IOSR Journal of Environmental Science, Toxicology and Food Technology (IOSR-JESTFT)
e-ISSN: 2319-2402,p- ISSN: 2319-2399.Volume 9, Issue 9 Ver. I (Sep. 2015), PP 05-11
www.iosrjournals.org

Effect of Heavy Metals on Gills of Fresh Water Bivalve Lamellidens marginalis.

Sandhya M. Sonawane
Department of Zoology,JDMVPS Arts, Science and Commerce College, Jalgaon, Dist. Jalgaon. (M.S.)

Abstract: *The effects of heavy metals on the histopathology gills of the fresh water bivalve L.marginalis was investigated. Samples were collected from the Godavari river at paithan. The bivalves were exposed to 1.6 ppm $CuSO_4$, 0.6 $Hgcl_2$ and 3.9 ppm $Cdcl_2$ for 72 hours acute treatment. For chronic exposure were exposed to 0.82 ppm $CuSO_4$, 0.32 ppm $Hgcl_2$ and 1.95 ppm $Cdcl_2$. Histopathological changes observed in the gills were edema and lifting of lamellar epithelia ,lamellar degeneration, necrotic changes in intercellular epithelial cells. This research simulate a reliable indicator of the aquatic ecosystem contamination and the possible negative impact of the surrounding environment*

Keywords: *Lamellidens marginalis, gills, histopathology heavy metals*

I. Introduction

Godavari river is an important river in India and it flows from western to southern India. The river is 1,465 km long and ranks as the second longest river in the country. Paithan is located 56 kms south of present day Aurangabad on the Godavari river. In last few decades increase in population density, heavy industrialization and agricultural activities have resulted in more and more waste entering in fresh water resources.

There are over 6500 spcies of marine bivalves in the phylum Mollusca(Robertson,L.J.2007). Marine bivalves are known to be natural unique accumulators of contaminants(Madkour,H.A.2005). There is an increased body of evidence implicating heavy metals as a potential threat to aquatic organism by way of studies on their physiology, behavior, biochemistry and ecology(V.R.Chavan and D.V.Muley,2014). Marine organisms are characterized by a greater spatial ability to accumulate some metals when compared with bottom sediments(Kaladharan P, Prema D. 2005). The shellfish represents an important source of protein for coastal communities. It has been predictable ,for instance, that over 90%of human health exposure to several contaminants occurs through diet primarily seafood and meat(Kim, M.,Wolt J.D. 2011, Smith A.G. 2002). Contamination of fresh water with a wide range of pollutants has become a matter of concern over last few decades (Vutukuru,2005). Heavy metals have devastating effects on ecological balance of the recent environment and a diversity of aquatic organisms (Farombi et.al,2007) . In order to evaluate the adverse effect of the pollutants on aquatic organisms ,there is a world wide trends to complement physical and chemical parameter with bio markers in aquatic pollution monitoring (Abdel et.al,2012). Bivalves are used in monitoring programmes in the marine environment due to their ability to concentrate pollutants to several orders of magnitude above ambient levels in seawater. O Connor,T.P.(2002) and Jung, K, Zauke G.P. (2008) reported that bivalves had been used as successful biomonitors of aquatic metallic pollutants levels internationally prominent examples including the United States National Oceanic and Atmospheric Administration Mussel Watch Programmes and the Australian Oyster Watch Programmes.

Effects of heavy metals on gills showed lamellar degeneration, epithelial lifting and necrotic changes in intercellular epithelial cells (V.R.Chavan and D.V.Muley,2014) .Necrosis of the epithelial cells on secondary lamellae, hyperplasia of the epithelial lining of gills filament, abnormal dilation of capillary lumen and destruction of blood cell in fish Carassius carassius (N.senol,S.Tekin Ozan,2013) .The gills of heavy metal expose group show some epithelial lesions when exposed to heavy metals. The filament regions constitute edema with intense lamellar vasso dilation. The gills exhibit a stratified fusion of stratified pigments in numerous spaces are found in the gill of heavy metal exposed common carp Cyprinus carpio (Vinodini,R and Narayanan,2008) .The present study was aimed to evaluate the histopathological effects of heavy metals copper, mercury and cadmium on gills of L.marginalis.

II. Material and methods

The bivalves, L.marginalis were collected from the Godavari River at paithan. The bivalves were acclimatized to laboratory conditions upto 2-3 days, before subjecting them to experiments. healthy active animals of approximately the same size and weight were chosen the acclimatized active bivalves were divided into two groups .one group of bivalves was exposed to lethal concentration(LC_{50} values of 96 hours) concentrations 1.9 ppm ,0.6 ppm and 3.9 ppm of $CuSO_4$, Hgcl2 and $Cdcl_2$ respectively upto 96 hours of treatment. Another group of acclimatized group of was exposed to sub lethal concentration of heavy metals, pollutants upto 20 days for chronic treatment. After acute and chronic exposure the animals were dissected and the gills were removed and fixed in Bouin's fluid. After dehydration they were embedded in paraffin wax and were sectioned at 5 microns. The sections were stained by the hematoxelene –eosin method and observed under a light microscope. All the precautions recommended by (ATSDR,2003) to minimize risks of sample contamination were followed during collection and treatment of sample.

III. Result And Discussion

Histopathological changes_after $CuSO_4$, Hgcl2 and $Cdcl_2$ exposes are anticipatory in nature. These abnormalities which occur in the tissues and cellular level are the results of complex physiological dis function since gills are the major respiratory organ . Gill surfaces are constantly exposed to water currents in aquatic bivalves; they have to face of all the toxic and harmful substances. The observations made in the present study indicate that thé damage in the tissue architecture in the gill is more severe in higher concentrations. This concentration dependent toxic intensity has also been reported by Ghate and Mulherkar (1979) in the gills of two species of fresh water prawns exposed to $CuSO_4$. Histopathological alteration of gills of bivalve tissues have been shown to be responsive and sensitive to wide range of contaminants because they play an important role in respiration and food collection.(Au,D.W.2004,Abdel Nabi,I.M,Taha I.A. & Moawad,T.I.2007).The histological changes of gill of bivalve in the present study are in agreement with (El Shenawy,N.S., Moawad,T.I.2009) as they reported irregularity of gill lamellae of the cells ,swelling of gill filament and haemocytes infiltration of bivalve.epithelial cells of gills play a crucial role (Choi H.J.,Ahn,I.Y.2003) the damage in the epithelium results in serious dysfunction of tissue consequently leading to deleterious effect on the organization levels(Madkour,H.A.2005) Bivalves possess different measures of defense against environmental hazards e.g. particle rejection and formation of pseudo faces, reduce filtration rate and valve closure (Waterman,B.T.,Herlyn,M....2008) Bonneris ,E,Fiala,Medoni(2005) reported that metal concentration recorded in soft tissue and muscle increased without a source of extra metals in water.

Manduzio,H.& Rocher,B.(2004) concluded that gills food preferentially be used in bio monitoring studies in blue muscles .The gill tissue of Mytillus galloprovincialis is responsible for the uptake of metal ions from water.(Znidaric,M.T.,Falnoga,I,and Turk,V.2005)Cd is not an essential element for animals (Madkour,H.A.2005)

Gills from copper sulphate (plate II.a)

The_first noticeable effect of copper sulphate in general on the gills of bivalve was the enlargement of respiratory epithelial cells. Nuclear size was greatly increased and cytoplasm shows granular structure, nuclei showed irregular shape and they also showed the granular appearance. Cytoplasm also showed disintegration, because of swelling of respiratory epithelium .the interlamellar space was reduce . in the central core there was conjection of blood cells. At certain places connective tissue core also showed the changes like enlargement of capillaries, vacuolization and disintegration of normal structure.

The mercury treated gills (plate II.b)

Exhibited severe alternate in the normal architecture of gill lamellae. The epithelial cells showed totally altered structure. The shape was lost, the nuclei show pycnotic conditions, moderate necrotic changes in interlamellar epithelial cells.,crysting of gill filament tips,infiltration of cells in primary axis. The shrinkage was prominent and because of shrinkage cavities were form between epithelial cells.at certain places mercury treated lamellae showed fusion and loss of interlamellar spaces.

Cadmium treated gills (plate II.c)

Showed enlargement of nuclei in the epithelial cells. Cytoplasm showed the degenerative changes and therefore mass of nuclei was observed in the lamellae .The central core of lamellae showed dialation.In certain regions the interlamellar space was filled with coagulated mucous and it was reduce due to enlargement of respiratory epithelium.

Gills from chronic exposure (plate III a)

Chronic exposure of copper sulphate showed drastic changes in the normal architecture of gill lamellae, cytoplasm showed disintegration because of swelling of respiratory epithelium. The gills of lead exposed fish showed dilation and congestion in blood vessel of primary gill filament (V.R.Chavan and D.V.Muley, 2014).due to copper sulphate there was overall enlargement of the lamellae which reduce the interlamellar space. Also the border of epithelial cells at some places showed degeneration. Some studies revealed that interstitial edema is one of the more frequent lesions observed in gill epithelium of fish exposed to heavy metals.(Mallatt,1985) edema with lifting of lamellar epithelium would be serve as a mechanism of defense (Arellano et.al.,1999)

Mercury treated gill lamellae (plate III.b)

Exhibited highly abnormal architecture, necrosis was evident in gill epithelium from the appearance of pycnotic nuclei .towards proximal and disintegration of nuclear material was more prominent than distal region. The blood capillaries also showed congestion of blood cells. At certain places blood cell show shrinkage and vacuolization.hyperplacia of epithelial cells between secondary lamellae led to fusion and separated from pillar system.vacuolisation and necrosis of lamellar epithelial cells ,congestion of central lamellar vein and hyperplasia of lamellar epithelial cells was evident in gill of fish exposed to lead (V.R.Chavan and D.V.Muley, 2014)

The chronic treatment of cadmium chloride altered (plate III.c)

the nuclei showed granular nature , the blood vessels in the connective tissue showed presence of leucocytes which also shows shrinkage. Some lamellae were fused at the proximal end and degenerative changes were observed in distal ends of lamellae. some studies reveals that histopathology evaluation of the toxic effect of heavy metals in the nucleus changes in the cell, lamellae structure ,necrosis inflammation hyperplacia,hypertrophy,athrophyad amount of mucous in epithelial cells were examined (N.Senol,S.Tekin Ozan,2013).the main histopathological changes observed in the gills were edema and lifting of lamellar epithelia(Vinodini,R and Narayanan M,2009).

Similarly Shanmugam A and Kesavan K(2007) reported that bioaccumulation of heavy metals such as magnesium, iron, zinc and copper concentration in different body parts and shell of C.melo from Cuddalore coast. Huang J.Y.(2007) reported that, level of accumulation of heavy metals such as mercury, cadmium, lead, zinc, copper and arcenic in soft tissues of 15 species of benthic invertebrates from Zhejiang costal waters, East China. The concentration of iron in different soft tissues and byssus and also studied the potential role of the byssus as an excretion route for iron in P. viridis(Yap CK, Tan SG 2007). Cadmium is widely distributed at low level in the environment and most foods have an inherently low level of Cd which has been shown to bind to the protein and accumulate significantly in higher level(FDA 2011).

Plate 1

Fig 1- L.S. of gill of Lamellidens marginalis showing normal architecture of gill, lamellae, respiratory epithelium, cilia,nuclei and interlamellar space x100.

GL- Gill Lamellae
RE- Respiratory Epithelium
C- Cilia
N- Nuclei
ILS- Interlamellar Space

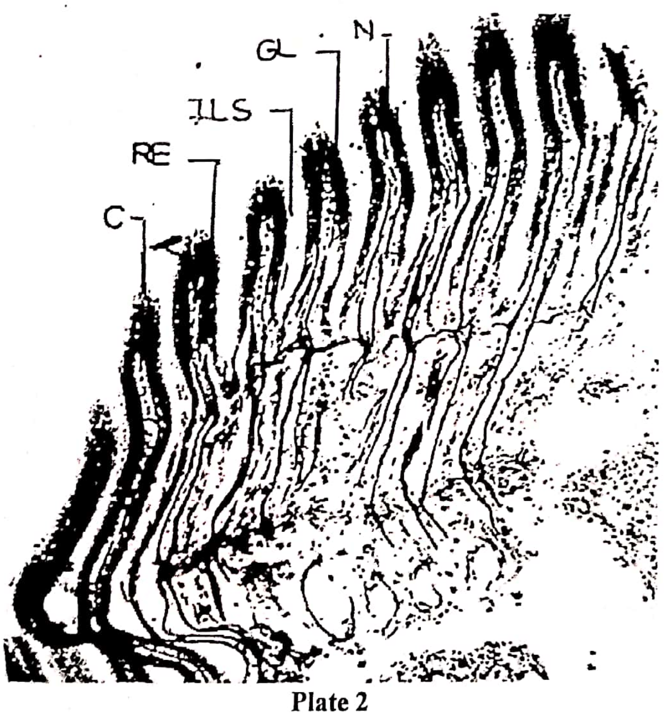
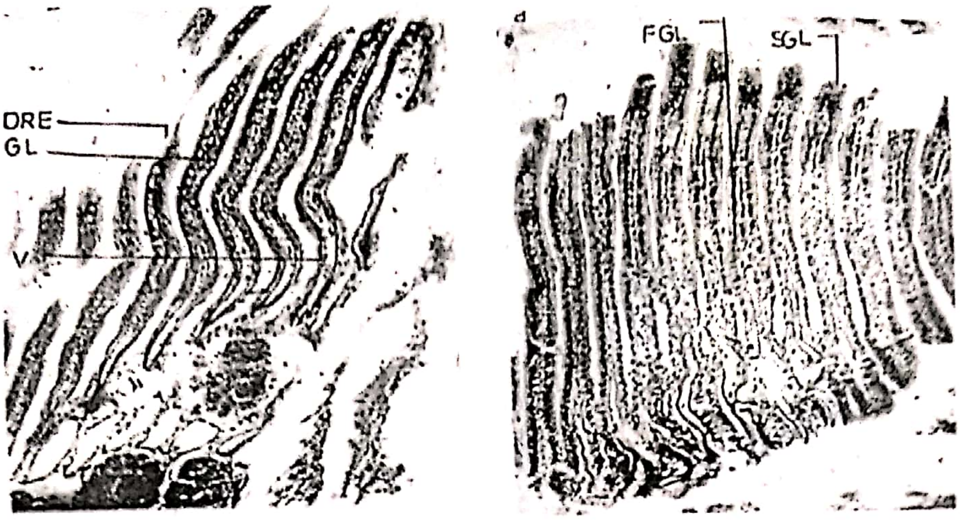


Plate 2



GL- Gill Lamellae
SGL- Swollen Gill Lamellae
DRE- Degenerated Respiratory Epithelium
FGL- Fused Gill Lamellae
CR- Chitinous Rod

PLATE 2:

L.S. of gill of L. marginalis exposed to acute treatment of (96 hrs) of copper sulphate, mercury chloride and cadmium chloride.

Fig:A Acute Copper Sulphate(96 hrs) x 100

Gill lamellae showing increased nuclear size, granular cytoplasm and nuclei, reduced interlamellar space, swollen gill lamellae and lamellar fusion.

Fig:B Acute Mercury Chloride (96 hrs) x 100

Gill lamellae showing vacuolization, degeneration of nuclei, necrosis and dissolution of respiratory epithelium.

Fig:C Acute Cadmium chloride (96 hrs) x100

Gill lamellae showing shrinkage of cytoplasm, vacuolization and degeneration of respiratory epithelium.

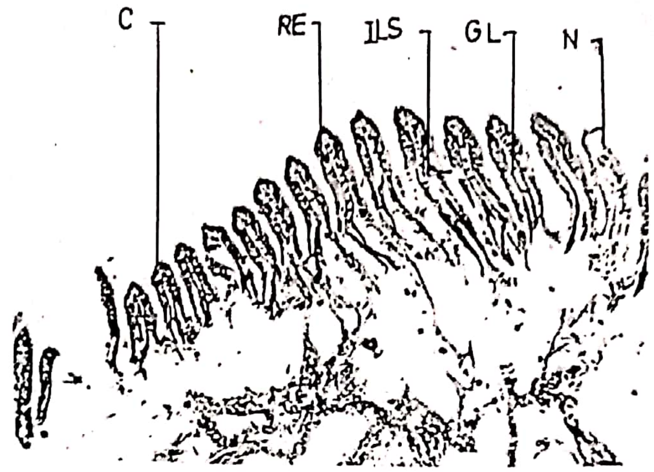
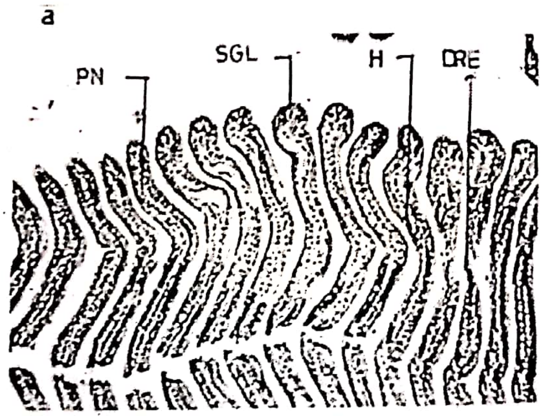


PLATE 3:
L.S. of gill of L.marginalis exposed to chronic treatment (20 days) CONTROL

PLATE 4:



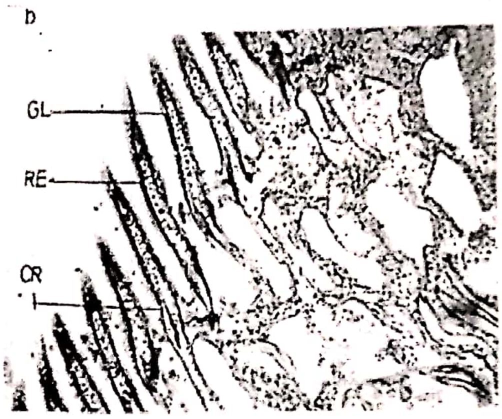
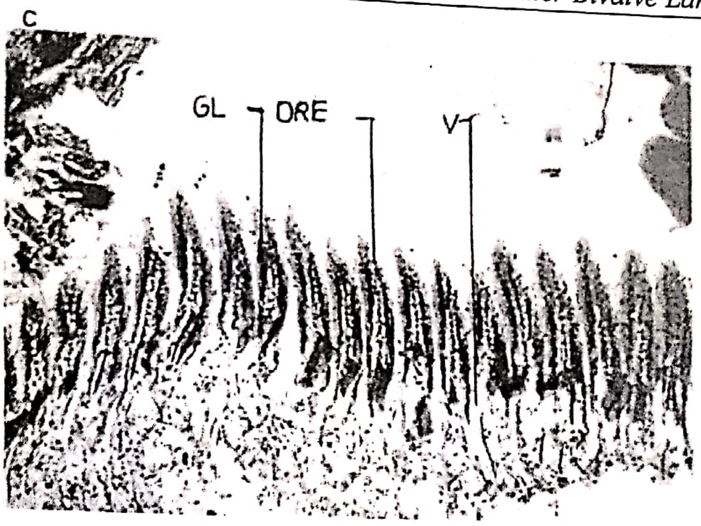


PLATE 4:
L.S. of gill of L. marginalis exposed to chronic treatment(20 days) of heavy metals, $CuSO_4$, $Hgcl_2$ and $Cdcl_2$.
Fig a; Chronic $CuSO_4$ x100
Gill lamellae showing swelling at their tips, pycnotic nuclei, vacuolization, congestion of haemocytes and degeneration of respiratory epithelium.
Fig b:Chronic $Hgcl_2$ x100
Gill lamellae showing degenerating nuclei , necrosis and dissolution of respiratory epithelium, vacuolization.
Figc: Chronic $Cdcl_2$, x100
Gill lamellae showing vacuolization, shrinkage of cytoplasm, degeneration of respiratory epithelium.
GL Gill Lamellae
SGL Swollen Gill Lamellae
H Haemocytes
PN Pycnotic Nuclei
DRE Degenerative Respiratory Epithelium
CR Chitinous Rod
V Vacuoles.



IV. Conclusion

Histopathological changes in bivalve L.marginalis due to effects of heavy metals can be used as a sensitive model to monitor the aquatic pollution and are widely used in water quality monitoring programmers in many countries. The current results indicate that heavy metal contamination definitely affects the gills by necrosis, edema, proliferation, hyperplacia, hypertrophy.It is recommended to treat the effluent (detoxification) before discharging to the resources to avoid negative impact on aquatic biota. The present research work served as an experimental tools and bio indicators for the first line evaluation of environmental pollution.

Bibliography

[1]. Abdel-Moneim A. M., Al-Kahtani M. A., and Dlmenshawy O. M. (2012.) Histopathological biomarkers in gills and liver of Oreochromis niloticus from polluted wetland environments, Saudi Arabia. Chemosphere, 88: 1028- 1035

[2]. Abdel Nabi, I.M., ElShenawy, N.S., Taha, I.A. and Moawad, T.I. (2007) Oxidative Stress Biomarkers and Bioconcentration of Reldan and Roubdup by Edible Clam Ruditapes decussatus. Acta Zoologica Sinica, 53, 910-920.

[3]. Agency for Toxic Substances and Disease Registry (ATSDR) (2003) For Fish and Shell-Fish Evaluation for (Isla) de vieqmes Bombiong Range Public Health Assessment (PHA).

[4]. Au, D.W. (2004) The Application of Histo-Cytopathological Biomarker in Marine Pollution Monitoring. Marine Pollution Bulletin, 48, 817-834. http://dx.doi.org/10.1016/j.marpolbul.2004.02.032

[5]. Blasco, J. and Puppo, J. (2005) Effect of Heavy Metals (Cu, Cd and Pb) on Aspartate and Alanine Aminotransferase in Ruditapes philippinarum (Mollusca: Bivalvia). Comparative Biochemi stry and Physiology Part C, Pharmacology, Toxicology and Endocrinology, 122, 253-263.

[6]. Bonneris, E., Giguere, A., Perceval, O., Buronfosse, T., Masson, S., Hare, L. and Campbell, P.G. (2005) Role of Calcium Concretions in Metal Sequestration: Bivalve, Pyganodon grandis.

[7]. Choi, H.J., Ahn, I.Y., Lee, Y., Kim, K.W. and Jeong, K. (2003) Histological Responses of the Antarctic Bivalve Lalemula elliptica to a Short-Term Subleathal Level Cd Exposure. Ocean and Polar Research, 25, 147-154. http://dx .doi.org/10.4217/ OPR .2003 .25.2.147

[8]. El Shenawy, N.S., Moawd, T.J.S., Mohallal, M.E., Abdel-Nabi, I.M. and Taha, I.A. (2009) Histopathologic Biomarker Response of Clam, Ruditapes decussatus, to Organ Phosphorous Pesticides Reldan and Roundup: Laboratory Study. Ocean Science Journal, 44, 27-34. http://dx.doi.org/10.1007/s12601-009-0004-5

[9]. FDA (2011) Fish and Fisheries Products Hazards and Controls Guidance – Fourth Edition. Centre for Food Safety and Applied Nutrition, US Food and Drug Administratio

[10]. Farombi E. O., Adelowo O. A.and Ajimoko Y. R.(2007). Biomarker of oxidative stress and heavy metal levels as induced by environmental pollution in African cat fish Clarias gariepinus from Nigeria Ogun river. Int. J. Environ. Res. Public Health, 4 (2): 158-165.

[11]. Ghate H. V.and Mulherkar L.(1979). Histological changes in gills of two fresh water prawns species exposed to copper sulphate. Indian J. Exp. Biol, 18 : 1040-1042

[12]. Goksu MZL, Akar M, Evik F, Findik (2005) Bioaccumulation of some heavy metals in two bivalvia species. Turk J Vet Anim Sci 29: 89-93.

[13]. Huang JY, Wu JH (2007) Heavy metal monitoring using bivalved shellfish from Zhejiang coastal waters, East China Sea. Environ Monitoring Assess 129: 315- 320.

[14]. Jung K, Zauke GP (2008) Bioaccumulation of trace metals in the brown shrimp Crangon crangon (Linnaeus, 1758) from the German Wadden Sea. Aquat Toxicol 88: 243-249.

[15]. Kaladharan P, Prema D, Valsala KK, Leelabhai KS, Rajagopalan M (2005) Trends in heavy metal concentrations in sediment, fin fishes and shellfishes in inshore waters of Cochin, southwest coast of India. J Mar Biol Ass 47: 1-7.

[16]. Kim M, Wolt JD (2011) Probabilistic risk assessment of dietary cadmium in the South Korean population. Food Aditives & Contaminents: Part A 28: 62- 70.

[17]. Madkour, H.A. (2005) Distribution and Relationships of Heavy Metals in the Gaint Clam (Tridacna maxima) and Associated Sediments from Different Sites in the Egyptian Red Sea Coast. Egyptian Journal of Aquatic Research, 31, 45-59.

[18]. Manduzio, H., Monsinjon, T., Galap, C., Leboulenger, F. and Rocher, B. (2004) Seasonal Variations in Antioxidant Defences in Blue Mussels Mytilus edulis Collected from a Polluted Area: Major Contributions in Gills of an Inducible Isoform of Cu/Zn-Superoxide Dismutase and of Glutathione S-Transferase. Aquatic Toxicology, 70, 83-93.

[19]. O'Connor, T.P. (2002) National Distribution of Chemical Concentrations in Mussels and Oysters in the U.S.A. Marine Environmental Research, 53, 117-143.

[20]. Ololade IA, Lajide L, Amoo IA, Oladoja NA (2008) Investigation of heavy metals contamination of edible marine seafood. Afr J Pure Appl Chem 2: 121-131.

[21]. Palpandi C, Kesavan K (2012) Heavy metal monitoring using Nerita crepidulariamangrove mollusc from the Vellar estuary, Southeast coast of India. APJTB 2: S358-S367.

[22]. Shanmugam A, Palpandi C, Kesavan K (2007) Bioaccumulation of Some Trace Metals (Mg, Fe, Zn, Cu) from Begger's Bowl Cymbium melo (Solander, 1786) (A Marine Neogastropod). Research J Environ Sci 1: 191-195.

[23]. Sherifa Shaker Hamid,Eman Hashem Radwan, Gaber Ahmed Saad(2014) Impact of selected Physicochemical Parameters and some Environmental pollutants on the Gill in Pinctada radiate from coastal zones, Egypt.open journal of Ecology,4,907-917.SciRes.http://www.scirp.org/journal/oje.

[24]. Soazig, L. and Marc, L. (2003) Potential Use of the Levels of the mRNA of a Specific Metallothionein Isoform (MT-20) in Mussel (Mytilus edulis) as a Biomarker of Cadmium Concentration. Marine Pollution Bulletin, 46, 1450- 1455. http://dx.doi.org/10.1016/S0025-326X(03)00283-2

[25]. Vutukuru S. S. 2005. Acute effect of Hexavalent chromium on survival, oxygen consumption, haematological parameter and some biochemical profiles of the Indian Major carp, Labeo rohita. Int. J. Environ. Res. Public Health , 2 (3): 456-462.

[26]. Vinodini R. and Narayanan M. 2008. Bioaccumulation of heavy metals in organs of fresh water fish Cyprinus carpio (Common carp) Int. J. Environ. Sci. Technol. 5: 179-182.

[27]. V.R.Chavan and Muley.2014.Effect of heavy metals on liver and gills of fish Cirrhinus marigala,International j.Current Microbiology and Applied Sci;3(5):277-288.

[28]. Waterman, B.T., Herlyn, M., Daehne, B., Bergmann, S., Meemken, M. and Kolodzey, H. (2008) Pathology and Mass Mortality of Pacific Oyster, Crassostrea gigas (Thunberg), in 2005 at the East Frisan Coast, Germany. Journal of Fish Diseases, 31, 621-630. http://dx.doi.org/10.1111/j.1365-2761.2008.00953.x

[29]. Yap CK, Cheng WH (2009) Heavy metal concentration in Nerita lineate: the potential as a biomonitor for heavy metal bioavailability and contamination in the tropical intertidal area. Marine Biodiversity Records 2: e49.

[30]. Znidaric, M.T., Falnoga, I., Skreblin, M. and Turk, V. (2005) Induction of Metallothionein-Like Proteins by Mercury and Distribution of Mercury and Selenium in the Cells of Hepatopancreas and Gill Tissues in Mussel Mytilus galloprovincialis. Biological Trace Element Research, 111, 120-140.

[31]. Zyadah, M., Ibrahim, M. and Madkour, A. (2004) Impact of Environmental Parameters on Benthic Invertebrates and Zooplankton Biodiversity of the Eastern Region of Delta Coast at Damietta, Egypt. Egyptian Journal of Aquatic Biology and Fisheries, 8, 37-52.