

ORIGINAL ARTICLE

IMPACT OF INDUSTRIAL EFFLUENT ON HISTOPATHOLOGICAL CHANGES IN KIDNEY  
TISSUE OF *ARIUS MACULATUS*

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ABSTRACT

The present study is carried out to observe the histopathological investigation in the kidney tissue of *Arius maculatus* exposed with sub lethal concentration industrial effluents collected from Cuddalore District. The histopathological changes were observed in the kidney tissues of *Arius maculatus* after exposure with industrial effluents (2% and 6%). The present study shows that shrinkage of glomeruli, Bowman's capsule, degeneration of epithelial cells of the renal tubules and vacuolization were observed in effluents treated fish, *Arius maculatus*

**Keywords:** Industrial effluents, Histopathology, Kidney, *Arius maculatus*

1. INTRODUCTION

The effluents released by various industries are causing a lot of problems and their disposal involves a complicated task. Many industries do not have proper facilities to treat the effluents and about 68.5 cubic million litres of industrial effluents are discharged as such into the environment (Natarajan, 1979). Alterations in the chemical composition of the aquatic environment usually affect behavioural and physiological activities of the inhabitants, particularly the fish population (O' Brien, 1967). In view of this, the need for toxicological testing has increased from 1970 and a standardization of this technique provides the usefulness, allowing the study of various aspects of acute toxicity (Stephen, 1982). The evaluation of acute toxicity test is worthwhile to know the levels, below which it may be considered safe for a particular toxicant in the aquatic environment.

The alarming increase in the volume of pollutants affect the living organisms and in particular pose a health hazard in humans. Toxic substances pollute the fishery reservoirs, sometimes causing heavy mortality of the fish population and their food organisms. Pandey *et al.* (1979) have reported that the biocides contain toxic substances alter the physiological conditions of the aquatic environment and destroy the utility and quality of the freshwater. The industrial effluents are constantly polluting the natural waters and their effects are manifold on living organisms including economically

important fishes. They are also responsible for various disturbances in physiological and biochemical parameters of fish (Shaffi, 1979). Industrial effluents containing different toxic substances also affect the aquatic system by interfering with their respiratory metabolism (Vaidyanathan *et al.*, 1995).  
Materials and methods

Fish *Arius maculatus* was collected from three different stations at Uppanar estuary and they were immediately transferred to ice box.

From the experimental station of Uppanar estuary, fish were caught either using cast net or hand lines. Cast net are made of nylon. The mesh varies from 10 to 15 mm. Hand lines with mono filament and hooks were used with prawn or mussel as bait for the capture of *Arius maculatus*. The fish were transported to the laboratory and maintained in the same way as the control fish collected from Perumal Lake. The water was renewed once in two days.

2. MATERIALS AND METHODS

Procurement of fish

Live specimens of *Arius maculatus* with an average length of  $8.5 \pm 0.50$  cm and weight of  $15.0 \pm 0.5$  g were collected from Uppanar brackish water by operating cast net. The fish were acclimatized in the aquaria of 120 litres capacity containing well aerated sea water (salinity 28 ‰; pH 7.69; oxygen content 4.32 mg/l) and water temperature (32.6°C) for a period of one week prior to experiment. During acclimatization, the fish were fed with chopped prawn and clams. Food was withheld one day before the commencement of the experiment. The water was changed along with waste feed and faecal matter

every 24 hours. Fish collected from Perumal lake (Plate 1A) were used as control and Uppanar brackishwater area was selected as experimental site (Plate 1B).

### Acute Toxicity Test

The raw and partially treated effluent was collected from the discharging point of industries surrounding the Uppanar estuary for acute toxicity test. In the acute toxicity bioassay, mortality could be observed within a short period.  $LC_{50}$  was calculated by the following method of Finney (1978) to observe mortality and behavioural response of the test fish, *Arius maculatus* on exposure to effluents of different concentrations.

Static acute toxicity was employed to evaluate the adverse effects of industrial effluents surrounding the Uppanar estuary on the fish, *Arius maculatus* under standardised laboratory conditions.

Food was withheld one day before the toxicity test with a view to avoid the possible change in the toxicity of the pollutants after addition of the effluent into the test tank with 100 litres of sea water having 10 fishes. Mortality was recorded after 24, 48, 72 and 96 hr and five replicates were maintained simultaneously for the purpose. Fishes showing respiratory and lack of response to tactile stimuli were considered, nearing dead and removed immediately. Percentage mortality was calculated and the values were subjected to Probit analysis (Reddy *et al.*, 1992). Confidential limits (upper and lower) of the Regression coefficient with Chi - square test were calculated.

### Design of sublethal toxic study

Sublethal studies are helpful to assess the response of the test organism to stress caused by the effluents. Based on acute toxicity test two sublethal concentrations (2% and 6%) on *Arius maculatus* were derived and used as the experimental concentrations. Sublethal of safe level concentration were derived from 96 hr  $LC_{50}$  value.

In the present study 2% (1/50) and 6% (1/30) dilution (in *Arius maculatus*) of the 96 hr  $LC_{50}$  were selected as sublethal concentrations. The experimental fish were exposed in each concentration for a period of 7, 15 and 30 days. A control batch corresponding to each test group was maintained simultaneously.

### Histological study

At an interval of 7, 15 and 30 days, a fish from each concentration (2% and 6%) of the effluent was sacrificed and the tissues of gill, liver, kidney, muscle and intestine were excised out and subjected to standard histological technique of Culling (1957). Each tissue was fixed separately in aqueous Bouin's fluid for histological examinations. After fixation, the tissue were dehydrated in ascending series of alcohol, cleared in methyl salicylate and embedded in paraffin wax. Serial sections were taken at 5 to 8  $\mu$  thickness and stained with Ehrlich's haematoxylin with aqueous eosin as the counterstain.

### 3.RESULTS

In the current pursuit on the effect of 2% concentration on the histology of kidney showed pathological manifestation only during 30 days of exposure while 7 days and 15 days exposure showed least probable variation in the renal histology. Nevertheless, dissolution of renal parenchyma was also observed at 7 days and 30 days (Plate 43-A and E). Dissolution or disassemblage of parenchyma with renal lesions characterized by oedema, excess shrinkage in glomeruli and fibrosis with excessive hyperemia were predominant and a partial necrosis was also common. The kidney exposed to 6% concentration showed marked variation. In the histology particularly in 15 and 30 days of exposure. Nevertheless, 7 days of exposure (Plate 43-B) showed least degree of histolysis with proliferating parenchyma and occasional enlargement of Bowman's capsule. Similarly 15 days of exposure revealed degeneration of epithelial cells of the renal tubules with marked vacuolization. A few of Bowman's capsules were damaged and appeared as large empty spaces and tubules of some places were devoid of epithelial cells. While 30 days of exposure (Plate 43-F) revealed scattering of renal parenchyma with occasional necrotic damage. The kidney exposed to field condition showed more histopathological manifestations and remarkable variation in the histology with damaged renal tubule, Bowman's capsule (necrotic Bowman's capsule) disorganised and distorted renal parenchyma and infected renal artery. Glomerulus was seen shattered as fragments with hyperplastic renal epithelium and varying degrees of degeneration and necrosis in the glomeruli, renal tubules and renal parenchyma were observed.

The mesonephric kidney of *Arius maculatus* consisted of number of uriniferous tubules, with a proximal glomerulus followed by shortened uriniferous tubules in the midst of the nephron and the bulk of the cortex was made of renal parenchyma. The nephrons arose in the cortex and then looped down in the interior for a variable distance and returned again into the cortex where it was continuous as the collecting duct descending into the interior.

The Bowman's capsules were held by renal parenchyma as intra Bowman's zones. Besides, in the Bowman's capsule there were lobular follicles or acinus with distinct lumen in the centre and it resembled a brush border of the urinary tubule. The Bowman's capsule consisted of a single layer of flattened cells (numbering 40) resting on a basement membrane and it formed the distended part of urinary tubule. The Bowman's capsule enclosed a densely packed network of anatomizing capillaries which invaginated into Bowman's capsule with the glomerulus being invested with a layer of epithelial cells called podocytes. The renal tubule extended from the Bowman's capsule to its junction with the end of the renal tubule or collecting tubule. The renal tubule was lined by a single layer of epithelial cells and concerned with selective re-absorption. Each kidney showed a single renal artery which was divided into 2 main branches, each of those gave rise to several interlobular arteries which ascended between the cone like zones of cortico - medullary junction. The interlobular arteries gave rise to numerous radial arteries which radiated towards the arterioles of the glomeruli. In the cortex numerous reno - corpuscles were visible at that magnification. Most of the cortical parenchyma surrounding the renal corpuscles consisted of different parts of renal tubules including collecting ducts which projected towards the tip of the renal papilla (Plate 42-A).

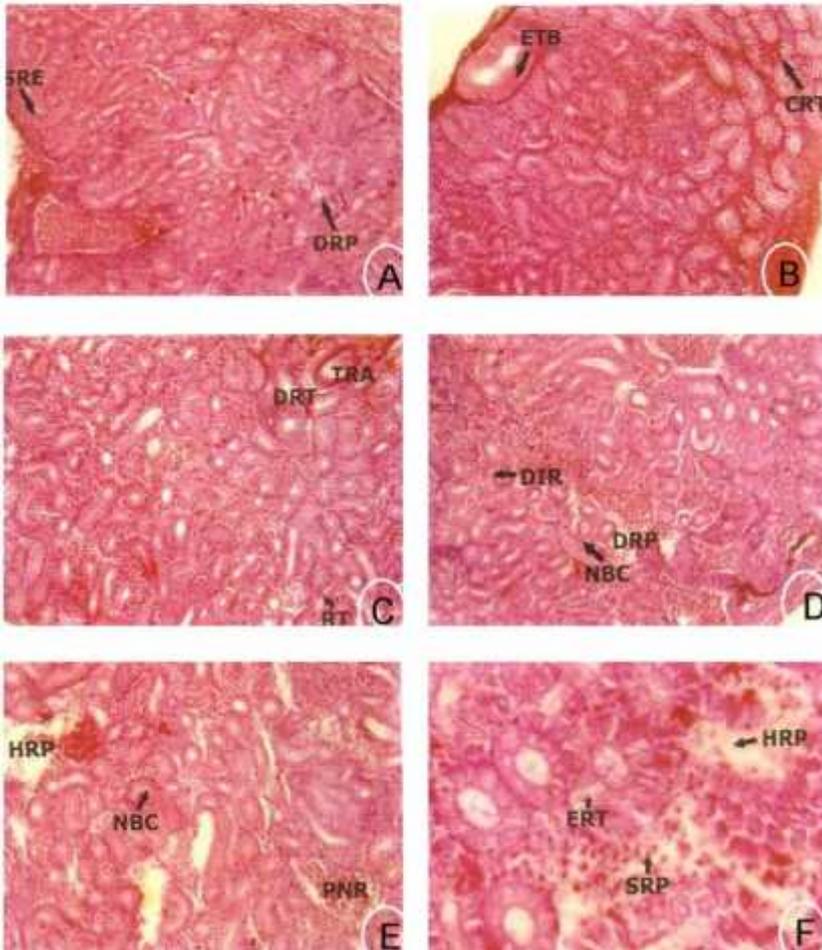
Fig.1 shows the kidney tissue of control fish



A: Light micrograph of the kidney of control fish *Arius maculatus* showing the histology (450X)

Plate 2 shows the kidney tissue of treated fish

Light micrographs on the histopathology of the kidney *Arius maculatus* exposed to 2% (A, C & E) and 6% (B, D & F) sublethal concentrations of the effluent for an exposure period of 7 days (A & B) 15 days (C & D) and 30 days (E & F) (180X)



## 4. DISCUSSION

Some of the most heavily industrialized areas of the world are situated near the estuaries which are particularly at risk from metallic infusion. Industrial discharges containing toxic and hazardous substances, including heavy metals (Woodling *et al.*, 2001) contribute tremendous to the pollution of aquatic ecosystem. Hence, the problem of toxicity in marine organisms from natural causes or manmade pollutants is coming into critical focus.

The pathological changes induced by the effluent on the kidney of *Arius maculatus* includes haemolysis of erythrocytes, karyolysis, rupture of cells, pyknotic nuclei, clumping of erythrocytes, tubular necrosis, and distortion of inner wall of tubules. The evidence of nephrotoxicity is indicated by changes like lesions, pyknotic nuclei, microvacuole degeneration, shrinkage of glomeruli, swelling and necrosis of the tubular epithelium in the second proximal tubule.

Sriwastawa *et al.* (1982), Karuppasamy (2000c) observed dilation of the renal tubules, cloudy swelling of the glomerular epithelial cells and hydropic degeneration in some of the tubular epithelial cells in mercury treated fish.

The most striking effect in kidney of metal treated fish was the enlargement of the lumen in the proximal and distal tubules due to hypertrophy. Disruption of the tubular cells resulted as the kidney had been induced to function actively under pollutional stress to eliminate the toxicants. Hyperplasia is seen as the defence mechanism of the fish to eliminate the toxicants.

There are five types of pollutants that affect the estuarine and coastal resources as under: (i) bacterial infection due to the discharge of untreated domestic sewage along with storm water runoff from the cities which can cause epidemics of water borne diseases such as dysentery, typhoid, cholera, poliomyelitis and hepatitis (Custodio, 2010); (ii) industrial wastes that deplete dissolved oxygen (Mondal and Singh, 2011); (iii) toxic chemicals of industrial wastes and land runoff comprising, pesticides and herbicides (Sankaran *et al.*, 2009) interfering with the metabolism; (iv) fertilizer's runoff that tend to stimulate growth of some life forms and cause eutrophication (Mondal *et al.*, 2011) and inert chemical sediments and counteracting with the delicate benthos of the estuary (Sankar *et al.*, 2010).

The priority list of pollutants compiled by the Environmental Protection Agency of United States contains the eight more widespread heavy metals such as arsenic, chromium, cadmium, copper, lead, mercury, nickel and zinc (Moore and Ramamoorthy, 1984), of which the last six are generally known as "toxic heavy metals" (Dara, 1997), while cobalt

manganese, molybdenum and selenium as "essential heavy metals." According to George (1987) aluminium, arsenic, antimony, cadmium, chromium, copper, iron, manganese, mercury, lead, uranium and zinc are more toxic to the aquatic system.

Sastry and Sharma, (1979) observed that the histopathological effect of arsenic on kidney tissue were enlargement of kidney tubules and damaged tubular epithelial cells in *Channa punctatus* treated with endrin. Similar histological damages in kidney tissue have been reported in *Ictalurus punctatus* exposed to methyl mercury (Kendall, 1975). Saxena and Sarin (1981) suggested that necrosis and damage of kidney tubules were mainly due to physiological response to increased excretory demands in cadmium treated *C. punctatus*.

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