Effect of Heavy Metal Pollutants on Fish Population

Shobharam Waskel
Department of Zoology, Dr. A. P. J. Abdul Kalam University, Indore – 452010

Dr. Meenakshi Solanki
Department of Zoology, Dr. A. P. J. Abdul Kalam University, Indore – 452010

Abstract

Aquaculture is perhaps the main financial endeavors in numerous nations across the world. Throughout the most recent couple of many years, there has been expanded interest for fish by purchasers, which has required the improvement of novel systems for upgraded creation of aquaculture. Notwithstanding, ecological and wellbeing challenges are expanding overall which are perceived as critical imperatives on aquaculture creation and exchange. Aimless utilization of synthetic compounds in agriculture, and modern effluents unfavorably influence the aquaculture and related climate. Heavy metals pollutants are dangerous to the environment. Some of these include essential elements that are required for the normal metabolism of the organism such as Cu, Fe and Zn, while others are non-essential and play no significant role including Cr, Pb and Cd. Their natural effects can be toxic (acute, chronic or sub-chronic), carcinogenic, mutagenic or teratogenic. Eliminating the synthetic impurities, from the climate is a troublesome errand. Hence, different techniques to remediate the substance impurities are being investigated. In this unique circumstance, bioremediation or the use of microorganisms to tidy up these toxins from a contaminated climate has showed up as a promising methodology for eliminating as well as diminishing the impact of the impurities. Ongoing headways made in this field of examination are talked about in this paper.

Keywords: *Bioremediation, Fish Culture System, Heavy Metals, Rivers.*

Introduction

Fish is a commodity of potential public health concern as it can be contaminated with a range of environmentally persistent chemicals, including heavy metals (Soliman, 2006). In India, the need for increasing fish production is necessary. Therefore, efforts have been made to increase fish production, at least to maintain the present level of consumption in view of the rapid population increase (El-Nemaki et al., 2008). The significance of Rivers fisheries lies not only in the supply of fish for regional domestic consumption (3,300 tons in 2003), but especially in regional employment and in export earnings (El-Bawaab, 1995). Rivers are source of good quality fish since it is a protected area. Rivers is economically the most important delta Rivers in India. It provides about 30% of domestic fish supply (Abd El-Hakim et al., 1999).

Heavy metals pollutants are dangerous to the environment. They include essential elements that are required for the normal metabolism of the organism such as Cu, Fe and Zn, and other non-essential elements that play no significant role including such as Cr, Pb and Cd. Their effect can be toxic (acute, chronic or sub-chronic), carcinogenic, mutagenic or teratogenic. Heavy metals are one of the five major types of toxic pollutants present in fresh water (Mason, 1991). The important environmental pollutants are those that tend to accumulate in organisms, those which are persistent because of their chemical stability or

poor biodegradability, and those which are readily soluble and therefore environmentally mobile (Hellawell, 1986). Heavy metals pollutants have been found to be dangerous to the environment (Phillips, 1991). Some of them are essential elements that are required for the normal metabolism of the organism including Cu, Fe and Zn, while the others are non-essential and play no significant role including Cr, Pb and Cd (Sanders, 1997). Their natural effects can be toxic (acute, chronic or sub-chronic), carcinogenic, mutagenic or teratogenic (Young, 2005). The present study investigated heavy metals levels (Cr, Pb and Cr) in Rivers and compared them with Rivers . It revealed the effect of heavy metal pollution on reproductive ability of thin lipped grey mullet fish (Indian fishes) which belongs to order Mugiliformes and family Moronidae (McDonough and Wenner, 2003).

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Materials and Methods:

Water samples of fish were collected from Rivers during the summer times. Cr, Pb and Cd concentration levels were determined using flame atomic absorption spectrophotometer with an air acetylene flame (Model 2380, Perkin-Elmer) according to Zottner and Seligson (1964). Statistical analysis was performed by using the student t-test (Murray, 1982). Fish tissue from both Rivers was dissected for inspection of the gonads (testes and ovaries). Specimens were fixed in 10% saline formalin, dehydrated in ascending series of alcohol and embedded in paraffin wax. Serial transverse sections at 5µm were stained by Haematoxylin-Eosin (Clayden, 1971). Total polysaccharides and protein contents were identified according to Causan and Pickett (1983) and Launa (1968) respectively. Masson's Trichrome Staining method was used for collagen fibers (Masson, 1929) and Methyl Green Pyronin Y method was used for detection of RNA/DNA content (Kurnick, 1955).

Results:

1. Heavy Metals in Water: Rivers water displayed the highest concentration levels (ppm) of tested heavy metals (Cr, Pb and Cd), which was showed by high significant difference (table 1). According to safety limits in the two environmental laws No. 48/1982 and No. 4/1994, concentration means of Cr, Pb and Cd in Rivers exceeded these recorded safety limits, but River water not exceeded them.

2. Histological and Histochemical Analysis:

I. Testes: Histologically, Indian fishes testes showed numerous seminiferous tubules exhibiting intensive spermatogenesis, reduced number of spermatogonia and many spermatids and spermatozoa (Figure 1). These seminiferous tubules showed both primary and secondary spermatocytes (Figure 1). Testes of Indian fishes from Rivers showed decreased number of seminiferous tubules along with some degenerative changes (Figure 2). They appeared empty different from primary and secondary spermatocytes. The interstitial cells and sertoli cells had invaded all seminiferous tubules sperm nests. Accordingly, seminiferous tubules showed lesser number of spermatids which had a radiated shape without any inner septa between nests (Figure 2). This can cause a marked decrease in spermatogonial mitosis and inhibition in the development of early stages of male germ cells resulting in delayed transformation of the spermatids to mature spermatozoa. Histochemically, testicular tissue of Rivers Indian fishes exhibited a strong PAS reaction for polysaccharides (Figure 3). It also showed a large amount of protein after application of mercury-bromophenol blue staining method (Figure 5). However, testicular tissues of Rivers fish exhibited lesser amount of polysaccharide (Figure 4) and protein after Mercury Bromophenol Blue reaction (Figure 6). Testicular tissues of Rivers Indian fishes showed greater RNA/DNA content (Figure 7) when compared with testicular tissues of Rivers studied fish after the use of Methyl Green Pyronin Y stain (Figure 8). They also showed a large amount of collagen fiber content by Masson's Trichrome stain technique (Figure 10). Conversely, very little collagen fiber content was found in testicular tissues of Indian fishes from Rivers by Masson's Trichrome stain method (Figure 9).

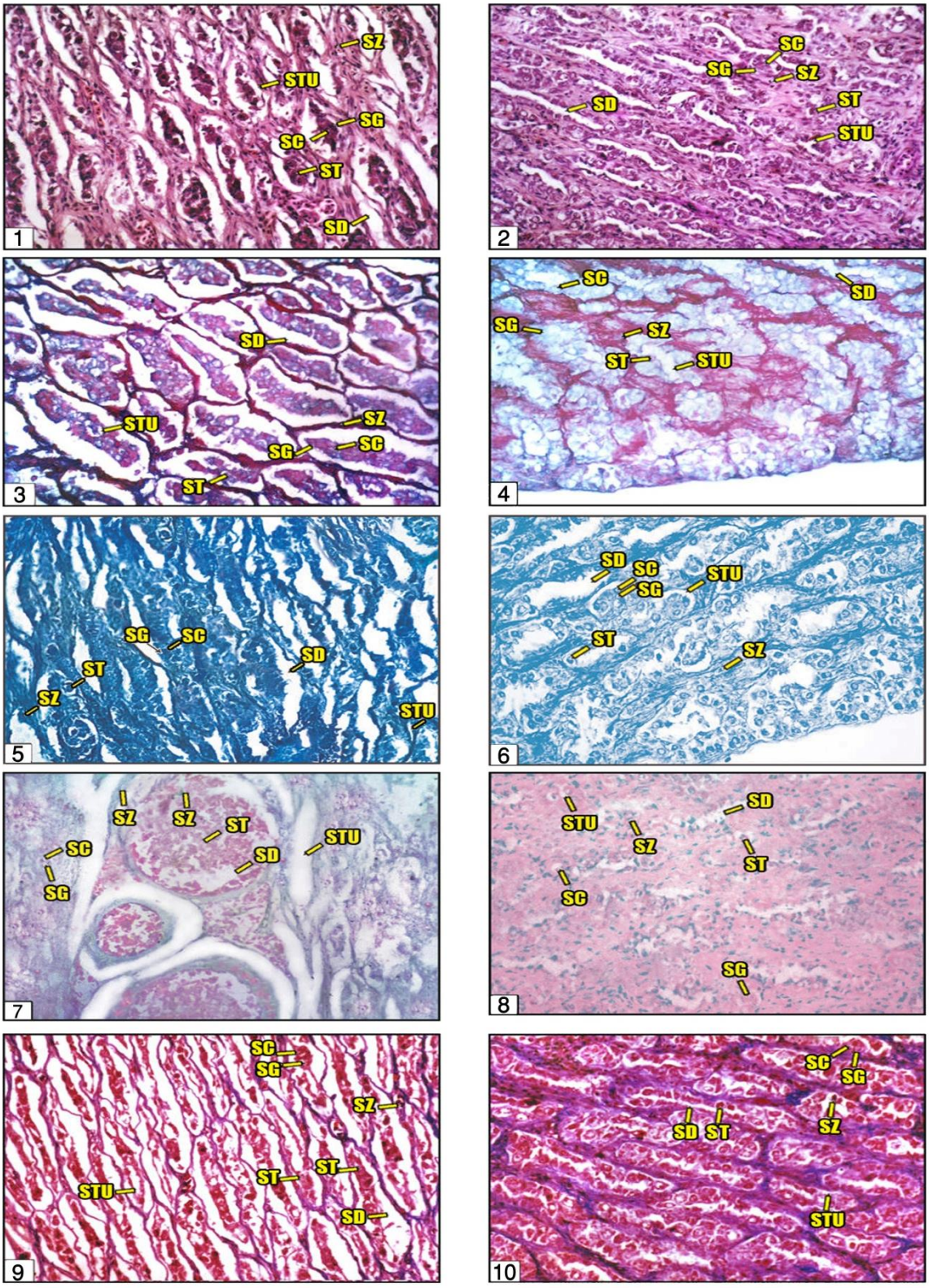
II. Ovaries: Ovaries of Indian fishes from Rivers showed no histological lesions. Minute polygonal undifferentiated oogonia with a little amount of oviplasm were observed in early and late peri-nucleolar stages. Oocytes were increased in size and enveloped by follicular epithelia and vitelline membrane (zona radiate) as a protective boundary for oviplasm (ooplasm). The vitellogenetic stage (lipid-yolk vesicles stage), was noticed with lipid vesicles in oviplasm. There was yolk protein in the central portion of oocytes (yolk spheres). The vitelline membrane (zona radiate) was visible between oviplasm and follicular layers (granulose cell layer and theca cell layer) as they were called zona granulose. Ovaries of Indian fishes from Rivers showed atresia (degeneration). Necrotic oocytes, a distinctive yolk and vacuolated cytoplasm; also, broken tunica albuginea were seen at all sites. Histochemically, ovarian tissue of Indian fishes from Rivers exhibited large amount of polysaccharide, proteins and DNA/RNA contents after application of PAS reaction, mercury-bromophenol blue and methyl green pyronin Y stain respectively. However, Rivers Indian fishes ovarian tissues showed lower amount of polysaccharides, proteins and DNA/RNA contents. They also showed less collagen fiber by Masson's stain method. Conversely, ovarian tissue of Indian fishes from Rivers showed a large amount of collagen fiber These collagen fibers were present in ovarian tunica albuginea.

Table (1): Mean concentrations (ppm) of Cr, Pb and Cd in Lake Bardawil water and Lake Manzalah at summer (2011). Lake Bardawil is a protected area, so it was included as the control. Values are expressed as mean ± standard deviation.

	Element					
	Cr		**Pb**		**Cd**	
Lake	Bardawil	Manzalah	Bardawil	Manzalah	Bardawil	Manzalah
Mean	0.0053±0.001	0.073±0.01	0.00095±0.0006	0.04±0.015	0.002±0.001	0.036±0.013
Standard Error	0.0005	0.005	0.0003	0.0075	0.005	0.006
t-Test	0.00172		0.00375		0.0054	
Significance	**		**		**	

(*): Significant: 0.01 P 0.05 and (**): Highly significant: P < 0.01.

Figures 1-10 Showing Cross section in Fish testis also showing seminiferous tubules (STU), spermatogonia (SG), spermatocytes (SC), spermducts (SD), spermatids (ST) and spermatozoa (SZ). (Hx & E., × 250).



Discussion:

Pollution of aquatic habitats is a major problem in India. In recent years, more toxic compounds were detected in aquatic ecosystem (Khare and Singh, 2002). Fishing is one of the most important industries and seafood is consumed by a large segment of Indiaian population (Soliman, 2006). However, fish population can be effected by a large range of the environmentally persistent heavy metals (Soliman, 2006).

Heavy metals are present at trace levels in aquatic environment (Al-Weher, 2008). Their levels increase due to industrial, agricultural and mining activities and they contribute to the pollution of aquatic ecosystems (Canli and Atli, 2003). Some heavy metals are essential (Fe, Zn and Cu…etc) and necessary in trace concentrations for normal growth and development (Kalay and Canly, 2000). They become toxic when their levels exceed required levels (Yilmaz, 2005) and be harmful to human health (ATSDR, 2004). However, the other non-essential heavy metals (Pb, Cd and Cr…etc) even at low concentrations are very toxic to aquatic ecosystems and human health (Jayakumar and Paul, 2006).

In the present study, the mean concentrations of Cr, Pb and Cd (non-essential heavy metals) which presented in Rivers water were 0.073-0.04-0.036 (ppm), respectively as they exceeded the recorded permissible levels in Indiaian Standards of Environmental Laws No. 48/1982 and No. 4/1994 which stated the maximum concentrations of Cr, Pb and Cd in fresh water should not exceed 0.001-0.01-0.05-0.05 (ppm), respectively. These observations showed that there were accumulated heavy metals in Rivers water. Cd exposure is linked with renal failure, bone fragility as osteoporosis and is considered as a cancer causing agent in the human health (ATSDR, 2004). There is adverse toxic effect of Pb on human health particularly children was recognized (Subramanian, 1988) in addition, neurological defects, renal tubular dysfunctions and anemia (Forstner and Wittman, 1983). Cr complexes, which are bounded to the other lower molecular weight legends, are mostly traversed to the cell membrane (Mertz, 1969) and concentrated in tissue organism and interacts with the cellular macromolecules, including DNA, or may be slowly released from the cell and then causes human carcinogen (Wiegand et al., 1985).

Heavy metals accumulate in fish tissues, as they can reach concentration levels up to 2000 fold higher than those found in the surrounding water environment (Popek et al., 2006) and transferred through the upper classes of food chain. Human, obtain the heavy metal contaminations from aquatic ecosystem food, especially fish (Langston, 1990). Toxic heavy metals can alter tissue histological structures, and cause reproduction defects (Weis and Weis, 1989). In addition, Bobek et al (1996) showed that pollution affects the normal structure of fish tissues. Heavy metals may have direct effects on fish gonads (testes and ovaries), resulting in a disturbed development of germ cells (Mohamed and Gad, 2008).

The present study demonstrated the histological configurations of Indian fishes testes from Rivers have the normal regular of spermatogenesis process stages (Weltzien et al., 2002). The environmental impacts on Rivers water caused a pronounced decline in gonad activity of the studied fish which reflected by decreasing sperm and spermatids counting in ripe testes, ripe oocytes degeneration (atresia) including spermatogonia, of the seminiferous tubules and necrosis as these histological changes in testes tissue were progressively increased with the increasing degree levels of heavy metal accumulation (Phillips, 1991) and that resulted in permanent testicular damage which reduce the fish ability to reproduce by suppressing sperm production (Hanna et al., 2008). Histochemically, L. ramada testicular tissues from Rivers showed an obvious decreasing of

the total polysaccharides and total protein, DNA/ RNA contents and increasing in collagen fibers composition according to the collapsed interstitial cells against the sperm nests due to severe heavy metals effect when compared with those collected from Rivers . Subsequently, the amount of the PAS-positive materials and mercury-bromophenol blue stained materials in the tissues indicated the degree of the total polysaccharide and protein contents in the testicular tissues. Since, the intensity of staining is greatly dependent on the amount of polysaccharides and proteins in such sites. That applied also on DNA/ RNA contents and collagen fibers (Chandra and Khuda, 2004).

The oogenesis cycle of Indian fishes is divided into five stages namely: (i) chromatin nucleolar stage, (ii) perinulceolar stage, (iii) yolk vesicle stage, (iv) vitellogenic stage and (v) ripe (mature) stage (Alne, 1999(. These were clearly apparent in Rivers fish. In the present study, pre-nucleolus stage was characterized by small size oocytes. At the end of this stage, the late per-nucleolus stage distinguished from the early pre-nucleolus stage by the enlargement of oocyte as the primitive wall of the oocyte was composed of one thin layer of flattened follicular cells (Zaki et al., 1993). The second growth phase was the vitellogenic stage which included yolk vesicle stage and was characterized by vacuoles in circular manner and the oocyte wall was consisted of two layers: an externally situated zona granulose and an internally located zona radiata layer near the cytoplasm (Yammamoto and Yoshioka, 1964). The overall patterns of oocyte development of the studied fish from Rivers showed marked degenerative changes. There was a distinctive form of yolk, and cytoplasm which was greatly changed and appeared highly vacuolated. Also, broken tunica albuginea was spread at all sites with greatly deformed cytoplasm. The degree of maturation of female fishes from Rivers was found higher than those obtained from in Rivers (Hatikakoty, 2002). The reduction in the size and development of oocytes in the fish from Rivers attributed to the affected vitellogenic and maturational enlargement of oocytes (Mousa and Mousa, 1999). The toxicity of heavy metals was presented as a disruption in gonadal development (Gordon et al., 2000). Histochemically, the ovarian tissues of the studied fish collected from Rivers gave a clear information about the internal changes in the total polysaccharide and total protein contents as well as DNA and RNA content and collagen fibers composition which resulted from the severe effect of the heavy metal pollution, when compared with those collected from Rivers as it is a protected area (Mousa and Mousa, 1999).

Conclusion:

The present study documented that toxic heavy metals can alter tissue histological structures, and cause reproduction defects. The environmental impacts on fish from Rivers were delayed spermatogenesis, spermatogenic cell disruption and spermatid reduction. The female fish subjects from Rivers showed atresia, deformed zona radiata, necrotic oocytes and broken tunica albuginea.

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