, *Labeo rohita* and *Cirrhinus mrigala* from Baloki Headworks because it receives high load of toxicants. Similar, observations have been recorded during the present investigations that river Sutlej receives polluted water of the Budha Nallah and Eastern Bein which are tributaries of the river Sutlej and ultimately this polluted water reaches at Harike wetland. Thus, high concentrations of toxicants have been observed in the scales of fish under report. Singh and Saharan (2010) conducted research on the water of the River Sutlej in Himachal Pradesh and observed various toxicants like S, Cl, Ca, Ti, V, Cr, Mn and Fe. The concentration of these toxicants is varied, but water sample showed little higher concentration of Ca and Fe. The concentration of Calcium may be due to nearby prevalence of mountains. Calcium is responsible for the hardness of the water. The occurrence of other element believed to be entered into this river through anthropogenic activities. During the present investigations, the presence of Calcium, Magnesium and Phosphorus may be entered into the water through local geological occurrence of these elements and occurrence

of heavy metals are resulted due to discharge of industrial and sewerage effluents.

Fishes are widely used to evaluate the health of aquatic ecosystems but pollutants bio-accumulate in the food chain and responsible for adverse effects and causes mass mortality of the fishes. Heavy metals also alter the physiological and biochemical parameters both in tissues and blood besides scales (Tort and Torres, 1988; Canli, 1995; Farkas *et al.*, 2002; Basa and Rani, 2003). The purpose of present investigations is to quantify the accumulation of various toxicants in the scales of the fishes. During the present course of work, the order of various elements accumulated in the scales of *Labeo rohita* was O>Ca>P>Cr>Si>Al>Mg>Pb>S. The result clearly indicates that the occurrence of heavy metals definitely affects the aquatic life of fresh water fishes of Harike wetland. Thus, on the basis of present investigations, it is observed that alteration in the calcareous structure and elemental composition of the scales can be successfully used to announce the pollution in the water of Harike wetland and scales can be used as reliable pollution indicator in aquatic ecosystems.

Conclusion

The toxicological studies of scales employed by scanning electron microscopy (SEM) and energy dispersive X-ray microanalysis (EDX) clearly indicate that structural damage and alterations in the elemental composition of the scales are excellent pollution indicator and announces pollution in the water of Harike wetland with authenticity.

Acknowledgements

Authors are very thankful to the Head, Department of Zoology and Environmental Sciences, Punjabi University, Patiala for providing necessary laboratory facilities.

References

- [1] Basa, S.P., and Usha Rani, A., 2003, "Cadmium induced antioxidant defense mechanism in freshwater teleost *Oreochromis mossambicus* (Tilapia)," *Eco. Toxicol. Environ. Saf.*, 56, pp. 218-221.
- [2] Basu, A., Rahaman, M.S., Mustafiz S., and Islam, M.R., 2007, "Batch studies of lead adsorption from a multicomponent aqueous solution onto Atlantic cod fish scale (*Gadus morhua*) substrate," *J. Environ. Enginen. Sci.*, 6, pp. 455-462.
- [3] Canli, M., 1995, "Natural occurrence of mettallothionein like protein n the hepatopancreas of the Norway lobster *Nephrons norvegicus* and effects of Cd, Cu and Zn exposure on levels of the metals bound on mettallothionein," *Turk. J. Zool.*, 22, pp. 149-157.
- [4] Chopra, R., Verma, V.K., and Sharma, P.K., 2001, "Mapping, monitoring and conservation of Harike wetland ecosystem, Punjab, India, through remote sensing," *Int. J. Rem. Sens.*, 22, pp.89-98.

- [5] Dua, A., and M.S., Johal, 1994, "Use of fish scale as indicator of pesticide pollution (Abstract). *Proc. National Symposium on Aquaculture For 2000 A.D.*, Madurai Kamraj University, Madurai, India.
- [6] Dua, A. and Gupta, N., 2005, "Mercury toxicology as assessed through fish scales" *Bull. Environ. Contam. Toxicol.*, 74, pp. 1105-1110.
- [7] Farkas, A., Salanki J., and Specziar, A., 2005, "Relation between growth and the heavy metal concentration in organs of *Abramisbrama* L. populating lake Balaton," *Arch. Environ. Contam. Toxicol.*, 43, pp. 236-243.
- [8] Johal, M.S., and Dua, A., 1994, "SEM study of the scales of freshwater snakehead *Channa punctatus* (Bloch.) upon exposure to endosulfan," *Bull. Environ. Contam. Toxicol.*, 52, pp. 718-721.
- [9] Johal, M.S., and Dua, A., 1995, "Elemental lepidological and toxicological studies in *Channa punctatus* (Bloch.) upon exposure to an organochlorine pesticide, Endosulfan," *Bull. Environ. Contam. Toxicol.*, 55, pp. 916-921.
- [10] Johal, M.S., and Mehta, R., 1994, "Mineral composition of the scales of three exotic carps using energy dispersive x-ray microanalysis (E.D.X.) technique," *J. Aqua. Biol. Fish.*, 1(2), pp. 25-29.
- [11] Johal, M.S., and Sawhney, A.K., 1997, "Lepidontal alterations of the circuli on the scales of freshwater snakehead, *Channa punctatus* (Bloch.) upon exposure to Malathion," *Curr. Sci.*, 72, pp. 367-369.
- [12] Johal, M.S., and Sawhney, A.K., 1999, "Mineral profile of focal and lepidontal regions of the scale of *Channa punctatus* as pollution indicator," *Poll. Res.*, 18(3), pp. 285-287.
- [13] Johal, M.S., Tandon, K.K., Kaur, H., Sonia, and Sandhir, E., 1994, "Mineral composition of the different regions of the scales of two minnows using energy-dispersive X-ray microanalysis (EDX) technique," *J. Aqua. Biol. Fish.*, 1, pp. 1-4.
- [14] Johal, M.S., and Grewal, H., 2006, "Lepidological and toxicological studies on the scales of a freshwater snakehead, *Channa punctatus* (Bloch.) upon exposure to an insecticide carbaryl," *Poll. Res.*, 25, pp. 317-321.
- [15] Kishore, B., Bhatt, S.P., Rawat, U.S., and Nutiyal, P., 1998, "Stream regulation : variation in the density of benthic macro-invertebrate fauna of Ganga in lateral canal at Haridwar," *J. Hill. Res.*, 11(1), pp. 62-67.
- [16] Ladhar S.S., and Braich, O.S., 2005, "Biological diversity in wetlands of Punjab- A check list" Punjab State Council for Science and Technology, Chandigarh.
- [17] Lanzing, W.J.R., and Higginbotham, D.R., 1974, "Scanning microscopy of surface structures of *Tilapia mossambica* (Peters) scales," *J. Fish Biol.*, 6, pp. 307 -310.
- [18] Palaniappan, P.R., Vadivelu, M., and Vijaysundaram, V., 2009, "Fourier transform raman Spectroscopic analysis of lead exposed muscle tissue of *Catla catla*," *Roman. J. Biophys.*, 19, pp. 117-125.
- [19] Rashed, M.N., 1999, "Cadmium and lead levels in fish (*Tilapia nilotica*) tissues as biological indicator for lake water pollution," *Environ. Monit. Assess.*, 68, pp. 75-89.

- [20] Rauf, A., Javed, M., and Ubaidullah, M., 2009, "Heavy metal levels in three major carps (*Catla catla*, *Labeo rohita* and *Cirrhina mrigala*) from the river Ravi, Pakistan," Pak. Vet. J., 29, pp. 24-26.
- [21] Rishi, K.K., and Jain, M., 1998, "Effect of toxicity of cadmium on scale morphology in *Cyprinus carpio* (Cyprinidae)," Bull. Environ. Contam. Toxicol., 60, pp. 323-328.
- [22] Singh, H.R., and Nautiyal, P., 1990, "Altitudinal change and impact of municipal sewage on the community structure of macrobenthic insects in the torrential sector of the river Ganga in the Garwal Himalayas (India)," Acta Hydrobiol., 32(3-4), pp. 407-421.
- [23] Singh, P., and Saharan, J.P., 2010, "Elemental analysis of Satluj river water using EDXRF," Nat. Sci., 8, pp. 24-28.
- [24] Tandon, K.K., and Johal, M.S., 1993, "Mineral composition of different regions of the scale of an endangered fish – *Tor putitora* (Hamilton) using energy-dispersive X-ray microanalysis technique," Curr. Sci., 65, pp. 72-573.
- [25] Tort, L., and Torres, P., 1998, "The effects of sub lethal concentration of cadmium on hematological parameters in dog fish, *Scyliorhinus canicula*," J. Fish Biol., 32, pp. 277-282.
- [26] Trivedy, R.K., Shrotri, A.C., and Khatavkar, S.D., 1990, "Physico-chemical characteristics and phytoplankton of the river Panchganga near Kolhapur, Maharashtra. In R.K. Trivedy (ED.), Rivers of India. New Delhi: Ashish Publishing House.