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Histopathological Changes in the Gill of *Labeo rohita* (Hamilton) Fingerlings Exposed to Atrazine

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Abstract: Atrazine (2-chloro-4-ethylamino-6-isopropylamino 1,3,5-triazine) is a widely used selective herbicide. Due to its persistence, it is present in the surface waters, contaminating non-target organisms such as fish. Herbicide atrazine was administered to *Labeo rohita* fingerlings in 120 hours. The used dose of atrazine was 0.18 mg/l for 120 hours (LC_0). The histopathological changes in the gill tissue like epithelial hyperplasia, curling of secondary lamellae and changes in chloride cells, besides these changes pyknotic nuclei, vacuolization, degradation of epithelial cells and pillar cells, were noticed.

Key words: Histopathological changes · *Labeo rohita* · Gill · Atrazine

INTRODUCTION

Atrazine (2-chloro-4-ethylamino-6-isopropylamino 1, 3, 5-triazine) is a white crystalline solid synthetic herbicide that is used as to kill broad leaf weeds in agricultural and roadway applications [1, 2]. In freshwater invertebrates atrazine has been found to affect hydromineral balance and gill function in crabs[3]. It can be detected in nearly every surface water system in the world. Fish are very susceptible to bioaccumulation in their fatty tissues, as they take up lindane residues from the water through the gills and skin [4]. The exposure to chemical contaminants can induce a number of lesions and injuries to different fish organs suitable for histopathological examination in searching for damages to tissue and cells [5, 6]. Although damages have been observed in the gill arches, liver, kidney, olfactory epithelium and nervous system[6].

In fish, gill are vital organs for their respiratory and osmoregulatory functions. Respiratory distress is one of the early symptoms of pesticide intoxication [4]. Considerable interest has been shown in recent years in histopathological study while conducting sub-lethal tests in fish. Tissue changes in test organisms exposed to a sub-lethal concentration of toxicant are a functional response of organisms which provides information on the nature of the toxicant. Histological changes associated with pesticides in fish have been studied by many authors [7-9]. In the present investigation was an attempt to observe the histological alterations in the gill of *Labeo rohita* exposed to atrazine.

MATERIALS AND METHODS

Healthy *Labeo rohita* fingerlings were procured from the freshwater form located in Puthur, Nagappattinam district. They were acclimatized for a maximum period of 15 days in the laboratory condition. The fingerlings each measuring 4.5 to 6.0 cm in length 5-6 g in weight are used. They were exposed to sublethal concentration of atrazine 0.18 mg/l for a period of 120 hrs. The gill pathology was examined after 120 hrs. An equal number of control fish were examined at similar intervals for histological comparison. The gill was removed and fixed in Bourin's fluid and then processed for microtome sectioning at 8µm and stained with haematoxylin, eosin and mounted in DPX.

RESULTS AND DISCUSSION

The gill is made up of filaments of primary lamellae arranged in double rows. Secondary lamellae arise from these filaments. The secondary lamellae are lined by a squamous epithelium. Histopathologcal changes in the gill of fishes due to pesticides and other contaminants have been reported earlier[10].

In the present study the observed changes are excessive mucus secretion, lifting up of the epithelium and lamellar fusion. The lifting of the epithelium increases the distance through which the toxicant has to travel to reach the blood stream (Fig. B, C). No recognizable changes were observed in the gills of the control fish. Each gill

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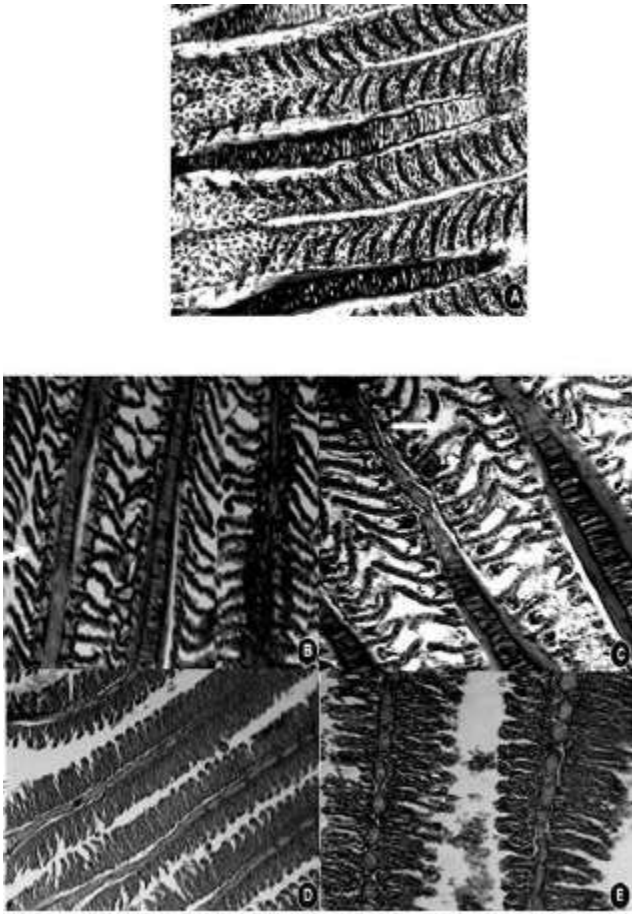

Fig. 1: A. Gills from control fish showing primary lamellae and secondary lamellae arising from these, parallel with them and perpendicular to the filament arcis (H and E × 400). B and C. Shortening of secondary lamellae and edematous separation of the epithelial layer (→) with necrotic cells occupying in the interlamelar space (H and E × 400). D and E. Swelling of the epithelium and fusion of secondary lamellae, hyperplasia, epithelial living in the base and tips of secondary lamellae (H and E × 400).

consisted of a primary filament and secondary lamellae (Fig. A). The primary lamellar epithelium was one or two cell layers thick. Chloride cells were visible along the primary lamellar epithelium especially at the bases of secondary lamellae. Since the gills are the primary route for the entry of pesticide. According to Leino *et al.* [11], the gill of pearl dace and fathead minnows from environmentally polluted Canadian lakes exhibited various cellular, histological and histopathological changes, which may contribute to problems related to respiration and acid-base balances. The severe damage in rupture of the gill epithelium resulted in hypoxia and respiratory failure. In addition, the fish showed problems in relation to ionic acid and base balance. Ruptures of the gill epithelium observed are direct responses to the action of atrazine. Roy and Datta, [12] reported that slight hyperplasia of gill epithelium in *Pineaus monodon* exposed to gusathion, a commonly used organophosphate. They further reported that inflammatory alterations of lamellar epithelium and hyperplasia were reported in the gills of freshwater major carp *Cirrhinus mrigala* (Hamilton) during 48 hrs exposure to sublethal dose of malathion.

In the present investigation freshwater fish *Labeo rohita* (Hamilton) exposed to sublethal concentrations of atrazine to hyperplasia of gill filaments, fusion of gill filaments due to separation, necrosis of gill epithelium, degeneration of pillar cells and the development of vacuoles in the epithelium are the prominent pathological changes (Fig. D and, E). The damage of gill of fish exposed to the sublethal concentrations were severe. Shortend and clubbing of

ends of the secondary gill lamellae, fusion of adjacent secondary gill lamellae and necrosis in the primary lamellae were well marked. Hyperplasia and hypertrophy of nuclei were also seen. Besides there changes pyknotic nuclei, vacuolization and degeneration of epithelial cells and pillar cells and lifting of the epithelial layer from the secondary lamellae were also significant (Fig. D and E).

Similar changes were observed in freshwater fish exposed to sublethal concentrations of monocrotophos to fenvalerate in *Labeo rohita* by Tilak *et al.*[13] and to cypermethrin in *Labeo rohita* Veeraiah, [14].

In fish exposed to 0.18 mg/l atrazine concentration (120 hours LC_0) in the present study the major changes in gills were edema, epithelial lifting, thickening of the primary lamellar epithelium and fusion of secondary lamellae. (Fig. D and E). Histopathological changes of gills such as hyperplasia and hypertrophy, epithelial lifting, aneurysm and increase in mucus secretion have been reported after the exposure of fish to a variety of noxious agents in the water, such as pesticides and phenols Nowak [15]. All these lesions may impair respiratory function. Filament cell proliferation and lamellar pavement cell hypertrophy reduce the inter lamellar space and may cause a complete lamellar fusion reducing the total surface area for gas exchange Nowak [15]. Otherwise, they increase in the distance of the water-blood barrier, which together with epithelial lifting and the increase in mucus secretion may drastically reduce the O_2 uptake and if the damping agent is not removed, can lead to the rupture of blood vessels with small haemorrhage focus.

In the present study gill histological changes have been related to atrazine concentration. It can be concluded that gill alterations as a result of herbicide exposition of fish may serve as a sensitive biomarker for the toxicity of sublethal concentrations of herbicides as well as other pollutants. However, complementary studies are necessary for a better understanding of its deleterious effects.

REFERENCES

1. USEPA [U.S. Environmental Protection Agency], 2002. Registration eligibility science chapter for atrazine environmental fate and effects chapter. Washington, DC, USA.
2. U.S. Center for Disease Control Agency for Toxic substances and Disease Registry (ATSDR), 2003.
3. Warining, C.P. and A. Moore, 2004. The effect of atrazine on Attantic Salmons (Salmo Salar) smolts in freshwater and after sea water transfer Aquatic Toxicol., 66: 93-104.
4. Ortiz, J.B., M.L. Gonzalez de Canales and C. Sarasquete, 2002. Histological alterations in different tissues of fishes under the impact of persistent chemical pollution. Ecotoxicol. Environ. Restor., 54: 45-52.
5. Rabitto, I.S., J.R.M.A. Costa, H.C. Silva de Assis, M.A.F. Randi, F.M. Akaishi, E. Pelletior, C.A. Oliveira Ribeiro, 2005. Dietary Pb(II) and TBT (tributyltin) exposures to neotropical fish *Hoplias mala*.
6. Oliveira Ribeiro, C.A., C. Rouleau, E. Pelletier, C. Audet, H. Tjalve, 1999. Distribution kinetics of dietary methyl mercury in the *Artic charr* (Salvelinus alpinus). Environ. Sci. Technol., 33: 902-907.
8. Bruno, D.W. and A.E. Ellis, 1988. Histological effects in Atlantic salmon, *Salmo salar* L. attributed to the use of tributiltin antifoliant. Aquaculture, 2: 14-20.
9. Mallatt, J., 1985. Fish gill structural changes induced by toxicants and other irritants. A statistical review. Can. J. Fish. Aquat. Sci., 42: 630-648.
10. Dutta, H.M., S. Adhikari, N.K. Singh, P.K. Roy and J.S. Munshi, 1993. Histopathological changes induced by malathion in the liver of freshwater catfish, *Heteropneuster fossilis* (Bloch.). Bull. Environ. Contam. Toxicol., 51: 895-900.
11. Leino, R.L., P. Wilkinson and J.G. Anderson, 1987. Histopathological changes in the gills of pearl dace, semotilus margarita and fathered minuows, pimephales promelas, from experimentally acidified Canadian lakes. Can. J. Fish Aqucat. Sci., 44: 126-134.
12. Roy, P.K. and J.S. Datta Munshi, 1991. Malathion induced structural and morphometric changes of gills of a freshwater major carp *Cirrhinus mrigala* (Ham.). J. Environ. Biol., 12(1): 79-87.
13. Tilak, K.S., K. Veeraiah and K. Yacobu, 2001. Studies of histopathological chanes in the gill, liver and kidney of *Ctenopharyngodon idellus* (Valenciennes) exposed to technical fenvalerate and EC20%. Poll. Res., 20(3): 387-393.
14. Veeraiah, K., 2001. Cypermethrin toxicity and its impact on histochemical and histological changes in the Indian Major Crop *Labeo rohita* (Hamilton).
15. Nowak, B., 1992. Changes in the gills induced by residues of endosulfan. Aquat. Toxicol., 23: 65-84.