Metal Uptake, Accumulation In Fish Living In Polluted Waters Impact On Health Risk: Approach To Remove Metal Contamination In Polluted Water Using Conjugated Heterocyclic Ligands

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Abstract

Heavy metals are frequently accumulated in the tissues of fish that live in contaminated waters. In general, the concentration of metals, the length of exposure, the mode of metal uptake, the environmental parameters (pH, salinity, hardness, and temperature of the water), and the intrinsic factors (feeding habits and age of the fish) all affect accumulation. Different metals have varying affinities for the tissues of fish. The majority primarily collect in the kidney, liver, and gills. When compared to other tissues, fish muscles typically have the lowest metal concentrations. Time influences the distribution of metals in different organs. Metal buildup in fish organs can result in functional abnormalities and structural damage. Inductively coupled plasma mass spectrometry (ICP-MS) was used to measure the concentrations of four metals—iron, nickel, cobalt, and copper—in five different fish tissues: the brain, liver, kidney, gills, and muscle. Water samples and ten distinct fish species were also included in the analysis. In order to lower the metal contamination in the contaminated water, metal chelation was applied to the synthesised ligand.

Keywords: Chelation, ligand, Fish, metal.

1. Introduction

Concerns about water contamination and the environment have grown throughout the world throughout the past three decades. Water serves as the foundation for the transport of nutrients across all ecosystems, a process that eventually endangers aquatic life and reaches humans through the food chain (Afshan et al., 2014; Garg et al., 2009; Nagarani et al., 2020). According to Pruß-Ustün et al. (2011), extended exposure to environmental pollution is the cause of about 25% of the diseases that affect humans today. According to reports, the two greatest global issues are waste disposal in aquatic environments and pollution (Anh et al., 2010; Arkoosh et al., 2010).Environmental pollutants represent a risk factor for human and animals in all areas of occurrence in the form of gas, solid and liquid forms as a single or synergistic action (Kovacik, 2017).

By creating metal complexes, heavy metals prevent structural proteins, enzymes, and nucleic acids from performing their activities (Jaishankar et al., 2014). Furthermore, according to Coen et al. (2012), it also causes chromosomal abnormalities, structural or morphological changes, and immune system dysfunction. According to Tao et al. (2001), the solubility, hardness, pH, and ecosystem complexity of heavy metals through their gills, food, and skin are the main physico-chemical characteristics that determine the nature of heavy metal toxicity in fish. Fish constitutes a major source of nutrients for humans. These fish can be the finest bioindicators for aquatic pollution because they were also at the top of the aquatic food chain.

Additionally, because this fish was at the top of the aquatic food chain, it can serve as one of the finest bioindicators of aquatic pollution. Fish can absorb and concentrate metals from the water around them directly, or indirectly, through other animals including small fish, crustaceans, and aquatic plants. Fish at the top of the aquatic food chain tend to accumulate pollutants in their fatty tissues, such as the liver. They can also accumulate metals, which they can then pass on to humans through food, leading to either acute or chronic illnesses (AL-Yousuf et al., 2000). Indeed, there can be serious health hazards for aquatic ecosystems and individuals who consume contaminated fish due to the uptake and buildup of metals in fish living in polluted waters. Bioaccumulation and biomagnification processes can cause heavy metals, including lead, mercury, cadmium, and arsenic, among others, to build up in fish tissues. Fish poisoning can cause a number of health concerns, including as cancer, renal damage, neurological difficulties, and reproductive problems.

A novel strategy for addressing metal contamination in contaminated water bodies and reducing related health hazards is the use of conjugated heterocyclic ligands for metal removal. Organic compounds having heterocyclic rings that have numerous functional groups and the ability to generate strong coordination connections with metal ions are known as conjugated heterocyclic ligands. These ligands can be engineered to attach to particular metal ions with selectivity, making it easier to remove those ions from water by adsorption, precipitation, or complexation.

2. Experimental: Materials and methods

The reactants and other supportive chemicals were procured from Aldrich-sigma, Himedia and utilized as such (No purification needed). The completion of chemical reaction was confirmed using TLC. The silica mesh size (60–120) was chosen and separate the reaction products. The elemental proportion was measured by Carlo Erba-EA1108 analyzer. The proportion of metal contents was measured using AAS. The identification of functional chromophores and metal coordinating sites were confirmed with the aid of Fourier Transform-Infrared spectra (Shimadzu FT-IR affinity-1). The electronic transitions and nature of geometrical arrangements metal chelates were arrived based on electronic absorption spectra. The NMR spectrum of 1,10-Phenanthroline molar conductance and metal complexes were recorded to arrive the nature carbon & proton, coordination environment around the metal ion using BRUKER 400 MHz spectrometer. The molecular weight and nature of fragmentation mode of 1,10-Phenanthroline scaffold and metal chelates were confirmed using mass analyzer JEOL SX 102/DA-6000. The Guoy's electronic balance and systronics conductivity bridge were used to find out the magnetic moment and molar conductance of metal chelates. The electrochemical redox characteristics of metal complexes using CHI 604D instrument and n-Bu_4NClO_4 electrolyte. The stability of metal chelates and structural core was also with the assistance of TGA under nitrogen atm.

2.1 Preparation of Ligand:

Reactants, 2-amino-5-nitrophenol (10 mM) and 1,10-phenanthroline-2,9-dicarbaldehyde (20 mM) are accurately weighed and thoroughly mixed in ethanol. The reaction admixture was stirred and refluxed for 6 hrs. The progress of reaction was checked using TLC. The obtained solid residue was separated through washings and dried (Scheme 1).

1,10-Phenanthroline derivative (H_2L): Molecular Formula: $C_{26}H_{16}N_6O_6$, Mass: 508. Productivity: 75%; CHN analysis: Theoretical; C 61.42, H 3.17, N 16.53; Found: N 16.48, C 61.32, H 3.05, UV (nm): 338, 250. FT-IR (KBr): 3300 υ(O–H); 3080-3070 υ(Ar–H); 2965, 2880 υ(C–H); 1658 cm^{-1} υ(>C=N); 1250 (C–OH). ^{1}H-NMR (ppm): 8.9 (HC=N<, s, 2H), 11.0 (-OH, s, 2H), 6.6-7.8 (Ar-H, m, 12H). FAB-Mass: 509 m/z. ^{13}C-NMR = 112.30, 124.8, 128.0, 132.30, 130.65, 150.10, 162.50 ppm.

Sample collection and preservation

At six in the morning, the local fish market in Local market Nagercoil, Tamilnadu, India, provided the fresh, nutritious fish. The study was carried out in 2019 from January to April. The study employed physical observation to choose the samples, which included being fully grown fish (fingerlings were not included), fresh and devoid of rotting odour, red-colored gills, smooth muscle free of mucus (which would indicate the presence of bacteria or pathogens), and chemical-odor-free. The moment samples were collected, they were kept in ice-cold (4 °C) storage. Each species was represented by three individuals for each analysis. Before being oven-dried for metal analysis, fish were carefully cleaned in distilled water that had been sterilised. Using an atomic absorption spectrometer, the metal concentrations were measured in accordance with the accepted double acid digestion procedures. Using approved solutions (Merck) acidified with HNO3 to the same pH as the samples, standards were created. New samples were kept for upcoming enzymatic research at − 80 °C. For lipid peroxidation analysis, the entire samples were homogenised in trichloro acidic acid (TCA), and for reduced glutathione analysis, in phosphate buffer (pH 7).

Environmental assessment (Degree of pollution) impact

Geoaccumulation index (Igeo)

To understand the current status of the environment and the heavy metal contamination with respect to the natural environment. The following equation applied for calculation of Geoaccumulation Index (Igeo) (Muller et al.)

$$Igeo = Log(Cn/1.5 \times Bn) \quad \text{---- (1)}$$

where, C_n is the measured concentration of metal n in sediment, B_n is the geochemical background concentration of metal[36] n and 1.5 is the background matrix correction factors due to lithogenic effects. As per Muller et al.[35], classified as per Igeo such as If Igeo value is 0 represent unpolluted sediments, Igeo is 0–1 represent unpolluted to moderately polluted, Igeo 1–2, moderately polluted, Igeo 2–3, moderately to strongly polluted, Igeo 3–4, strongly polluted, Igeo 4–5 strongly to extremely polluted and Igeo more than 6 represents extremely polluted.

Contamination index (CI)

Contamination index is used for identification of enriched heavy metals with respect to the maximum admissible limit (MAL) standard as SON[37]; WHO[38]. The contamination index in water samples were calculated as.

CI=[(Concentration of the Total studied metals/MAL of each metal)/total no.of studied metal]

The contamination index is classified as $CI > 5$ contaminated, CI 1–5 slight contaminated and $CI < 1$ represent not contaminated.

3. Results and discussion

3.1 Structural elucidation

The work was carried out to assess the environmental stress in the marine fish at local market. This study also measures the bioaccumulation of pollutants and its effect during transport. The list of fish samples collected from the local market is given in Table 1.

Table 1 List of fish collated to study the environmental risk assessment

S. no	Vernacular name	Scientific name	Class	Order
1	Emperor Long Face emperor bream	*Lethrinus olivaceus*	Actinopterygii	Perciformes
2	Indian goat fish (Nagarai Meen)	*Parupeneus indicus*	Actinopterygii	Perciformes
3	Malabar Trevally/Jack Fish—Paarai Meen	*Carangoides malabaricus*	Actinopterygii	Perciformes
4	Blue Fin Travelly	*Alectis indica*	Actinopterygii	Perciformes
5	Silver Pomfret	*Pampus argenteus*	Actinopterygii	Scombriformes
6	Chaalai or Sardine	*Sardinella longiceps*	Actinopterygii	Clupeiformes
7	Ailai/Dolphin	*Coryphaena hippurus* (Linnaeus, 1758)	Actinopterygii	Perciformes
8	Mural, Needle fish, Viraal, Gar fish	*Hemiramphus far* (Forsskal, 1775)	Actinopterygii	Beloniformes
9	Barracuda (ooli)	*Sphyraena forsteri* (Cuvier, 1829)	Actinopterygii	Scombriformes
10	Kilanga, Lady Fish	*Elops machnata* (Forskal, 1775)	Actinopterygii	Elopiformes

An effective indicator of overall lipid peroxidation is the intermediate of polyunsaturated fatty acid oxidation, malondialdehyde (MDA). Using a spectrophotometer, malondialdehyde (MDA) and thiobarbituric acid generate an adduct that can be measured at 532 nm. Malondialdehyde (MDA) equivalent, which is shown in Fig. 1, is the practical unit of measurement for TBARS. The sample has TBARS ranging from 2 to 200 µM MDA. Alectis indica displayed reduced MDA formation among the Periciformes order. Measured in the reduced form were the non-antioxidant substances glutathione, a portion of glutathione peroxidase, and glutathione reductase enzymes. Figure 2 illustrates that the species under investigation had variable glutathione contents, ranging from 5 to 48 µM.

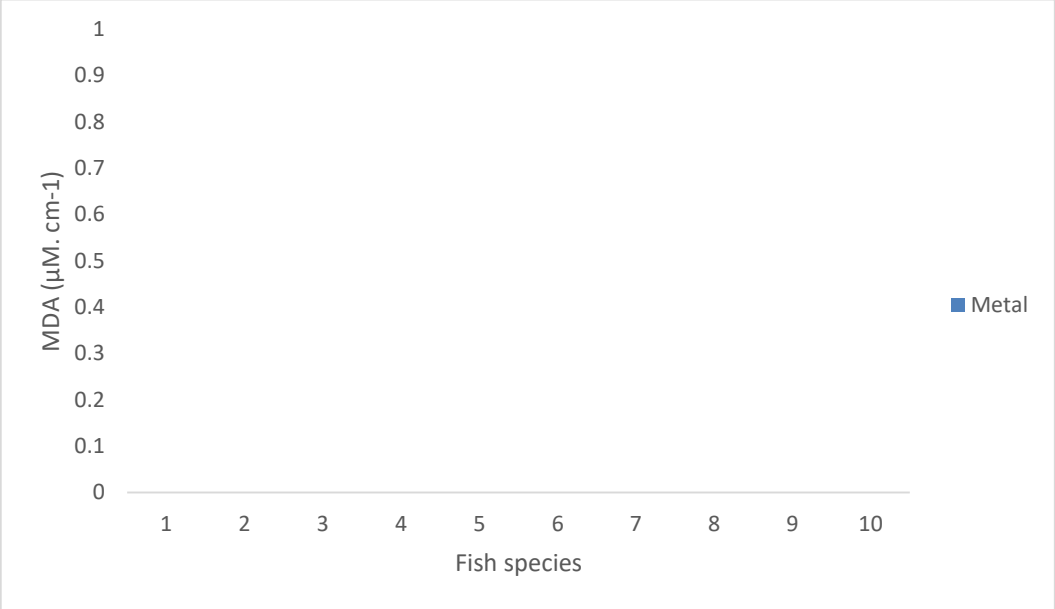

Fig.1 Level of MDA formation in the collected fish

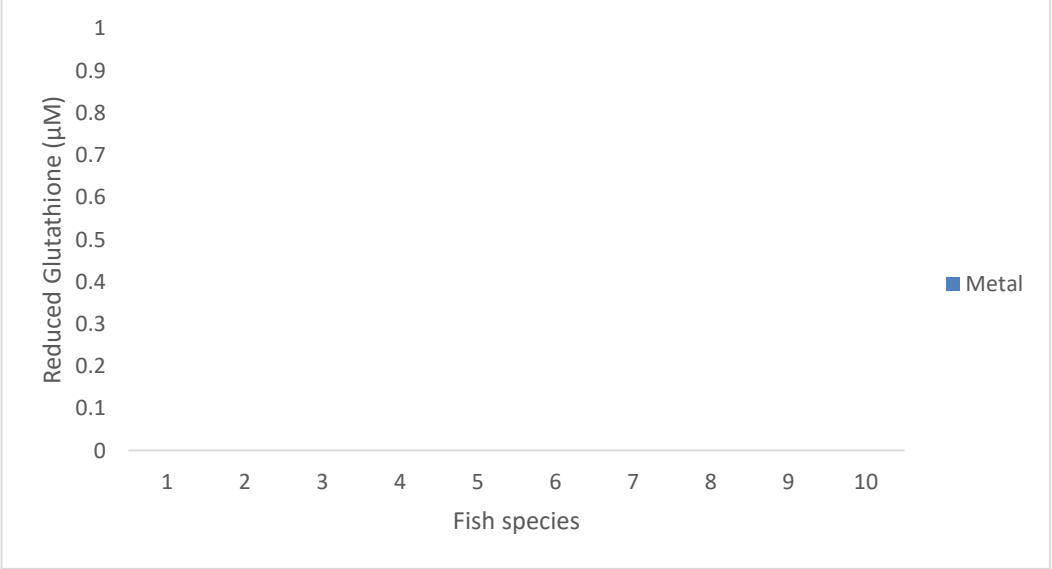

Fig.2 Level of reduced glutathione in the collected fish

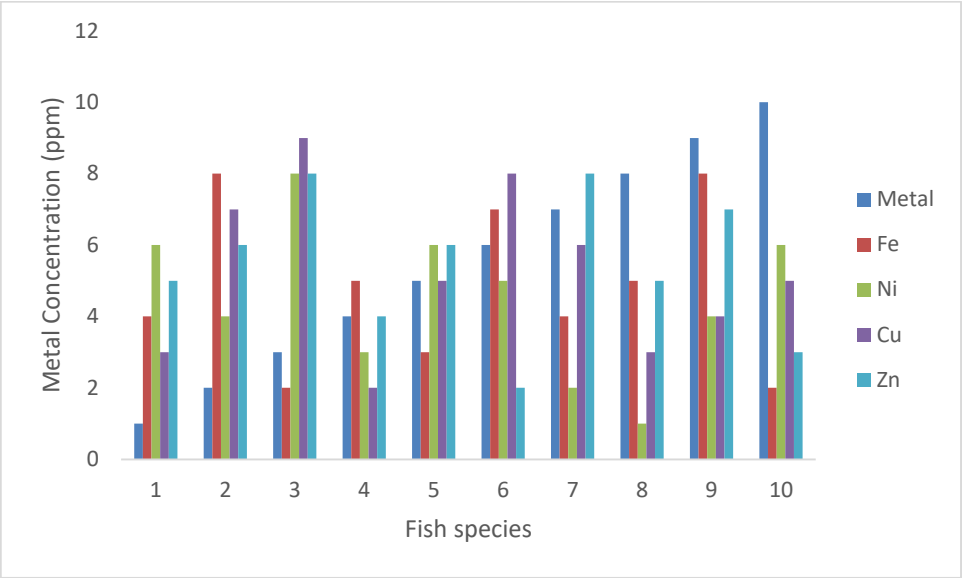

Fig.3 Heavy metal studies in the collected fish samples

Fish from the polluted water may accumulate varying concentrations of metals. Variations in metal buildup between species may be correlated with food and living patterns. Benthivore species had higher levels of zinc and cadmium, according to Kidwell et al. (1995). Voigt (2004) similarly discovered higher mercury contents in predatory fish compared to non-predatory fish. Zinc and lead concentrations were shown to be greater in benthic fish by Ney and Van Hassel (1983). Campbell (1994) found that while benthivores had more cadmium, predators accumulated higher amounts of zinc and nickel.

With the exception of mercury, most metal concentrations in fish are typically inversely correlated with fish size and age. Pseudocrenilabrus philander was used to measure the bioaccumulation of iron, manganese, zinc, copper, nickel, and lead in an impounded mine. The results showed that the body mass of fish and metal concentrations were inversely correlated (De Wet et al., 1994). The age of Coregonus clupeaformis was shown to be inversely correlated with its lead content (Allen-Gill and Martynov, 1995), and the age of Catostomus commersoni was found to be inversely correlated with the accumulation of zinc, lead, cadmium, and nickel (Ney and Van Hassel, 1983).

According to field data, fish's concentration of mercury rises with size and age. Fish age and size-related increases in body mercury levels are likely caused by the metal's attraction for muscle tissue (Goldstein et al., 1996; Munn and Short, 1997; Green and Knutzen, 2003; Voigt, 2004). Numerous authors' data suggest that different metals have varying affinities for different organs. The majority of the total body burdens that are accumulated in the liver, kidney, and gills at varying exposure times and concentrations of metals in the water.

According to several writers (Al-Mohanna, 1994; Kock et al., 1998; Giguere et al., 2004), fish from natural water bodies had significant metal concentrations in their digestive tracts. Elevated amounts of metals in fish gut are associated with the mode of food absorption. Yamazaki et al. (1996) reported that substantial concentrations of copper, chromium, and silver were found in the gall bladder of Carassius auratus. Organs like the brain, gonads, and bones may also exhibit elevated metal levels. In contrast, muscles typically exhibit low metal concentrations but are frequently tested for metals because they are consumed by humans.

Different metals may have varying affinities for different fish organs. In particular, organ-specific accumulations of vital metals including cobalt, iron, zinc, copper, manganese, or zinc are observed. For instance, zinc concentrates in the gonads since these organs are where they carry out their primary metabolic functions, whereas copper exhibits a clear predilection for the liver even at low ambient quantities. However, in situations when there is metal pollution, the metals typically accumulate in the same organs, where they might have harmful effects.The kidney and liver are the main organs where cadmium accumulates, but the spleen, digestive system, and gills can also contain large concentrations of the metal. Lead buildup in the digestive tract, gills, liver, kidneys, and spleen, among other organs.

The distribution of metals in different organs is also related to time. Fish organ accumulation of metals is dependent on uptake and disposal rates, and different organs may exhibit different patterns of metal concentration changes during and after exposure. Because different metals have varying degrees of attraction for different fish species' tissues, the impact of time on the distribution of metals within an organism is a complicated topic. Numerous findings suggest that there may be differences in the dynamics of metal concentrations following exposure and depuration in different organs (Fig. 3). Metal concentrations in the gills rise quickly at the start of aquatic exposure and then typically decrease.

In the study was focused on the extraction or removal of metal ions using conjuagated heterocyclic ligands which contains ionic functionality makes favourable of chelation.

Conclusions

This study evaluated four metals in fish tissues, water samples, and soil sediments from ten different fish at the Nagercoil local market. This study determined the contamination index (CI) in water samples and the geoaccumulation index (Igeo) in soil sediments as indicators of the environmental assessment impact (degree of pollution). Risk assessment is offered for the fish culture in the Nagercoil local market. Thirteen sampling sites were used to detect the presence of the following metals in muscle, gill, liver, kidney, and brain: chromium, vanadium, cobalt, manganese, copper, nickel, zinc, silver, molybdenum, arsenic, selenium, tin, strontium, cadmium, mercury, and lead. Furthermore, below the lethal threshold, metal contamination was also discovered in various tissues, including the brain, kidney, liver, and gills. Metal concentrations in water samples were also determined to be below the hazardous threshold. Fish from metal-contaminated water are generally safe to eat since their muscular tissue accumulates very little metal, with the exception of mercury. However, predatory fish, birds, and mammals that consume contaminated fish may be at risk from these fish. In the end, these investigations need to concentrate on quantifying pollution levels that could cause aquatic habitats to undergo permanent biological alterations. The toxicity levels were moderate up to this point, and they were approaching danger. Both the government and the general public can work to maintain and regulate the activities that unnaturally discharge toxins into the environment in order to preserve a clear and clean environment.

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