Chapter – VII

Histological, Histopathological and Histochemical Observations and Discussion on Gill of Channa Striatus (Bloch.) Exposed to various Concentrations of Phosphate

Chapter – VII

Histological, Histopathological and Histochemical Observations and Discussion on Gill of Channa Striatus (Bloch.) Exposed to various Concentrations of Phosphate

Gill :

Review of literature :

Gill is one of the vital organ which comes in direct contact of water and aquatic pollutants. Gills play an important role in respiration and osmo regulation. Exchange of gases takes place across the thin epithelium of the gills and it also regulates salts and water apart from excretion of nitrogenous remarkable sensitive index to toxicity.

Studies on the histopathological observations in gills are histopathological insecticidal and pesticidal pollution (Mahajan and Singh, 1973). The epithelial sloughing off gill lamellae has been observed in phenol by Mitrovic et. al., (1968) and in pesticides by Dalela et. al., (1979). Joining of respiratory folds of gill lamina resulting in reduction of respiratory folds has also been observed in C. carpio in aldrin toxicity (Ratnakar and Awasthy, 1979). The change in gill colour and mucus secretion in gills of T. mossambica were recorded under lindane toxicity by Murthy and Rao (1983).

Srivastava and Srivastava (1985) had published a series of papers on the effects of urea induced histopathological changes in gills of C. punctatus and C. mrigala. He observed hypertrophy of mucous cells, clumping and fusion of cells, separation of epithelial layer of secondary lamellae and fluid filled spaces.

Histopathological changes in the gill epithelial tissues in S. fontinalis including hypertrophy, excessive mucous secretion, extensive epithelial necrosis and sloughing of with increased pH stress (Daye and Garside, 1976). Histopathological and ultrastructural studies in cases of aflatoxin intoxicated fish indicated alterations in the gill tissue (Sahoo et. al., 2003). In aflatoxin treated rainbow trout, hyperplasia is seen and can also be caused by a variety of heavy metals, pesticides and phenols as well as high pH, heat stress and presence of microorganisms (Chevalier et. al., 1985).

The distillery effluents effects on B. stigma were observed including partial rupture and voluminous mucus secretion (Haniffa and Sundervadhanm, 1984).

From the review of literature it seemed that there are almost no studies related to phosphate induced histopathological changes in the gill of fresh water fishes and alterations in the mucosubstances.

Therefore a commonly found increased phosphate concentration due to sewage was selected for the present investigation to show its pathological effects on the histology and mucusubstance distribution and alterations in the gill of a fresh water fish C. striatus.

Histological observations on the control fish gill :

The gill of C. striatus comprised of no. of thin primary gill filaments attached to arch. Large no. of semi lunar projections on both dorsal and ventral sides of these primary filaments called as secondary gill lamellae (Plate No. 4, Fig. 1 and 2).

Each secondary lamellae consisted of middle vascular layer with covering of epithelial cells. The gill epithelium is primary made up of basal cells,

PLATE NO. 4

Histology, histopathology and histochemistry of Gill of control and experimental fish C. striatus (Bloch.)

Fig. 1 : T.S. of gill of control fish stained with H-E X 240

Fig. 2 : T.S. of gill of fish exposed to 0.01 M phosphate for 96 hrs. stained with H - E X 280.

Fig. 3 : T.S. of gill of control fish stained with H. E. X 280

Fig. 4 : T.S. of gill of fish exposed to 0.01 M phosphate for 96 hrs. stained with H. E. X 280.

Fig. 5 : T.S. of gill of control fish stained with AB pH 1.00 H - E X 280.

Fig. 6 : T.S. of gill of fish exposed to 0.01 M phosphate for 96 hrs. stained with AB pH-1 X 240.

Fig. 7 : T.S. of gill of kidney fish stained with AB pH-1 X 240.

Fig. 8 : T.S. of gill of exposed fish to 0.01 M phosphate for 96 hrs. stained with AB pH -1 X 240.

Abbreviations :

CAC - Cartilageneous cells

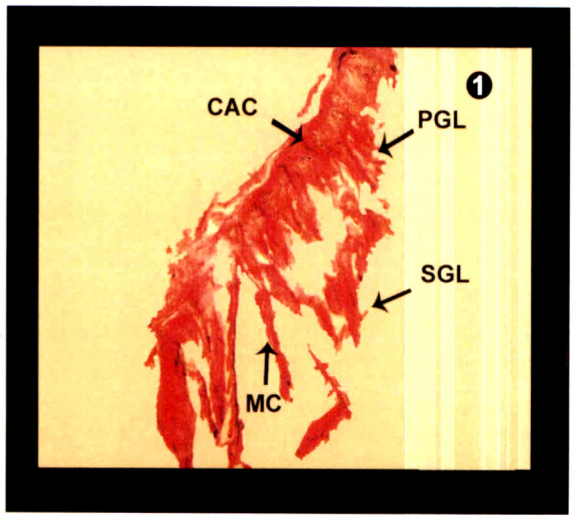
SGL - Secondary gill lamellae

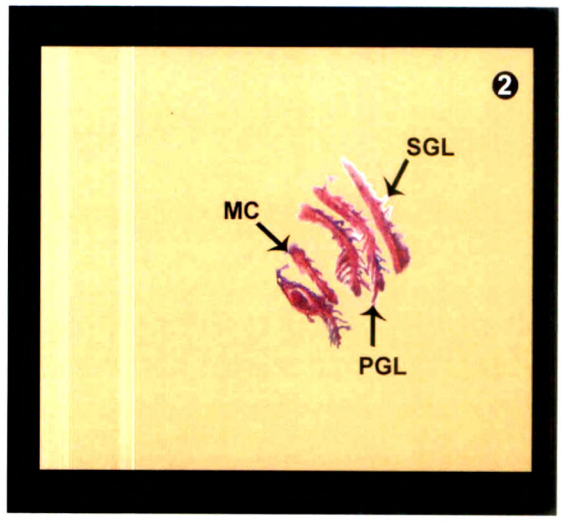
PC - Pillar Cells

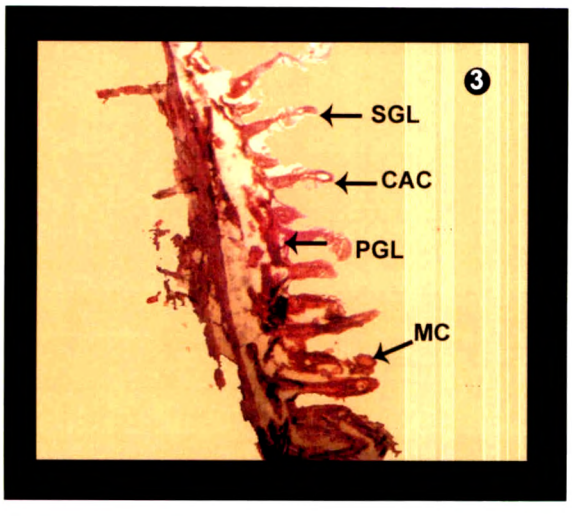
MC - Mucous Cells

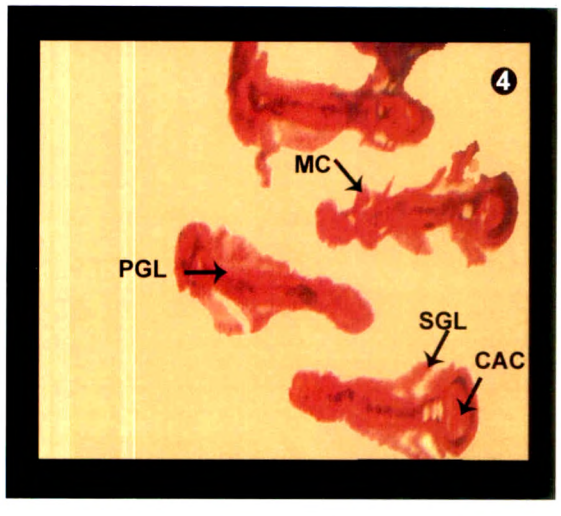
PGL - Primary gill lamellae

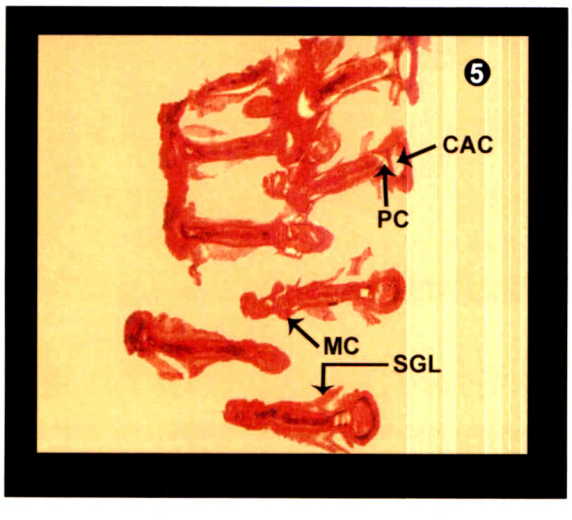
PLATE NO. 4

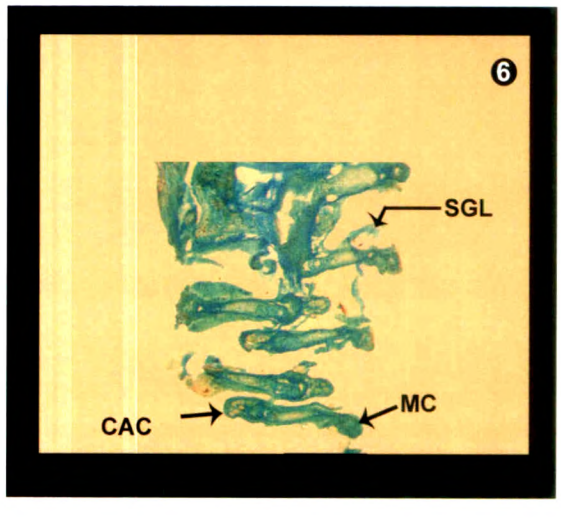

1
CAC
PGL
SGL
MC

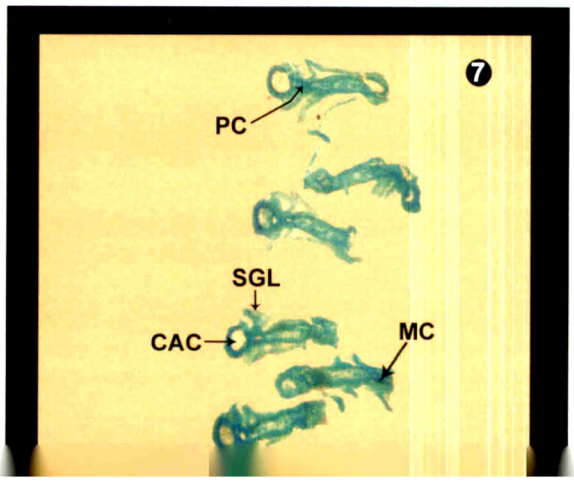

2
SGL
MC
PGL

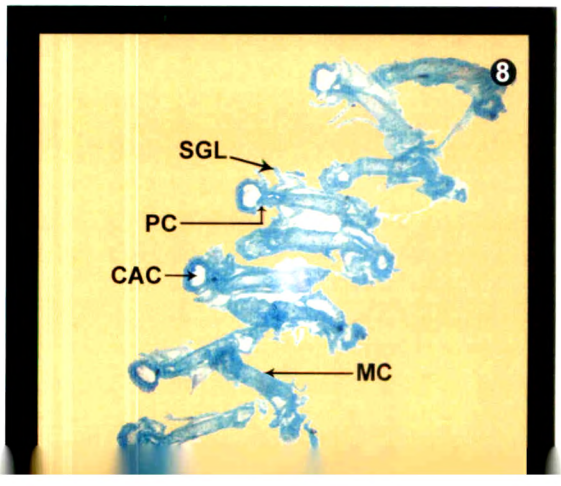

3
SGL
CAC
PGL
MC


4
MC
PGL
SGL
CAC


5
CAC
PC
MC
SGL


6
SGL
CAC
MC


7
PC
SGL
CAC
MC


8
SGL
PC
CAC
MC

pillar or supporting cells, mucous cells and chloride cells. The position of the pillar cells were fixed by the presence of cartiligenous substances on both the sides. Mucous cells were scattered through out the gill epithelium, mainly towards the tip and base of the secondary lamellae.

Histopathological alterations in the gills due to phosphate intoxication :

The histopathological alterations in the various tissues and cells in the gills due to phosphate intoxication are recorded in Plate No. 4, Fig. 1 and 2. The important histopathological alterations are described hereafter.

0.006 M Phosphate :

No much effect of this concentration is observed on the gills of C. striatus and fish appears control.

0.007 M Phosphate :

The gills exposed to 0.007 M phosphate intoxication showed minor hypertrophy and swelling of the epithelium. The microscopic picture revealed thickening and swelling of gill epithelium, particular mucus cells in the secondary gill lamellae. Interlamellar filament secreted large quantities of mucus.

0.01 M Phosphate :

Gill of C. striatus expose to 0.01 M phosphate for prolonged time showed prominent hypertrophy of gill epithelium along with swelling and thickening of epithelium. The mucous secreting cells on secondary gill lamellae and interlamellar filament epithelium also showed hypertrophy resulting in massive mucus secretion. Mucous cells exhibited enhanced stained affinity to eosin. The epithelial cells were enlarged in size with vacuolated cytoplasm and prominent nucleus (Plate No. 4, Fig. 2, 4, 6, 8).

0.015 M Phosphate :

0.015 M phosphate intoxication modified the architecture of gill by degeneration and flattening. The secondary lamellae as a whole including pillar cells have been dialated an fused together forming hollow spherical vacuoles at the tip cellular debris at the bases of secondary lamalles was observed. Degeneration of gill filament, cartilage cells, blood cells was seen. Mucous cells enlarged and were concentrated at the tips of secondary gill lamellae.

Histochemical observation in the gills due to phosphate intoxication :

Histochemical study of gill of C. striatus revealed that the gill cells can be differentiated in surface epithelial cells and mucous cells. The large amount of mucosubstances secretion three types of mucous cells can be identified. Histochemical observations recorded in Table No. 7.1.

Histochemical observations on the gill of control fish C. striatus :

The surface epithelial cells. These cells show moderate PAS activity and have no reactivity with ABpH-1 and ABpH-2.5 indicating presence of only neutral mucosubstances in them. Comparatively these cells are more in number.

Mucous Cells :

From the reactivities the mucous cells are of three types MC, MC_2 and MC_3 depending on the type of mucous secretion.

M_1 – Strongly sulfated mucins

M_2 – Sialomucins

M_3 – Sulfo-sialomucin

With the help of few methods employed here the types can be formed in broader sense.

Cartilage cells :

Cartilage cells show both reactivities of surface epithelial cells and type I mucous cells indicating presence of neutral and sulfomucins.

Histochemical alterations in the mucosubstance in the gill after intoxicaton with various concentrations of phosphate :

Overall pattern of mucosubstance indicated that surface epithelial cells contain moderate neutral mucins which was increased progressively increased in higher concentrations of phosphate and duration of exposure.

Similarly, mucous cells M_1 – secreting strongly sulfated mucins M_2 - producing sialomucins and M_3 – producing sulfosialomucins.

The mucous cells showed increased intensities of staining which progressively increased concentration of phosphate and prolonged exposure showed more secretion of mucosubstances.

Cartilage cells :

These cells show moderate to intense reactivities to all histochemical techniques employed. Indicating presence of neutral, sulfo and sialomucins and the secretion of the mucous remain constant through out the phosphate exposure.

Table No. 7.1

Histochemical observations on mucosubstances in the gill of freshwater fish

Channa striatus (Bloch.)

Sr. No.	Histochemical Techniques	Conc. of Phosphate	Generalized reactivities considering 24, 48, 72 and 96 hrs taken together				
			Surface epithelial cells	Mucous Cells			Cartiliage
				M1	M2	M3	
1	PAS	C	++P	++++P	+++P	++++P	+++P
		0.006 M	++P	++++P	+++P	++++P	+++P
		0.007 M	++P	++++P	+++P	++++P	+++P
		0.01 M	+++P	++++P	+++P	++++P	+++P
		0.015 M	+++P	++++P	++++P	++++P	+++P
2	AB pH 1	C	-	++++B	-	++++B	+++B
		0.006 M	-	++++B	-	++++B	+++B
		0.007 M	-	++++B	-	++++B	+++B
		0.01 M	-	++++B	-	++++B	+++B
		0.015 M	-	++++B	-	++++B	+++B
3	AB pH 1 - PAS	C	++P	++++B	+++P	++++BP	++BP
		0.006 M	++P	++++B	+++P	++++BP	++BP
		0.007 M	++P	++++B	+++P	++++BP	++BP
		0.01 M	+++P	++++B	+++P	++++BP	+++BP
		0.015 M	+++P	++++B	+++P	++++BP	+++BP
4	AB pH 2.5	C	-	+++B	+++B	+++B	+++B
		0.006 M	-	+++B	+++B	+++B	+++B
		0.007 M	-	+++B	+++B	+++B	+++B
		0.01 M	-	+++B	+++B	+++B	+++B
		0.015 M	-	+++B	+++B	+++B	+++B
5	AB pH 2.5 - PAS	C	++P	++++B	+++B	++++BP	+++BP
		0.006 M	++P	++++B	+++B	++++BP	+++BP
		0.007 M	+++P	++++B	+++B	++++BP	+++BP
		0.01 M	+++P	++++B	+++B	++++BP	+++BP
		0.015 M	++++P	++++B	++++B	++++BP	+++BP

Discussion :

In fishes the gill is the primary site of osomoregulation and respiration and also main target organ for aquatic toxicants (Woodword, et. al., 1983). Gill of C. striatus comprised of primary gill filaments attached to an arch. Large number of semilunar projections on both dorsal and ventral sides form the primary filaments. The gill epithelium is primary mode up of epithelial cells, pillar cells mucous cells and chloride cells. The histological structure observed in C. striatus is similar which other teleosten fishes. Cartilage cells can also be observed.

Histopathological alterations in response to the phosphate intoxication were evident. Severe hyperplasia, hypertrophy of epithelial cells, clubbing and fusion of secondary lamellae as well as oedema have been observed in the gills of channel cat fish, Ictalurns punctatus caged below the waste water treatment plant. Srivastava, et. al., (1998), Sinhaseni and Tespratap (1987) described marked swelling of secondary gill lamellae and hydropic vacuolation of epithelial cells following exposure to paraquat.

In the present study, after exposure to phosphate at different dose and time internal showed thickening and swelling of gill epithelium. Secondary gill lamellae and interlamellar filament showed hypertrophy resulting in massive mucus secretion. Epithelial cells enlarged in size with vacuolated cytoplasm.

Degeneration of gill filaments cartilage cells, blood cells was seen mucous cells enlarged and concentrated to the tip.

Fusion of lamellar epithelial lining of adjacent lamellae may be due to hyperosmotic external medium and through physiologically impaired epithelial cells (Rombough and Garside, 1977).

Similar histopathological alterations have been observed in the gills of fish exposed to different water pollutants (Mallat, 1985, Gill et. al., 1991, Srivastva et. al., 1998).

Histochemically in control fish surface epithelial cells contain only neutral mucosubstances. Mucous cells are of three types M1, M2 and M3 elaborating strongly sulfated, sialomucin and respectively. Cartilage cells indicate the presence of neutral and sulfomucins.

The gills exposed to phosphate intoxication showed that all the mucous producing cells increase in number and size producing voluminous amount of mucus. This is obviously protective act against the intoxication.