Chapter – I

Introduction

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Limnology is the study of fresh and saline waters contained within the continental boundaries (Goldman and Horne, 1983). It is also referred as Hydro-biology or Aquatic biology. Hydrobiological study plays an important role in decision making processes for problems of dam construction, pollution and aquaculture practices. In order to utilize fresh water body successfully for fish production it is very important to study the biotic and abiotic factors.

The evaluation of the physico-chemical parameters and its annual fluctuations in aquatic ecosystem is very significant to determine species composition of fish.

Pollution of surface waters with sewage, combined liquid wastes discharged from all domestic, municipal and industrial sources is now very common. Composition of sewage vary considerably from place to place due to marked differences in the dietary habits of people, composition of trade wastes and water consumption. The required to oxide the whole organic matter and ammonia present in it. (Jingran J. L., 1991)

Domestic sewage have been reported to contain about 250 – 400 ppm of organic carbon and 80 – 120 ppm of total nitrogen, thus giving a C : N ratio of around 3 : 1 (Klein, 1962). Industrial sewage may contain much more organic carbon and hence may have a higher C : N ratio. Sewage present in present day civilization contains appreciable amounts of detergents.

Aquatic organisms are affected by pollutants both directly and indirectly in various ways. Following some of effects summarized by Zingran J. L. (1991).

Increase in osmotic pressure, violent alterations in the pH. A decrease in oxygen content of the water sometimes becoming a limiting factor. Specific toxic ingredients may injure the gills or other external structures, causing death either from anoxemia or by intake or absorption. The fish is affected indirectly when its habitual food organisms are destroyed.

Mechanical injury or blocking of gills from slit or other suspended material. Long distance migratory fishes during some phase of their life history may be adversely affected by highly localized pollution of the river and cumulative effects insecticides, pesticides etc.

Thus poisonous substances from various sources such as industrial waste, domestic and municipality waste, pesticides, detergents, chemicals fertilizers and agricultural drainage water reach to the river water have a fatal effects on aquatic ecosystem particularly on fish. Pollution in river water change the physical, physico-chemical properties of water, destroy the biological balance of self purification closely linked with bacteria algae and protozoans.

A brief review of the work on effects of water pollution due to anthropoid activities on histology, histochemical architecture especially distribution of mucosubstances and histopathological alterations in some of the vital organs of fish it is hoped, will provide basic information for discussion and comparison of the results obtained in the present investigation.

Hence this brief review of the literature with respect to each of the vital organs such as stomach, liver, kidney and gill is incorporated in the respective chapters.

Phosphate :

Phosphorus is the chemical element in the periodic table that has the symbol P and atomic number 15. Phosphorus is commonly found in inorganic phosphate rocks and in all living cells. The most important commercial use of phosphorus is in the production of fertilizers. It is also widely used in explosive, nerve agents, friction matches, fire works, pesticides, toothpaste, and detergents.

Phosphorus exists naturally in rocks. Phosphate rocks are an important source of phosphorus, which contains the mineral apatite. Rocks release phosphorus as they erode under normal weather conditions phosphorus enters freshwater systems in four main ways.

i) Atmospheric Source : Rain and dust

ii) Discrete source : Sewage treatment plants and industrial effluents.

iii) Diffuse sources : Storm water, agricultural and land clearing runoff.

iv) Non point sources : Sources within the water system, including washouts from river banks, re-suspension from sediments.

The rate at which phosphate loads enter fresh water systems varies with land use, geology, morphology of the drainage basin, soil productivity, human activities and pollution. Products such as laundry detergents used to be a large source phosphorus to freshwater systems.

In fresh water systems, phosphorus occurs in three forms –

i) Inorganic phosphorus

ii) Particles of organic phosphorus and

iii) Dissolved organic phosphorus

Aquatic algae and plants use an inorganic form of phosphorus for their nutrition. In most lakes and rivers, phosphorus is the primary nutrient that limits the growth of algae and plants. In most of the system the nutrient form of phosphorus is taken up very quickly hence it becomes difficult to measure phosphorus accurately.

Excessive phosphorus in a freshwater system increases plant and algal growth, which leads to changes in number and types of plant and animals, increases in animal growth and size, increases turbidity. Therefore more organic matter fall to the bottom of the system in the form of dead plants and animals and leading to losses of oxygen in the water. When there is no oxygen at the bottom of a freshwater system, phosphorus that previously had been locked in the sediment can be released back into the water. This is called internal loading which accelerates the problem of excessively high productivity.

Phosphorus is generally recognized as key nutrient in maintaining the productivity of water and phosphorus is one of the major nutrient responsible for biological productivity. The main supply of phosphorus in natural waters comes from weathering of phosphorus bearing rocks, leaching of soil of catchment area by rain, cattle dung and night soil (Jhingran, 1991). Other factors regulate the phosphorus content of the water bodies are the rate at which inorganic phosphate lost in the mud, bacterial activity, sewage contamination, agriculture fertilizers, pH of water and soil, aquatic vegetation phytoplankton etc.

There are several forms of phosphate which may be present in aquatic systems is soluble or particulate forms. These include orthophosphate poly or metaphosphates and organically bound phosphates. But 85% of total

phosphorus is present in organic form (organismal matter) only inorganic phosphorus (as orthophosphate) plays a dynamic role in an aquatic ecosystem.

The phosphorus more than 2 mg/l in open water gives sign of organic pollution. Phosphate can also interfere with water treatment processes, for example concentration as low as 0.2 mg/l interfere with the chemical coagulation of turbidity, Pomeroy, et. al., (1965).

Channa striatus (Bloch, 1793) :

Common Names :

Chevron snakehead, striped snakehead, banded snakehead, common snakehead. Soali (Pakistan); Murrel (India), Sohr, dekhu (Maharashtra, India), Shol (West Bengal), Pooli Kuchi, Koochinamarl (Karnataka), hal path maha, Lulla (Sri Lanka), tomam paya (Malaysia), Dalag, delak (Philippines)

Native range :

Most drainages of India, Pakistan, Southern Nepal, Sri Lanka, Bangladesh, Myanmar, Thailand, Southern China, Malaysia, Sumatra Borneo etc. This is an amazingly extensive 'native' distribution for an any freshwater fish, indicating that Channa striatus is quite probably a species complex. Native range lies between about 32^0N and 7^0 N indicative of a fish that is temperate to trophical.

Size :

It can attain a length of 30 – 36 cms in 1 year. (Bhatt, 1970). Talwar and Jhingran (1992) stated that this species is sexually mature at 30 cm, but added that 2 years were required to reach that size. In rivers of Uttar Pradesh, Chevron snakehead grow to 32 cm in 2 years.

Habitat :

Freshwater ponds and streams, usually in stagnant muddy waters, primarily found in plains in India (Talwar & Jhingran, 1992). It occurs in reservoirs but also exists in rivers, lakes swamps, rice paddies, mining pools, and roadside ditches (Mohsin & Ambat, 1983). In India, it can be found in reservoirs and rice paddies (Jhingran, 1984). Channa striatus is an obligate air breather. Breathing organ is developed in about 60 days during growth. The fish breathes aquatically until 18-20 days following hatching and thereafter becomes a bimodal breathers. Verma (1977) recorded a pH range for Channa striatus of 4.25 to 9.40 with 100% survival over 72 hrs and 90% survival at pH 3.10 for the same period.

Feeding habits :

Carnivorous, feeding on worms, prawns, frogs, and especially other fishes. Young fly feed on algae and protozons, juveniles feed on small crustaceans and adults are highly carnivorous, dreaded predators of other fishes. Jhingram (1984) cited larvae as feeding on insects, water fleas and fishfry. Juveniles preferring dipteran larvae, zooplankton and fishfry and adults as piscivorous.

Characters :

Gular region of head without patch of scales. Mouth large, lower jaw with 4 – 7 canines behind a single row of villiform teeth that wider to 6 rows at the jaw symphysis. Enlarged canine teeth are ideal for gripping, killing and tearing. Pectoral fin about half of head length. Dorsal fin with 37 – 40 rays, anal fin rays 23 – 29, pectoral rays 15 – 17, pelvic rays 6 and caudal fin rounded. Scale on top of head large with a rosette of head scales between orbits (Talwar & Jhingran, 1992). Colouration is quite variable. The dorsum is often dark brown to black, typically obscuring the chevron like markings dorsally.

PLATE 'M'

Photograph showing Channa striatus (Bloch.)

used in the present investigation

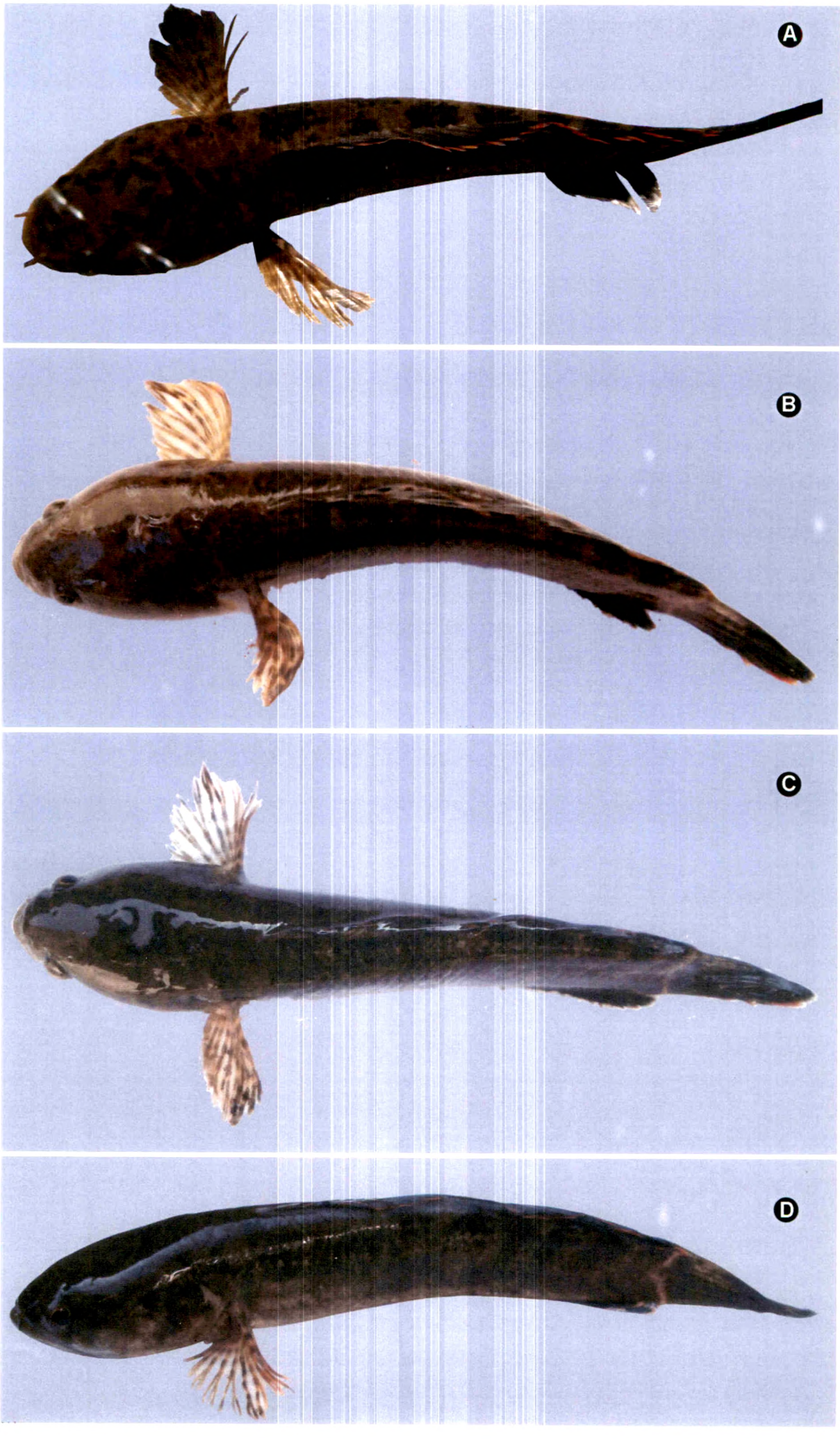
A) Dorsal view

B) Dorsal View

C) Dorsal View

D) Lateral View

PLATE NO. 'M'



Commercial importance :

Chevron snakehead is a popular and highly prized fish, widely distributed and the most economically important species of the genus Talwar & Jhingran (1992). Certain Channa species have been investigated or utilized as biological controls. Channa punctata, were used of different levels of pH and salinity in controlling mosquito larvae (Kehar, et. al., 1995). A more common use of snakehead in biological control has been as a predator in fish culture.

Milstein and Prein (1993) reported that Channa striatus are used to control overproduction of tilapias in the Phillippines. Wee (1982) also noted that they are also used in carp culture to control unwanted "Pest fish" in culture ponds and in turn they can be sol as products of culture.

Reasons for undertaking the present investigation :

A critical evaluation of the available literature on the fish toxicology, reveal that there are several avenues open for working in the field of histopathology and histochemistry of mucosubstances. Municipality sewage dumped in the river creates havoc and the life of common edible fishes like Channa striatus almost become impossible. Following are the reasons that led to the selection of present study.

1. River Krishna is lifeline of Satara district. It is the source of food in the form of number of fishes and drinking water for the people living surrounding it. Municipality sewage is dumped in the river Krishna near Satara city which causes pollution problems in the river. Hence to study the quality of water it was decided to undertake hydrological study of river Krishna.
2. In most of the studies where industrial waste pollutes the river the toxic pollutants are studied. The toxic chemicals such as pesticides, insecticides,

detergents, heavy metals polluting the river and their effects on common edible fishes are studied but less attention is paid to phosphate as a pollutant.

3. Channa striatus is the commonly available edible fish in river Krishna. The sewage water from Satara city pollutes the river Krishna and life of edible fish Channa striatus is put in danger. Hence it was decided to study this commonly available and effects of phosphate concentration on it.
4. The phosphate loads enter fresh water system depends on land use, geology, morphology of the drainage basin, soil productivity, human activities and pollution. Products like laundry detergents used to be a large source of phosphorus to fresh water system. Normally phosphate do not have detrimental effects on the fishes within normal limits. Now it is clear that 2 mg/l of phosphate acts as pollutant and the water near sewage entry have much higher values of phosphate that led to undertake this investigation.
5. Scanty work is being done on the phosphate toxicity. The toxic effect being phosphorus from pesticides and detergents have been studied recently on some fishes. No work is done on sewage derived inorganic nutrients such as phosphates especially common edible fish like Channa striatus.
6. A critical review of phosphate toxicity to the vital organs such as stomach, liver, kidney and gill shows that almost no work is done in this direction. Generally the large concentrations of phosphate in pollutants had profound effect on the architecture of the vital organs leadings to the death of a fish. Hence it will very useful in future of make efforts for survival of species in river Krishna.

7. Almost no research work at present is available on the effects of phosphate on histology and mucin histolchemistry of the common edible fish Channa striatus.

Hence knowing the importance of water quality of river Krishna and survival of fish species.

Based on above mentioned reasons and knowing the importance of water quality of river Krishna and survival of fish species the present investigation of phosphate toxicity and effects on vital organs such as stomach, liver, kidney and gills was undertaken.

Plan of the present work and presentation of the dissertation :

After reviewing and finding that meagre amount of work done especially phosphate effects of sewage toxicants on the vital organs of viz. stomach, liver, kidney and gill of Channa striatus, it was decided to work out histology, histopathology and characterization of mucosubstances and the effect of phosphate concentration on it.

a) Choice of the animal :

For present investigation the fish Channa striatus was selected. The fishes can be regularly collected locally from river Krishna and they are available throughout the year. These fishes are comparatively sturdy and were considered appropriate to study the effects of phosphate concentration on it.

b) Choice of the toxicant :

Sewage, industrial effluents, fertilizers, laundry detergents are the major sources of phosphate concentration. Locally available fishes from river Krishna are always subjected to dumping of sewage of Satara city in river

Krishna. Hence phosphate was selected to investigate its effects on histopathology and histochemistry of the stomach, liver, kidney and gills of Channa striatus.

c) Choice of techniques :

1. **Hydrological studies :** Well known methods for hydrological studies were used. Various physical and chemical parameters were observed through out the year. Studies on temperature, pH, transparency electrical conductivity, dissolved oxygen, free carbon dioxide, total alkalinity, total hardness, total dissolved solids and total suspended solids and phosphates were done.
2. **LC 50 :** The LC 50 valves were calculated by method given by Metelev et.al. (1971)
3. As the present investigation aims at details study of histology of stomach, liver, kidney and gills in normal fish and histopathological changes occurring in different cellular sites of different vital organs after exposure of the fish to different concentrations of phosphate, standard histological technique (H-E) was employed.
4. The present investigation also aims at a detailed histochemical study of mucosubstances in different cellular sites in the vital organs stomach, liver, kidney and gill. For the characterization of mucosubstances well established histochemical techniques were employed. These histochemical techniques are currently used to identify glycogen, neutral mucosubstances, acidic mucosubstances i.e. carboxy and sulfo mucins etc.

d. Critical evaluation of the observations :

It was decided to analyse critically the results obtained in the present investigation on the vital organs of fish Channa striatus in relation to -

1. Histology of the stomach, liver, kidney and gills.

2. Hydrological studies on physical and chemical parameters of the water from river Krishna from where the fishes were collected for study and sewage from Satara city is being dumped.
3. Histopathological effects of various phosphate concentrations on the vital organs of the fish.
4. Histochemical effects of various phosphate concentrations on the vital organs of the fish.
5. Histochemical studies include effect of phosphate concentration on the distribution of mucosubstances in the vital organs of the fish under study.
6. To compare results obtained in the present investigation and the existing literature on the other fishes and other toxicants so as to find out similarities or differences if any.
7. To find out cellular specializations if any in a given vital organ.
8. To project idea about the functional significance of mucosubstances after exposure to various concentrations of phosphate based on circumstantial evidences.

Presentation of the Dissertation :

The thesis of present investigation is divided into eight chapters. The first chapter is on introduction, reviews of literature on histology, histopathology and histochemistry of stomach, liver, kidney and gill of fish Channa striatus and related work. The reasons that stimulated to undertake the present investigation and plan of the proposed research work. The second chapter deals with material and methods employed in the present work. Chapter third is confined to hydrological observations of river Krishna abused with sewage dumping near Satara city. Chapter four, five, six and seven will include review of

research work histological, histopathological and histochemical observations on stomach, liver, kidney and gills respectively exposed to various concentrations of phosphate. Chapter eight is comprised of summary and concluding remark. The references cited in the various chapters will be summarized at the end of the dissertation as bibliography.