IJCRT.ORG ISSN : 2320-2882

INTERNATIONAL JOURNAL OF CREATIVE RESEARCH THOUGHTS (IJCRT)

An International Open Access, Peer-reviewed, Refereed Journal

Histological Changes In Digestive Gland Of Freshwater Bivalve Mollusc *Lamellidens Marginalis* Exposed To Tributyltin Chloride In Summer Season

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Abstract:

Freshwater bivalve, Lamellidens marginalis exposed to 3.5ppm, 2.5ppm, 1.8ppm and 1.0ppm LC_{50} concentration at 24 to 96 hrs of periods of intervals. In experimental digestive tubules, channels, and connective tissue of the gland various changes were recorded. Something degenerative changes observed in control groups of bivalves. From this study we concluded that toxicity of TBTCL was responsible for histological changes in freshwater bivalve, Lamellidens marginalis.

Keywords*: Histology, Lamellidens marginalis, Digestive gland, tributyltin chloride, acute toxicity, Summer Season.*

INTRODUCTION:

All metals are, however, toxic to aquatic organisms when present at elevated level, causing direct or indirect effects such as histological damage, or a reduction in the survival, growth and reproduction of the species it influences (Heath, 1987). Biomarkers are important tools in the detection of various stresses in aquatic species. Stresses may include chemicals, metals and other environmental pollutants. Biomarkers are indicators that can be used to assess the effects of these disturbances (world health organization, Geneva 1993).

The majority of environmental organotin pollution is from biocidal antifouling paints that reach TBT to protect ship hulls from algal and mollusc growth. Nevertheless, organotins enter both freshwater and marine environments through treated woods, run-off from landfill, sewage and industrial discharges (Fent, 1996; O'Halloran *et al.*, 1998a). Once in the aquatic environment, they are bioaccumulated and biomagnified by aquatic invertebrates and vertebrates and can reach extremely high levels in the tissues of these organisms (Focardi *et al.*, 1999; Tsuda*et al.*, 1988).

TBT affects the *O. gigas* was first noticed by Alzieu and Heral, (1984). Bruno and Ellis, (1988) observed histopathological changes in different tissues of Atlantic salmon, *Salmo salar*,, attributed to use of tributyltin antifoulant. Sarojini *et al.*, (1989) worked on histological changes in gills and ovary of prawn, *C. rajadhari* exposed to TBTO.

Lipofuscin granules are being used as a biomarker of cellular stress in association with lysosomal alterations in the digestive gland of bivalves, and have proved to be an efficient biomarker (Au, 2004; Zorita *et al.*, 2006). The digestive cells of the tubules are primary organ of endocytic absorption and intracellular digestion. In gastropods and bivalves; the digestive gland is the major site of heavy metal storage (Simkiss and Moson., 1983, Pozzi and Merlini, 1977). Reduction in epithelial thickness of the digestive gland of gastropods and bivalves was proposed by some authors as indicator of environmental quality assessment (Tripp *et al.*, 1984, Marigomez*et al.*, 1990). Usheva *et al.*,(2006) worked on histopathology of the digestive gland of bivalve molluscs, *C. grayanus*.Kharat, (2007) observed the changes in digestive gland of freshwater prawn, *Macrobrachiumkistnensis* exposed to organotin tributyltin chloride. Songyot et al., (2016): observed Histopathologically changes due to the effects of pulp and paper mill effluent on the digestive glands of river

snail, *Filopaludina martensi*. Ustina et al., (2018) worked on Histological and ultrastructural alternations in the digestive gland of the Egyptian slug, *Limax maximus* treated with botanic molluscicidal thymol.

Bivalves are filter-feeders and thus uptake heavy elements not only from food and water but also from ingestion of inorganic particulate materials (Ei-Sikaily et al., 2004). Moreover, they have been well established as bioindicators for monitoring the concentration of heavy trace metals in many areas in the world (Neuberger-Cywiak et al., 2003).There is paucity of literature available on the histopathological effect of tributyltin compounds on the freshwater bivalve species, vital tissues digestive gland.Thus in the present investigation attempt has been made to study the effect of tributyltin chloride in digestive glands of freshwater bivalve, *L. marginalis*exposed up to 96hrs. in summer.

MATERIAL AND METHODS:

The freshwater bivalves, *Lamellidens marginalis* were collected from the Godavari river at Paithan. The site of the collection is 45Km. away from Aurangabad city, of Maharashtra state. They were collected and acclimatize to laboratory conditions, they were kept in a plastic troughs containing water and acclimatize to laboratory condition for 3 to 4 days. 1ppm stock solution of tributyltin chloride was prepared in acetone Laughlin *et al.,* (1983). After the acclimatization, healthy medium sized bivalves were selected for experiments.

For each experiment 10 animals of approximately similar size were exposed.In summer the LC50 values for 24, 48, 72 and 96 hoursfound to be 3.5ppm, 2.5ppm, 1.8ppm and 1.0 ppmrespectively. Bivalves in each experimental group were sacrificed and their tissues digestive glands were dissected out from control and experimental bivalves.Tissues were fixed in Bouins hollande. After fixation tissues were washed in running tap water, so as to remove the Bouins hollande from tissues. The washed tissues were dehydrated in different grades of alcohol (from 30% to absolute alcohol). The cleared tissues were embedded in paraffin wax (58 to 60^0c) and blocks were prepared.

Blocks of the tissues were treamed and serial sections of 8µ thickness were cut with the help of microtome. Sections were spread properly on the slides, and were stained with Mallory's triple stain. The stained sections were examined under light microscope for histological effect of tributyltin chloride.

OBSERVATIONS AND RESULTS:

The freshwater bivalve, *Lamellidens marginalis* were exposed to acute concentrations of organotin tributyltin chloride up to 96hours to observe the histological changes in digestive glands.

Histopathological changes of hepatopancreas in summer season:

In summer, the digestive gland in bivalves belonging to control group showed few degenerative changes indicated by loss of cytoplasmic density of secretory and digestive cells. Digestive diverticula's were flattened and some digestive tubules line by strong basophilic cells. Decrease in height of epithelial digestive diverticula's(Plate-1. fig-1).

The *L. marginalis*were exposed to tributyltin chloride in summer at 3.5ppm, 2.5ppm, 1.8ppm and 1.0ppm for 24, 48 72 and 96 hours respectively. Results were compared with control group and illustrated histopathological changes

The basement membrane ruptured and necrosis was observed in both the type of cells which were mostly detached from basement membrane. Picnoid nuclei were observed and few were released from the cells. The high vacuolization and necrosis were observed in the epithelial wall of digestive diverticula. Cuboidal cell present in large unstained empty vacuoles and dilated lumen. Degenerative changes were indicated by loss of cytoplasmic density and vacuolization in all types of cells. Advanced necrotic changes were evident in 96hours exposure (Plate-1. fig-2, 3, 4 and 5).

PLATE - 01

Histological changes in digestive gland due to tributyltin chloride (TBTCl) stress in *Lamellidensmarginalis*during summer season.

Fig. 1- Control

Fig. 2 - Experimental (24Hrs.)

Fig. 3 - Experimental (48Hrs.)

Fig. 4 - Experimental (72Hrs.)

Fig. 5 - Experimental (96Hrs.)

CT = Connective tissue

DC = Digestive cell

SC = Secretory cell

A = Amoebocytes

W = Wall of the tubule

L = Lumen of the tubule

DW = Degenerative wall of the tubule

Photo plate -01

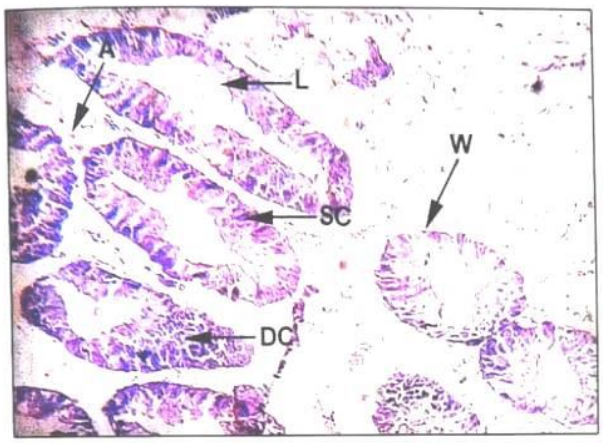

Fig - 1 CONTROL

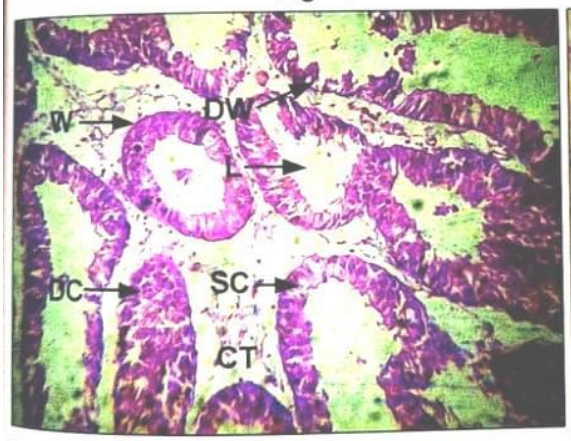

Fig - 2 24 Hrs

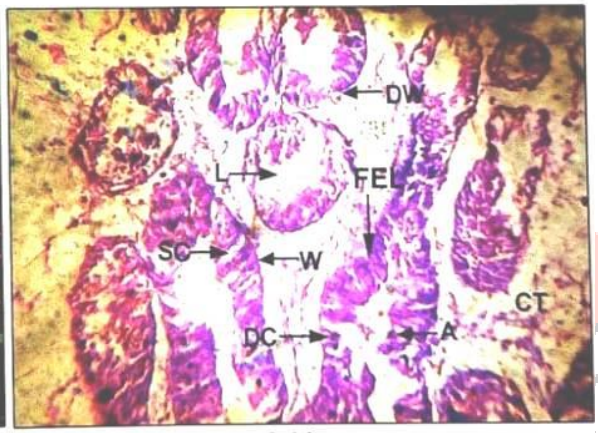

Fig - 3 48 Hrs

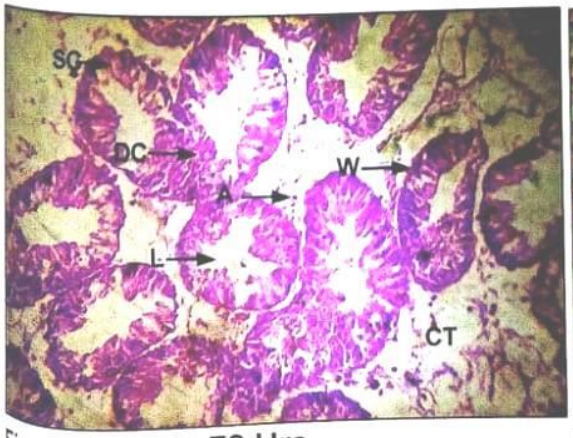

Fig - 4 72 Hrs

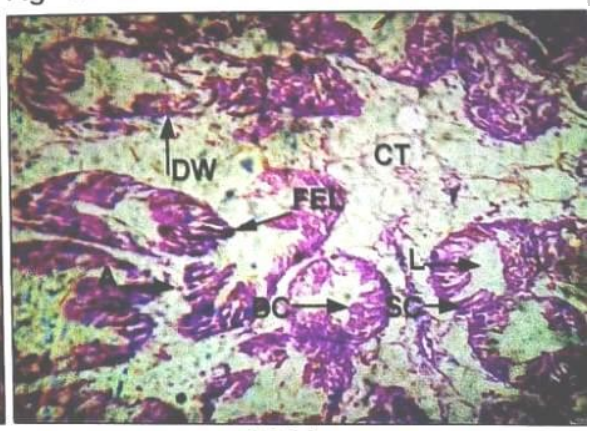

Fig - 5 96 Hrs

DISCUSSION AND CONCLUSION:

L. marginalis were exposed to tributyltin chloride in summer at 3.5ppm, 2.5ppm, 1.8ppm and 1.0 ppm for 24, 48, 72 and 96 hours respectively. Severity of damage was found to be more in 96hours,as compare to 72, 48, 24hours exposures.

In the present study, it is revealed that the initial impact of the antifouling organometallic compounds exposed for 24hours is less when compared with other exposure periods. The 48 hours exposure to TBT shows that damage caused is at a higher rate to the tissue structure as it metabolizes into the tissues. After 72 hours exposure the damage is still there and not so high which might be due to the adaptation of the tissues to the pollutant and development of resistance to some extent. At 96 hours exposure the damage was increased which may be due to the lost of resistance of power of the tissues. This may be either due to the defense mechanism of cell becoming weak, or due to the high accumulation of the pollutant. The degree of toxic effect depends mainly on levels of pollutant and metabolites in the target tissues. The effect of tributyltin chloride may be highest, because obviously high temp in summer, decreasein oxygen content, low food availability and in this situation toxicity of tributyltin chloride is very high.

Although the observed histopathological changes can not be linked with the relatively high mortalities observed during exposure and probably due to spawning, their severity appears to be both dose and time dependent. Furthermore, the data acquired suggest that poisoning results from different mechanisms. Although there are few studies devoted to histopathological effects of pollutants in molluscan species, digestive diverticula's modifications such as intensive fragmentation, vacuolization, epithelial thinning have been noted (Tripp *et al.*, 1984; Couch, 1984; Rasmussen *et al.*, 1985). Such modifications could be considered as a general molluscan response to stress (Moore *et al.*, 1979; Lowe *et al.*, 1981) and have been interpreted as a physiological survival mechanism of bivalves subjected to stress (Moore *et al.*, 1979; Henry, 1987). Kharat, (2007) observed the changes in digestive gland of freshwater prawn, *Macrobrachiumkistnensis*, there was reduced the size of lumen, less secretory globules compare to control and epithelial lining ruptured, after exposure to tributyltin chloride.

Kumar et al., (2011): showed alterations indicate hyperactivity reactions of the hepatopancreas in fresh water mussels to combat the stress of dimethoate exposure. Impact of mercury on hepatopancreas of fresh water bivalve, *Lamellidens marginalis* showed swelling of the tubules, which was distinct from the connective tissue observed by Suryawanshi et al., (2020). Sheikh Yasmeen and U.H. Mane, (2013) repoted in Control group large number of amoebocytes in interlobular connective tissue. Destruction of digestive secretory cells, infiltration of amoebocytes in tubules and digestive cells and karyolysis were most common due to cadmium, especially in Lc50.

Our results confirm the extreme harmfulness of TBT for *L. marginalis* histological notifications on the cells of digestive glands the primary target organ, were observed when *L. marginalis* are exposed to acute concentrations of tributyltin chloride.

REFERENCES:

1. Alzieu C. and Heral M. (1984): Ecotoxicological effects of organotin compounds on oyster culture. In : Persoune G. Jaspers E. and Claws. C (eds) Ecotoxicological testing for and claws. C. (eds). *Ecotoxicological testing for the marine Environ.*, 1. 87- 196.

2. Au, D.W.T., (2004): The application of histo-cytopathological biomarkers in marine pollution monitoring: a review. *Mar. Poll. Bull.* 48, 817–834.

3. Bruno, D. W. and A. E. Ellis (1988): Histopathological effects in Atlantic salmon, *Salmo salar* L.., attributed to use of tributyltin antifoulant. *Aquaculture* 72 : 15 – 20.

4. Couch J. A., (1984); Atrophy of diverticular epithelium as an indicator of environmental irritants in the oyster, Crassostrea virginica. Mar. Environ. Res., 14, 525-526.

5. El-Sikaly A, Khaled A, El Nemr A, (2004): Heavy metals monitoring using bivalves from Mediterranean Sea and red sea. *Environ. Monit. Assess.* 98 : 41-58.

6. Fent, K., (1996): Ecotoxicology of organotin compounds. *Crit. Rev. Toxicol.* 26, 1–117.

7. Focardi, S., S. Corsolini, S. Aurigi, G. Pecetti, and J.C. Sanchez-Hernandez. (1999): Accumulation of butyltin compounds in dolphins stranded along the Mediterranean coasts. *Applied Organometallic Chemistry* **14:** 48-56.

8. Heath AG, (1987): Water pollution and fish physiology. *CRC. Press, Boca Raton. 245pp.*

9. Henry M., (1987): La glande digestive de la palourde Ruditapes decussates L. Recherches wtrastructurales, Cytochimiques, Ecophysiologist, Ecotoxicologist. These dr.es sciences, Uni; Aix-Marseille, 439p.

10. Kharat P.S. (2007): Some physiological aspects of freshwater prawn, *Macrobrachium kistnensis* exposed to organotin tributyltin chloride. Thesis of Dr. B.A.M.U. Aurangabad. pages 1-185.

11. Kumar Saurabh, Rakesh Kumar Pandey, Shobha Das and Vijai Krishna Das (2011): Pathological changes in hepatopancreas of freshwater mussel *Lamellidens marginalis* exposed to sub-lethal concentration of Dimethoate. GERF Bulletin of Biosciences 2011, 2(2):18-23.

12. Laughlin, R. B. JR., W. French., and H. E. Guard. (1983): Acute and sublethal toxicity of tributyltin oxide (TBTO) and its pu- tative environmental product, tributyltin sulfide (TBTS) to zoeal mud crabs, *Rhithropanopeus harrisii. Water, Air, and Soil Pollution* 20:69-79.

13. Lowe D.M., M.N. Moore, K. R. Clarke. (1981): Effects of oil on digestive cells in mussels: quantitative alterations in cellular and lysosomal structure. Aquat. Toxicol., 1, 213-226

14. Marigomez, J.A. V.Saez, M.P. Cajaraville and E. Angulo. Helgolander Meeresunters. (1990): Vol. 44 p. 81-94.

15. Moore M. N.. D. M Lowe, S. L. Moore, (1979): Induction of lysosomal destabilization in marine bivalve mollusks exposed to air. Marine. Biol. Letters, 1, 47-57.

16. Neuberger-Cywiak L, Achituv Y, Garcia EM (2003): Effects of Zinc and Cadmium on the Burrowing Behavior LC50 and LT50 on *Donax trunculus* Linnaeus (Bivalvia - Donacidae). *Bull. Environ. Contam. Toxicol.* 70: 713-722.

17. O'Halloran, K., Ahokas, J.T. and Wright, P.F.A. (1998a): The adverse effects of aquatic contaminants on fish immune responses. *Australasian Journal of Ecotoxicology* **4:** 9- 28.

18. Pozzi, G.and Merlini: Malacol, M. (1977): 16 p. 227-230. Purchon, R.D. (1977): The Biology of mollusca. II ed. Pergamon, Oxford, pp. 1-5.

19. Rasmussen L. P. D., Hage E., Karlog O., (1985): Light and electronmicroscopic of the acute and long-term toxic effects of N-nitrosodipropylamine and N-methylnitrosurea on the marine musscl Mytilus edulis. Mar. Biol., 85, 55-65.

20. Sarojini, R. Nagabhushanam, R. Khan, A. K. Sambasivarao S. and K. Jayalakshmi (1989) : Effect of benzene on the maturation of the ovaries of crab, *Barytelphusa cunicularis. Environ. Series* Vol. II. 11- 15.

21. Sheikh Yasmeen and U.H. Mane, (2013): Changes in hepatopancreas of the bivalve Molluscs Lamellidens marginalis Exposed to Acute Toxicity of Cadmium in summer. World Journal of Fish and Marine Sciences 5 (4): 437-440.

22. Simkiss, K. **and Mason, A. Z.** (1983.): *Metal ions : Metabolic and toxic effects* . In " The Mollusca" . Vol . 2 . *Environmental Biochemistry and Physiology.* PP. 110 - 164 , edited by Hochachka , P. W. (Academic Press . New York , London.

23. Songyot Kullasoot, Tinnapan Netpae and Chitchol Phalaraksha, (2016): Histopathological effects of pulp and paper mill effluent on the digestive glands of river snail, Filopaludinam artensi (Gastropoda

Vivviparidae) in the male klong river, western Thailand. International Journal of Applied Environmental Sciences. Volume 11, Number 4 (2016), pp. 903-913.
24. Suryawanshi, G.D and S.G. Kamble (2020): Impact of mercury on hepatopancreas and gonads of fresh water bivalve, Lamellidens marginalis. Journal of Global Biosciences. Vol. 9. No. 06. Pp. 7550-7559.
25. Tripp M. R., C. R. Fries, M. A. Cravcn, C. E. Grier, C. E. Gates, (1984): Histopathology of Mercenaria mercenaria as an indicator of pollutant stress. Mar. Environ. Res., 14, 521 524.
26. Tripp M. R., C. R. Fries, M. A. Cravcn, C. E. Grier, C. E. Gates, (1984): Histopathology of Mercenaria mercenaria as an indicator of pollutant stress. Mar. Environ. Res., 14, 521 524.
27. Usheva, L.N., M.A. Vaschenko and V.B. Durkina (2006): Histopathology of the digestive gland of the bivalve mollusks, *Crenomytilus grayanus* (Dunker, 1853) from southwestern peter, The *Great Bay, Sea of Japan. Russian Journal of Marine Biology*, Vol. 32: 166-172.
28 Ustina N. T. Habib; Ahmed A. Abdel-Haleem; Omaima M. Mustafa and Eman H. Ismail. (2018): Histological and ultrastructural alternations in the digestive gland of the Egyptian slug, *Limax maximus* (Linnaeus, 1758) treated with botanic molluscicidal thymol, with reference to biological studies. Egyptian Journal of Aquatic Biology & Fisheries. Vol. 22(5): pp137- 148.
29. WHO (1993): *Biomarkers and Risk Assessment: concepts and principles* (World Health Organization, Geneva.
30. Zorita, I., Ortiz-Zarragoitia, M., Soto, M., Cajaraville, M.P., (2006): Biomarkers in mussels from a copper site gradient (Visnes, Norway): an integrated biochemical, histochemical and histological study. *Aquat.Toxicol.* 78, 109–116.