Bioaccumulation of Zn in freshwater fishes worldwide and their possible impacts on human health: A review

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Abstract

Zinc (Zn) is an essential micronutrient for many metabolic processes in organisms, including edible fishes. However, excessive Zn bioaccumulation in these fishes can pose potential health risks to humans consuming them. This review explores the bioaccumulation of Zn in edible fishes and their implications on human health. Different species of edible fishes have varied abilities to accumulate Zn in their tissues, depending on factors such as their feeding habits, environmental conditions, and metabolic processes. Numerous studies have investigated the uptake and accumulation of Zn in fishes, aiming to determine safe levels for human consumption. Zn bioaccumulation in fishes is influenced by both environmental and biological factors. Environmental factors such as water quality and sediment composition play a crucial role in determining Zn concentrations in fish habitats. Biological factors such as fish size, age, and species influence the uptake, compartmentalization, and elimination of Zn. Furthermore, Zn bioaccumulation can vary in different fish tissues, with liver, gills, and muscles being the most commonly studied organs. These tissues serve as important indicators for Zn bioavailability and potential risks to human health. Excessive Zn concentrations in fish tissues may result in various adverse effects, including hepatotoxicity and impairment of the immune system. In conclusion, understanding the bioaccumulation of Zn in edible fishes is vital to safeguard human health. This review is a systematic study of Bioaccumulation of heavy metal Zinc (Zn) in edible fishes mainly to understand the harmful effects of Zn Bioaccumulation and Biomagnification. Biomagnification might not show symptoms presently but will eventually appear years down the line if situation keeps worsening at current rates. At that point of time we will hardly have any options of escaping it therefore it is important that we take precautions in advance to keep this from happening. We aim to understand current levels of Zn poisoning in edible fishes worldwide through previous studies and understand how close we are and what should be done to prevent this from happening.

Keywords: - Heavy metal, Bioaccumulation, Biomagnification, Zinc poisoning, health impacts.

Introduction

Heavy metals are pestilential pollutants that are toxic for us. They cause deleterious effects on animals, plants and human beings. Not only that they are persistent in nature but also have the propensity to accumulate in living organisms at various levels through food chain. Heavy metals collect in fish body over the time and increase as the animal ages, a process known as Bioaccumulation. Another process called Biomagnification takes place when these heavy metals are transferred from lower trophic levels to higher tropic levels. Within a Food Web the process of Biomagnification leads to higher concentration in apex predators. They amplify as the trophic levels increases and can lead to health defects when consumed beyond safe consumption limits. In nature metal contaminants are found in low concentrations and are not harmful. High concentrations are due to various anthropogenic activities like effluent of industries, hospitals, sewage treatment etc. Dumping them in our water bodies leads to heavy metal Bio-accumulation and Biomagnification in freshwater fishes making them unfit for consumption. Heavy metal accumulation depends on various water parameters like pH, temperature, salinity, solubility and complexity of the ecosystem. In fishes heavy metal accumulation causes diseases and deleterious effects. Such fishes if consumed directly are harmful for the health of an individual. Metals can enter inside the body of a fish through gills, skin or digestive tract. Within their body, kidneys, liver, gonads and gills are the organs that accumulate higher concentrations in comparison to other parts of the body like muscle [1, 2]. Big fishes harbor higher concentrations of heavy metal in comparison to small ones. The magnitude of accumulation depends upon species, age and trophic level of the fish as well. Bioaccumulation can vary within the same species. Since, fishes are at the top trophic levels of food chains and are consumed by larger part of population they serve as best bioindicators for metal contamination [3]**.** Above threshold levels bioaccumulation of any metal is harmful and it can bring physiological changes that are irreversible and lethal [4]. Zinc (Zn) is a micronutrient and is needed in small quantities for healthy growth but at high concentrations it can lead to deleterious effects on vital organs. Higher concentrations might lead to death as well. Physiochemical characteristics of water also play significant role in metal accumulation [2, 3, 4]. After Iron (Fe), Zn is the most important micronutrients in our body needed for proper functioning of immune system. Seawater and freshwater generally contain very low levels of Zn [5, 6]. Zinc sulphate plays important role in cellular metabolism and Zn acts as co-factor for a number enzymes [7,12]. At higher concentrations Zn has been shown to cause lethal effects. Zn toxicity has been studied in a variety of organisms starting from unicellular algae and bacteria to advanced animals like Zebra Fish and Rainbow trouts. Zn toxicity is caused by the release of Zn ions that disrupt homeostasis [7], release reactive oxygen species (ROS) [8] causing oxidative stress and membrane disorganization [9, 10]. It was observed that organisms are more susceptible to soluble Zn in comparison to suspended solid particles [11] and animal on higher trophic levels are much more susceptible in comparison to lower level micro-organisms [12]. In case of Zebra fish (*Danio rerio*) Zn is required for its embryo development [13] but overexposure leads to developmental delays and inhibited embryonic development. This delay was found to be dependent on the Zn concentrations the animal was exposed to [14, 15]. Use of $ZnCl_2$ (Zinc Chloride) causes hatching delays [12]**.** Zn is capable of reducing Cadmium (Cd) toxicity also by inhibiting its accumulation and reducing hepatotoxicity [16,17] and hence it can be used as countermeasure to check Cd toxicity [18]. Studies using Zn-Fe layered double hydroxides for toxicity assessment are being done to understand its effect on different trophic levels [19]**.** Most studies involve algae, bacteria, zooplanktons, lower crustaceans or small fishes like Zebra fish [12-17]. Very few studies involve freshwater fishes that contribute as food and hence actual effects of Zn poisoning on higher trophic level fishes are not very clearly understood. There have been very few studies on rainbow trouts and carps where their response to acute exposure of Zn was studied [20, 38, 40]. Permissible amount of Zn in freshwater fish is 50 mg/kg of fresh meat [21]. Zn easily enters in early developmental stages like larvae and eggs. Gametes and small fry are considered most sensitive to Zn [22]. Within fish body Zn accumulates in gonads, liver and gills mainly [23, 24, 25]. Fishes play important role in assessing the condition of a contaminated water body. They are very sensitive to such change in their environment. Upon exposure to sub-lethal concentrations, the heavy metal enters their body and cause deleterious changes in their physiology [26]**.** In recent years the accumulation of heavy metals in aquatic ecosystems has become an issue of great concern world-wide. Heavy metals accumulate to very high toxic levels and cause serious effect on the aquatic organisms without any visible signs. Expansion in urbanization, population, agricultural practices and industrialization have further aggravated the situation [27, 28, 29, 30]. The effects are visible now and therefore it is of considerable interest. Edible freshwater fishes like *Clarias batrachus* are good source of protein, vitamin -D, amino acid, calcium, iodine, omega-3 polyunsaturated fatty acid, and more [31, 32, 33]**.** It is found in tropical and temperate waters with temperatures 18 degree

Celsius and above. It is an omnivore (eats food of both animal and plant origin), detritivore (feeds on dead organic material, especially plant detritus) and an opportunistic feeder that prefers shallow waters. It is available in India and is one of the most important dietary component consumed by humans. Therefore, they can also be the biggest contributors to heavy metal poisoning threat. Best biomarker of metal pollution in the aquatic ecosystems and hence best models to study heavy metal poisoning as well [34, 35, 36, 37, 38]**.** Water affects various aspects of the society. The focus on heavy metals and their detection has been much discussed these days because of the excessive leaching of these non-biodegradable metals. According to the recent report of the Central Water Commission (CWC), from May 2014 - Apr 2018, two-third of drinking water samples have been found to be contaminated and polluted by deadly heavy metals [41]. In this review we aim to summarize the studies done on Zn bioaccumulation in freshwater fishes.

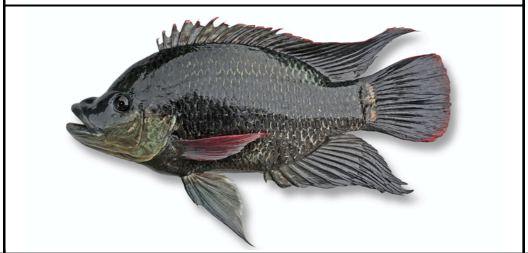
Review Methodology

The purpose of this systematic review is to examine the existing literature on bioaccumulation of Zn in freshwater fishes. Zn deficiency is relatively rare in developed countries but can occur in certain populations, such as those with malnutrition, gastrointestinal diseases, or genetic disorders that affect Zn absorption. Sufficient Zn is of particular significance to the vulnerable system. Zn influences the lymphocyte response to mitogens and cytokines, serves as a co-factor for the thymic hormone, and is involved in leukocyte signal transduction, T cell activity and stimulates monocytes to secrete pro-inflammatory cytokines [50]. Serious Zn deficiency can be inherited or acquired. ***Acrodermatitis***, a rare autosomal recessive metabolic disorder caused by a mutation in the intestinal Zip4 transporter, is the most severe of the inherited forms [51]. Skin lesions, alopecia, diahorrea, neuropsychological disturbances, weight loss, reduced immune function, and hypogonadism in men all are symptoms of this condition, which can be fatal if not treated [52, 53, 54]. Zinc (Zn) bioaccumulation can have significant impacts on aquatic ecosystems. When Zn enters a freshwater ecosystem through various sources such as industrial runoff, agricultural activities, or atmospheric deposition, it can accumulate in the sediment and water column and taken up by aquatic plants and algae, which form the base of the food chain. This can lead to adverse effects on organisms at higher trophic levels, including fish, birds, and mammals. In aquatic organisms, such as fish, Zn bioaccumulation can cause a range of physiological and behavioral effects that can be transferred to humans as food. Studies all over the world report accumulation of heavy metals. In order to do this review we searched PubMed database using keyword "Zinc bioaccumulation and freshwater fishes" and found 63 publications. 58 were found relevant to this study.

Bioaccumulation studies done on fishes *Pelmatochromis guentheri* and *Pelmatochromis pulchur* fish of Mbaa river, Nigeria indicated that concentration of heavy metal Zn was way higher as compared to recommended limits by World Health Organization (WHO) and Food and Environment Protection Act (FEPA) indicating that fishes from this river are not fit for human consumption anymore [63]. Similar studies were done on 5 marine fish species of Riga, Latvia, to study effects of heavy metals Zn, Pb and Mercury (Hg). They tried to co-relate concentration of heavy metals in fish tissues with fish body size at different locations and time periods. They could do so by degrading performance of their statistical tests yet limited success was achieved specially in case of pelagic fishes [64].

Table 1A: Heavy metal studies done around the world and contribution of Zn bioaccumulation in contaminating our aquatic ecosystems. Out of all publications discussed below one is from marine water and rest are from freshwater environments. Places included are Mbaa River (Nigeria), Latvia, Paraoppeba river (Brazil), Tamil Nadu (India), China, Egypt, Taiwan, Nile river (Egypt), Pakistan. Iran and Brazil [63-77]. Fish images were taken from database FishBase (https://www.fishbase.se/).

S.No.	Zoological name of freshwater fish	Location and Heavy Metals	Freshwater fish specimen	Reference
1	*Pelmatochromis guentheri* and *Pelmatochromis . pulchur*	Mbaa river, Nigeria Zn, Pb, Cd and Cu		[63]
2	5 Marine fish species	Latvia, Zn, Pb and Hg		[64]
3	*Geophagus brasiliensis* (Pearl Cichlid)	Brazil **Muscles:** Al > Cu > Zn > Fe > Co > Mn > Cr > Ag > Ni > Pb > Cd > As		[65]
4	*Pseudoplatystoma corruscans* (Catfish)	Paraopeba River, Brazil Hg, Cd, Zn, Cr, and Pb		[66]
5	*Prochilodus lineatus*	Brazil Zn, Mn and Fe		[67]
6	*Rhodeus sinensis* (Chinese bitterling) and *Ctenogobius giurinus/ Rhinogobius giurinus*	China Zn, Cu, Pb, Cd, As, and Hg	 	[68]
7	*Cyprinus carpio,*	Tamil Nadu, India ZnO Nanoparticles		[69]
8 9	*Orechromis niloticus*	Taiwan Zn and Cu Nile river, Giza, Egypt (near metal related factories), Cu, Zn, Pb, Fe, Mn and Cd		[70] [116]
10	30 marine water fish species	New Ferry Whorf, Sassoon dock and Versova (Mumbai) Zn, Cr, Mn, Co, Cu, Se, As, Sr, Cd, Sn, Sb and Pb		[71]

11	*Schizothorax plagiostomus*	River Swat, Pakistan Zn, Pb, Cr and Ni		[72]
12	*Carassius gibelio* and *Esox lucius*	Anzali, Iran Zn>Cu>Pb> Cr> Cd. **Tissues**: liver > kidney > gill ~ intestine > muscle	 	[73]
13	*Pelteobagrus. fulvidraco*, *O. mossambicus* and *Cyprinus carpio*	Dabaoshan mine in South China Zn, As, Cd, Cr, Cu, Pb, Ni and Tl	 	[74]
14	*Bryconamericus iheringii*	Sinos river, Brazil Zn, Al, Fe and Pb		[76]
15	*Cyprinus carpio*	China Zn and Cd		[77]

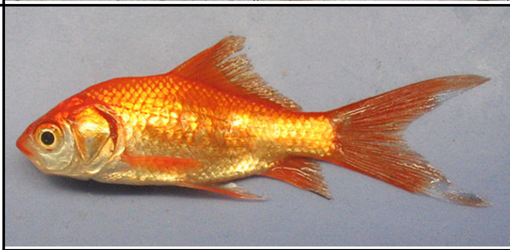
From Brazil it was reported that Zn bioaccumulates in tissues of fishes more than most heavy metals as indicated by gills, liver, and muscles of *Geophagus brasiliensis.* High levels of Zn can impair growth and reproduction, disrupt functioning of vital organs, and compromise immune systems [65]. In *Pseudoplatystoma corruscans* fish from Paraopeba River, Brazil, it was found that viscera (liver and spleen) accumulate more of heavy metals [66]. In *Prochilodus lineatus* a mixture of Zn, Mn and iron (Fe) exposure lead to oxidative stress and DNA damage of blood cells [67]. Zn showed highest bioconcentration factor (BCF) in two small size fishes, *Rhodeus sinensis,*and *Ctenogobius giurinus* in a study from Southwest China [68]. In *Cyprus carpio,* Zinc Oxide (ZnO) nanoparticle exposure alters hematological parameters, induces reactive oxygen species (ROS) and alters histomorphology [69]. In *Orechromis niloticus* from Taiwan, highest bioaccumulation factor (BAF) was estimated to be above 1000 for Zn and was attributed to industrial activities in the area [70] From Giza, Egypt, same fish in metal factory areas of Nile showed heavy metal concentrations that were within safe limits despite high concentrations in the surrounding waters [116]. Heavy metal contamination isn't limited to fresh water bodies, even marine aquatic system is being reported as contaminated by heavy metals on the basis of study done on 30 marine fish species collected from New Ferry Whorf, Sassoon dock and Verso fishing harbour in Mumbai, India [71].

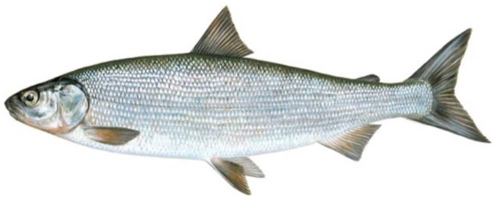
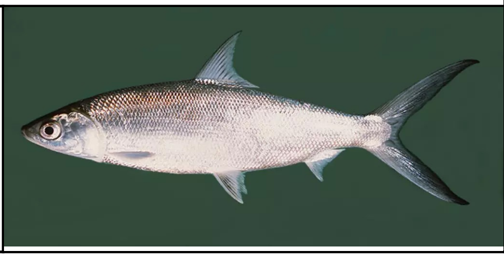
Snow trout (*Schizothorax plagiostomus*) from River Swat of Pakistan report Zn as major bioaccumulating heavy metal after lead (Pb), and Nickel (Ni) [72]. In freshwater fishes *Caracas gibelio* and *Esox lucius* metal concentrations in descending order were: Zn> Cu > Pb > Cr > Cd, similarly tissues followed: liver > kidney > gill ~ intestine > muscle [73]. In rivers near Dabaoshan mine in South China metals in tissues followed: intestines > gills > muscles and nearly all intestine tissue samples exceeded the safe limits [74]. Asian clam, *Potamocorbula amurensis*, is a preferred food of the sturgeon, *Accipenser transmontanus* and effective bioaccumulator of Selenium (Se) which increases Se loads in sturgeon [75]. Similarly higher levels of Zn were found in *Bryconamericus iheringii* caught from Sinos river, Brazil. Histopathological alterations were observed in their gills as well [76]. Zn-enriched *Bacillus cereus* has the potential to combat Cd toxicity in mirror carp *Cyprinus carpio* [77].

Heavy metal accumulation in *Labeo rohita*, (liver>kidney>gills>muscles) and *Ctenopharyngodon idella*, (gills>liver>kidney>muscles) from Bhopal showed Zn accumulates highest while Hg is lowest [78]. In Shaoguan, China the freshwater fishes of constructed wetlands show high amounts of heavy metal accumulation [79]. Another interesting study that came out in two strains of *Oreochromis niloticus,* suggested that Zn bioccumulation can be regulated by certain strains of fishes [80].Such species can be cultured in water bodies where heavy metal pollutants are present. Freshwater fishes from Kolkata were found high in Pb values, Zn and Cd were found below the water quality guideline levels for the protection of freshwater aquatic life proposed by CEQG (Canadian Environmental Quality Guidelines) and AENV (Alberta Environment)[81]. From Slovakia, fish *Perca fluviatilis* and its parasites were analyzed for heavy metals As, Cd, Cr, Cu, Hg, Mn, Ni, Pb and Zn. Organs used were muscle, liver, kidney, brain, reproductive organs and adipose tissue. It was observed that Zn concentrations were exceeding all other metals. Among the organs, kidney received the highest concentrations [82]. In case of whitefish (*Coregonus lavaretus*) from subarctic lake Imandra, Ni, Al and Sr were found in high concentrations in fish organs. These organs during the period of heavy concentrations showed symptoms like nephrocalcinosis (deposition of calcium in kidney), scoliosis (sideway curvature of spine), and myopathy (disease of muscles that control voluntary movement in the body) [83]. A study from Lake Nahuel Huapi, an unpolluted ultra-oligotrophic system in North Patagonia suggests that even though Zn present in volcanic ash leached into this lake in 2011 still post 2 years of recovery the lake showed no signs of significant increase in Zn concentration. This is first report of Zn bio dilution in lacustrine environments [84]. Sewage treatment plants also play role in adding metals to our water bodies. A study done on Ripoll River, Catalonia, Spain shows heavy metals Fe, Hg, Cd, Zn, Pb, Ni and Cu accumulation in the liver and muscles of *Barbus meridionalis* also known as Mediterranean barbel [85].

In fresh water because Ca^{2+} and Zn^{2+} compete for uptake at the gills Ca ameliorates Zn toxicity. Combination of 100 % salinity and High Ca always protects against Zn toxicity as shown in fish *Fundulus heteroclitus* (killifish) in both freshwater and marine environments [86]. Toxicity caused by quantum dots (nonmaterial) of ZnS was evaluated in freshwater fish *Danio rerio* and it was found that when applied individually they cause oxidative stress in fishes [87].

Table 1B: Heavy metal studies done around the world and contribution of Zn bioaccumulation in contaminating our aquatic ecosystems from Bhopal (India), Shaoguan (China), Rizal (Philippines), Kolkata (India), Košice (Slovakia), Subarctic Lake Imandra, North Patagonia, Ripoll river (Catalonia Spain), Caparica (Portugal), Finniss river (Sydney, Australia), Hugli river (West Bengal, India), and Southwest Taiwan [78-90]. Fish images were taken from public databases like FishBase (https://www.fishbase.se/).

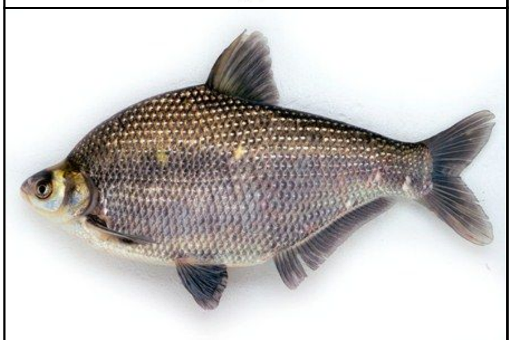
S.No.	Zoological name of freshwater fish	Location and Heavy Metals	Freshwater fish specimen	Reference
16	*Labeo rohita* and *Ctenopharyngodon idella* (grass carp)	Bhopal Zn, Pb, Cd, Ni, Cu, Cr and Hg		[78]
17	*Carassius auratus* (Gold fish), *Cirrhinus molitorella* (mud carp or dace), *Ctenopharyngodon. idellus* (grass carp), *Cyprinus carpio, Sarcocheilchthys kiangsiensis*	Shaoguan (Guangdong, China) Zn, Pb, Cu and Cd.		[79]
18	*Oreochromis niloticus*	Rizal Philippines Zn, Cd and Mercury		[80]
19	*L. rohita, C. catla, Cirrhinus mrigala, O. mossambicus, and C. carpio*	Kolkata, India Zn, Pb and Cd		[81]

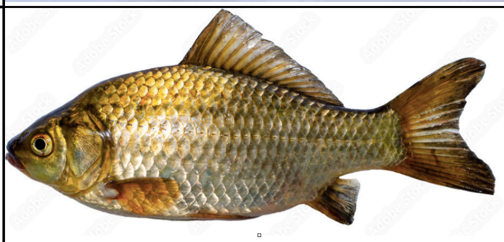
S.No.	Zoological name of freshwater fish	Location and Heavy Metals	Freshwater fish specimen	Reference
20	Perca fluviatilis	Košice, Slovakia. Zn>Cu>Mn>Hg>As> Cr>Cd>Ni>Pb		[82]
21	*Coregonus lavaretus*	Subarctic Lake Imandra Zn, Cu, Al, Sr, Cr, Pb, and Hg		[83]
22	Freshwater fish and macro invertebrates.	North Patagonia, Zn		[84]
23	*Barbus meridionalis*	Ripoll river, Catalonia Spain. Zn, Fe, Hg, Cd, Pb, Ni and Cu		[85]
24	*Fundulus heteroclitus* (Killifish)	Canada Zn and Ca		[86]
25	*Danio rerio* (Zebra fish)	Caparica, Portugal ZnS quantum dots (nanomaterial)		[87]
26	*Nematalosa erebi* (Bony bream) and *Neosilurus ater* (black catfish)	Finniss river, Sydney, Australia Co, Cu, Pb, Mn, Ni and Zn	 	[88]
27	Freshwater fish	Hugli river, West Bengal, India. Ca, K, Mg, Fe, Zn and Se		[89]
28	Chanos chanos (Milk fish)	Southwest Taiwan Zn, As and Cu		[90]

From Sydney, Australia diminished metal accumulation was reported in fishes *Nematalosa erebi* (Bony bream) and *Neosilurus ater* (black catfish) that were exposed to acid mine drainage containing high concentrations of mine-related metals (Co, Cu, Pb, Mn, Ni, U and Zn) for over five decades. It was interpreted that both fish species have probably modified their kinetics via adaptation or tolerance responses and reduced their body burdens of metals. [88]. Due to pollution caused by tanneries along Hugli river in West Bengal, India, wild caught fishes are high in concentrations of Fe, Ca and Zn as compared to the farmed fishes [89] which is alarming and needs to be addressed in public interest. As, Zn and Cu levels in milk fish (*Chanos chanos*) in Southwest Taiwan were found positively co-related to the concentrations found in pond watering were enough to pose carcinogenic risk [90].

A study on omnivorous and carnivorous fishes done in Pearl River Delta, China indicate that there was no health risk associated to ingestion of the freshwater fish from the river as heavy metal concentration decreases heavily with increasing trophic levels. Omnivorous fishes showed more metal accumulation as compared to carnivorous fishes [91]. East Kolkata Wetlands (EKW) also known as Ramsar site is a waterbody where wastewater-fed natural fish aquaculture showed that heavy metals bioaccumulation in fish muscles here are within safe limits for human consumption as recommended by WHO and FAO [92]. Methylmercury concentration in *Micropterus dolomieu* and *Hypentelium nigricans* was found high enough to be considered as threat to human health [93]. *Thryssa vitrirostris* and *Johnius belangerii* showed high levels of Zn is their liver tissues. Highest level of metals were found in pelagic species in two estuaries and Arvand river of northeast Persian Gulf [94]. Livers of Carp species found in ponds near Dabaoshan mines of South China contained heavy metals in high concentrations. The BAF in descending order was found to be Cd>Zn>Cu>Pb for site 1 and Zn>Cd>Cu>Pb for site 2 suggesting potential human health risk [95]. A study done on gill ectoparasites of two yellow fishes *Labeobarbus aeneus* and *Labeobarbus kimberleyensis* suggested that bioaccumulation of Zn in these parasites was 15 to 90 times more as compared to their host fishes [96]. Dimon lake, Italy being a mountain lake was found less polluted and presence of heavy metals was found way lesser in *Cottus gobio* a predator fish as compared to Diptera Chironomidae suggesting biodilution and not bioaccumulation along the increasing trophic levels [97]. Liver, gills and muscle tissues of *Schizothorax niger* and *Cyprinus carpio* from river Jhelum of Kashmir Himalaya were studied for heavy metals. Zn was found highest in the liver tissue and was recorded lowest in muscle tissue in *Schizothorax niger* whereas in *Cyprinus carpio* Zn was recorded highest in liver and lowest in gills [98]. Heavy metal assessment of muscles of *Oreochromis amphimelas* and *Clarias gariepinus* from Lake Manyara, Tanzania showed that fish from this area are safe in terms of metal toxicity [99]. A study done on *Solea senegalensis* from Sado estuary, Portugal suggests that although bioaccumulation depends on sediment contamination it is not linearly dependent. Heavy metal uptake depends upon its bioavailability and synergistic effects in organisms [100]. Studies done on two edible species of *Carassius carassius* and *Triplophysa kashmirensis* from Srinagar, India revealed that heavy metal concentrations are higher in small size fishes as compared to the large size fishes of the same species. They found Zn concentrations in these fishes high enough to pose a threat to human health upon consumption [101]. From France comes a study where they are trying to understand the mechanism of bioaccumulation and their interdependencies in *Brachydanio rerio* [102]. Data from a Copper Mine area in Purple mountains of Southeast China done on *Ctenopharyngodon idellus* suggest high concentrations of Zn in liver, gill and kidneys. Some of the values exceed safe limits for human consumption [103].

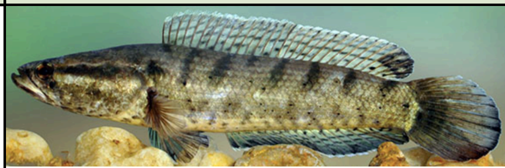
Table 1C: Heavy metal studies done around the world and contribution of Zn bioaccumulation in contaminating our aquatic ecosystems from Pearl River Delta (China), Ramsar Site, Kolkata (India), Ozark stream, Columbia (USA), Persian Gulf, Dabaosan mine, (South China), Auckland Park (South Africa), Dimon Lake (Carnic Alps, Austria, Italy), Jhelum River (Himalaya, India), Lake Mnayara (Northern Tanzania), Sado estuary (Portugal), Dal Lake (Srinagar, Kashmir, India), Talence (France) and Purple Mountain Cu mine in Southeast China (References 91-103). Fish images were taken from public databases like FishBase (https://www.fishbase.se/).

S.No.	**Zoological name of freshwater fish**	**Location and Heavy Metals**	**Freshwater fish specimen**	**Reference**
29	Omnivorous and Carnivorous fishes	Pearl River Delta, China, Zn, Cd, Pb, Cr, Cu, Ni and Mn		[91]
30	Freshwater fish	East Kolkata Wetlands or Ramsar site, Cr, V, Co, Mn, Co, Ni, Zn, Ag, Mo, Ar, Se, Sn, Ga, Ge, Sr, Cd, Hg, and Pb		[92]
31	*Hypentelium nigricans* (northern Hog sucker) *Micropterus dolomieu* (smallmouth bass)	Ozark stream, Missouri, Columbia, MO, USA. Hg	 	[93]
32	*Thryssa vitrirostris* and *Johnius belangerii*	Gulf Arvand river, Meleh estuary and Musa estuary in the northeast Persian Gulf. Zn, Cd, Cu, Ni and Pb	 	[94]
33	5 Carp species: *Hypophthalmichthys molitrix, Ctenopharyngodon idellus, Megalobrama amblycephala, Aristichthys nobilis,* and *Carassius auratus auratus*	Ponds near Dabaoshan mine, South China. Zn, Pb, Cd and Cu	  	[95]
34	Ectoparasites of *Labeobarbus aeneus* (Smallmouth yellow fish)	Auckland Park, South Africa Zn and Cu		[96]

S.No.	Zoological name of freshwater fish	Location and Heavy Metals	Freshwater fish specimen	Reference
34	and *Labeobarbus kimberleyensis* (Yellow fish)			
35	*Cottus gobio*	Dimon Lake, Carnic Alps, Austria, Italy. As, Cd, Pb, and Zn		[97]
36	*Schizothorax niger* (snowtrout) and Cyprinus carpio (common carp)	River Jhelum of Kashmir Himalaya Zn, Cu, Pb and Fe		[98]
37	*Oreochromis amphimelas* and *Clarias gariepinus*	Lake Mnayara, Northern Tanzania Cu, Pb, Ni and Zn	 	[99]
38	*Solea senegalensis*	Sado estuary, Portugal. As, Cd, Cr, Cu, Ni, Pb and Zn		[100]
39	*Carassius carassius* (Crucian carp) and *Triplophysa kashmirensis* (Thick lip loach)	Dal Lake, Srinagar, Kashmir, India. Cd, Mn, Cu, Zn, Pb, and Cr	 	[101]
40	*Brachydanio rerio*	Talence, France Cu, Zn, Ag, Se, and methyl Hg		[102]
41	*Ctenopharyngodon idellus* (grass carps)	Purple Mountain Cu mine in Southeast China. Cr, Ni, Mn, Cu, Zn, As, Cd,Hg and Pb		[103]

Two cat fish species, *Channa punctatus* and *Aorichthys aor* from river Ganges, Prayagraj, India were studied and it was found that Zn accumulates maximum in muscles of fishes as well as in sediments [104]. Studies done on fish tissues of Busko Blato, a shallow reservoir located in Bosnia and Herzegovina taking four fish species *Aulopyge huegelii* (Dalmatian barbel gudgeon), *Chondrostoma nasus* (the nase) , *Leuciscus souffia* (the souffie) and *Salmo trutta* (brown trout) indicated all heavy metals within the safety range [105]. From river Kabul Khyber Pakhtunkhwa Pakistan, freshwater fish *Cyprinus carpio* tissues indicated levels of Zn beyond permissible limit. Zn was found to be accumulating maximum. Metal burden is in sequence blood > intestine > skin > liver > gills > muscles [106].

Table 1D: Heavy metal studies done around the world and contribution of Zn bioaccumulation in contaminating our aquatic ecosystems from Prayagraj (India), Busko Blato (Bosnia and Herzegovina), and Khyber River, Pakistan (References 104-106). Fish images were taken from public databases like FishBase (https://www.fishbase.se/).

S.No.	Zoological name of freshwater fish	Location and Heavy Metals	Freshwater fish specimen	Reference
42	*Channa punctatus* and *Aorichthys aor*	University of Allahabad, Prayagraj, India. Cu, Cr, Cd, Pb and Zn	 	[104]
43	*Aulopyge huegelii* (Dalmatian barbelgudgeon), *Chondrostoma nasus* (the nase) , *Leuciscus souffia* (the souffie) and *Salmo trutta* (brown trout)	Busko Blato reservoir Bosnia and Herzegovina Hg, Pb, Cd, As, Cu, Zn and Cr	   	[105]
44	*Cyprinus carpio* (Common carp)	River Kabul Khyber Pakhtunkhwa Pakistan, Zn, Ni, Cr, Cd, Pb and Hg		[106]

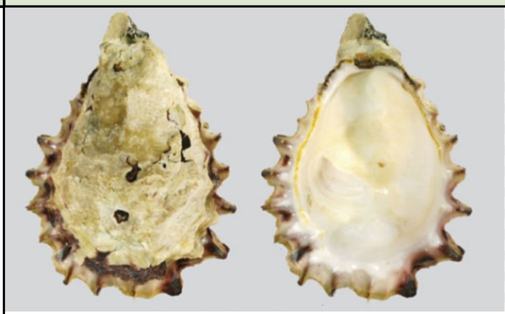
From Prague, Czech Republic report of Acanthocephalan parasite (*Acanthocephalus lucii*) that feed on freshwater fish *Perca fluviatilis* accumulate way more amount of heavy metals than the fish itself. Authors state that this might benefit the host by saving it from excess heavy metal accumulation [107]. Using N-isotope analysis, a study done in New England on freshwater food web that included zooplanktons and freshwater fishes indicated that Zn concentrations were higher in fish as compared to zooplanktons, but overall variation in Zn concentration was found to be low [108]. In Red swamp crayfish (*Procambarus clarkii*) from River Guadiamar, Iberian Peninsula overall metal concentration was found in the order : hepatopancreas/viscera>exoskeleton/gills>abdominal muscle. Zn and Cu being essential metals were found in high concentrations and independent of their quantities in the environment [109]. Such studies were also reported in prawns *Macrobracium* species [115]. Aznalcóllar mine, owned by the Canadian-Swedish Company Boliden Ltd. burst its banks into the nearby River Guadiamar of south Iberian Peninsula. High concentrations of metals, especially Zn, entered the River Guadalquivir. A biomonitoring program has been carried out to evaluate the incidence of this spill on the fauna of the River Guadalquivir. One month after the accident, concentrations of Zn were higher in C. angulata and in Palaemon longirostris [110]. Process of effect of acclimation was studied in juvenile *Oncorhynchus mykiss* (Rainbow trout). Cu exposed trout shows cross-acclimation whereas Zn exposed did not [111].

Table 1E: Heavy metal studies done around the world and contribution of Zn bioaccumulation in contaminating our aquatic ecosystems from Prague (Czech Republic), New England, Guadiamar river (Spain), River Guadalquivir (Iberian Peninsula) and Canada [107-111]. Fish images were taken from public databases like FishBase (https://www.fishbase.se/).

S.No.	Zoological name of freshwater fish	Location and Heavy Metals	Freshwater specimen	Reference
45	*Perca fluviatilis* and *Acanthocephalus lucii* (acanthocephalan parasite)	Prague (Czech Republic) Cd, Cu, Mn and Zn		[107]
46	Freshwater food web	New England lakes,	As, Cd, Hg, Pb, and Zn	[108]
47 48	*Procambarus clarkii* (red swamp crayfish) *Macrobrachium australiense*, *M. rosenbergii* and *M. latidactylus* (prawns)	River Guadiamar, SW, Spain. Cd, Cu, Zn, Pb and As		[109, 115]
49	*Liza ramada*	River Guadalquivir, Iberian Peninsula Cd, Cu, Zn		[110]
50	*Oncorhynchus mykiss* (Juvenile Rainbow trout)	Waterloo, Canada Zn, Cu and Cd		[111]

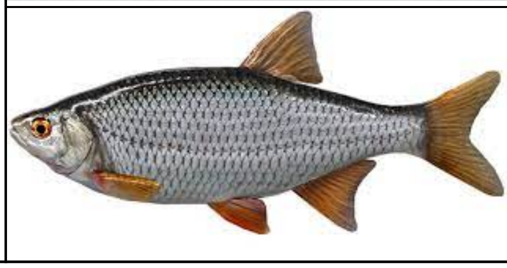
Another study done on a seagrass ecosystem in Australia, denies Biomagnification. Benthic microalgae had higher metal concentrations than those that fed on detritus [112]. Study done in the area of Gangetic delta on shellfish and Finfish indicates that bioaccumulation i species dependent [113]. Four fish species of family Cyprinideae from Enne Dame Lake Kütahya,Turkey were analysed for heavy metal contamination using inductively coupled plasma-optic emission spectroscopy (ICP-OES), and atomic absorption spectrophotometer (AAS). Bioaccumulation factor for Zn was found remarkably high [114]. In a study done near Animas River watershed Colorado, USA associated with acid drainage and hence heavy metal pollution indicated that highest concentrations of Zn, Cu and Cd were in *Rithrogena* (Mayfly) that feeds on periphyton and detrivores. Zn concentration decreased with each trophic transfers unlike Cu and Cd [117]. Acanthocephalan endoparasite, *Pomphorhynchus laevis* of the *Barbrus barbuls* (cyprinid barbel) and the host fish were tested for heavy metal contents and it was fond that in comparison to host the parasites had way more heavy metal load. It was concluded that these parasites are most sensitive as indicators. [118].

Table 1F: Heavy metal studies done around the world and contribution of Zn bioaccumulation in contaminating our aquatic ecosystems from Australia, Gangetic Delta (Kolkata, India), Dame Lake (Turkey), Animas River (Colorado, USA) and Austria (References 112-118). Fish images were taken from public databases like FishBase (https://www.fishbase.se/).

S.No.	**Zoological name of freshwater fish**	**Location and Heavy Metals**	**Freshwater specimen**	**Reference**
51	Seagrass ecosystem (oyster *Saccostrea glomerata*, seagrass *Zostera capricorni*, mud oyster *Ostrea angasi* and fish)	Australia Zn, Cu and Cd		[112]
52	Finfish and Shellfish	Gangetic Delta, Kolkata, India. Zn, Cu, Pb and Cd		[113]
53	*Carassius carassius* (Crusian carp), *Chondrostoma nasus* (Common nase), *Leuciscus cephalus* (European chub) and *Alburnus alburnus* (Common bleak)	Enne Dame Lake, Kütahya, Turkey Cu, Zn, Cd,Mn, Fe, Co, Mg, Ni, Cr and B (ICP-OES and AAS).	 	[114]
54	Salvelinus fontinalis (Brook trout) liver, Benthic invertebrates and Periphytons	Animas River, Colorado, USA. Zn, Cd, Cu and Pb.		[117]
55	Endoparasites (*Pomphorhynchus laevis*) of the *Barbrus barbuls* (cyprinid barbel)	Austria Zn, Cd and Pb.		[118]

Palaemonetes pugio and *Nereis acuminata,* two common preys of fish *Fundulus heteroclitus* (mummichogs) from chronically metal-polluted salt marshes in New York, USA, showed higher concentrations of non-essential metals that are less trophically available whereas essential metals (Cu and Zn) varied relatively little [119]. From Nitra River, Slovakia a study of 5 freshwater Cyprinid fishes revealed that heavy metal concentration in muscles of these fishes was in order Zn>Cu>Fe>Mn>Ni>Pb>Cd [120]. Gill, liver, kidney, intestine and muscle of edible fishes captured from Kharkai river, Jamshedpur were analyzed for heavy metals. They suggest heavy metal pollution beyond safe limits and call for immediate addressing of the problem [121].

Table 1G: Heavy metal studies done around the world and contribution of Zn bioaccumulation in contaminating our aquatic ecosystems from Salt Marshes, New York (USA), Nitra River (Slovakia), and Kharakai River (Jamshedpur) [119-121]. Fish images were taken from public databases like FishBase (https://www.fishbase.se/).

S.No.	Zoological name of freshwater fish	Location and Heavy Metals	Freshwater specimen	Reference
56	*Fundulus heteroclitus* (mummichogs)	Chronically metal-polluted salt marshes in New York, USA. Cd, Cu, Pb, and Zn		[119]
57	Cyprinid fish species: *Gobio gobio* (Gudgeon), *Leuciscus cephalus* (European chub), *Barbus barbus* (Common Barbel), *Rutilus rutilus* (Common roach), *Chondrostoma nasus* (common nase)	Nitra River, Slovakia Zn>Cu>Fe>Mn>Ni>Pb>Cd	 	[120]
58	Freshwater edible fishes	Kharkai River, Jamshedpur, Zn, Fe, Ni, Pb, Cu, Mn, Cr and Co.		[121]

Results and Conclusions

Scientists around the world consider Zn as an important heavy metal pollutant responsible for contamination of our water bodies. Countries around the world are reporting adverse effects of Zn bioaccumulation on freshwater fishes. Zn bioaccumulates most among heavy metals and in high concentrations can impair growth and reproduction of fishes. Zn can cause oxidative stress and DNA damage. Viscera (liver and spleen) accumulates more of heavy metals as compared to muscles. Different studies report different sequences of bioaccumulation in fish tissue. Even Zinc Oxide (ZnO) nanoparticles are capable of altering hematological parameters and histomorphology of fish organs. Quantum dots of ZnS cause oxidative stress in fishes. Fish parasites and prey accumulate Zn 15 to 90 times more than their host fishes [96, 107, [115]. Zn metabolism is complex. It can help combat Cd toxicity as a supplement. Ca ameliorates Zn toxicity. Reports of possibility of Zn concentration regulation by certain strains are also being reported. Wetlands, metal mines, Sewage plants and tanneries add metals to waterbodies. Trophic transfer of Zn is shown to be co-related positively in some studies whereas others report bio dilution. First report of Zn bio-dilution in lacustrine environments comes from North Patagonia where Zn present in volcanic ash leached into a lake in 2011 and after 2 years the lake showed no signs of significant increase in Zn concentrations in fishes which means that water bodies recover on their own as long as contamination is within certain limits that nature can handle.

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