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Bioindicators in Heavy Metal Detection

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Abstract - *Heavy metals pose a threat to plant and human life, because of their toxicity, bioaccumulation, and non-biodegradability. Metal contaminants have two significant effects: pollution of the environment and health concerns. The use of bioindicators as observation devices to monitor natural pollution with hazardous metals has grown in popularity. To measure the build-up of heavy metals, bioindicators, such as flora and animals, are collected and evaluated. To screen dangerous metals from air, water, soil, and other sources, different living creatures from the five kingdoms – Monera, Protista, Fungi, Plantae, Animalia – are used. They should be able to concentrate the pollutant in their tissues to a level that is higher than the permissible limit for the surrounding environment. Here, we are surveying bioindicators and biological impacts of 11 heavy metals-Copper (Cu), Mercury (Hg), Chromium (Cr), Manganese (Mn), Cadmium (Cd), Lead (Pb), Zinc (Zn), Iron (Fe), Arsenic (As), Cobalt (Co) and Nickel (Ni).*

Key Words: Heavy metal detection, bio accumulation bioindicators, pollution, environment, heavy metal toxicity, harmful effects, bioremediation, biological impact.

1.INTRODUCTION

Living creatures such as plants, planktons, animals, and bacteria are used as bioindicators to monitor the health of the natural ecosystem in the environment [1]. The worldwide increase in environmental pollutants requires new and optimized methods of detection and control. Heavy metals are one form of hazardous industrial contaminant that can have long-term consequences for ecosystems and species.

Detecting environmental contamination with biological material as indicators is a low-cost, dependable, and straightforward alternative to traditional sampling approaches. Several organisms such as green algae, arthropods, lichens, and hydrophytes have been successfully used to detect heavy metals from industries.

Effective and reliable bioindicators of heavy metal pollution should react with the contaminant in a quantitative manner such that the measured strength of the biomarker response is proportionate to the amount of pollutant present. They should be easy to test and should accumulate the contaminant in their tissues to a much greater concentration than the surrounding environment. Lastly, they should be able to distinguish between excess synthetic compounds and natural ecological stresses and also measure potentially toxic substances [1].

The advantages associated with the use of bioindicators are that they are useful in quickly ascertaining biological impacts, both on the environment and on specific organisms, abundantly prevalent and easy to utilise as well as much cheaper alternative to specialized measuring systems [2].

This review classifies bioindicators based on the heavy metals they accumulate and detect. Each metal contains examples of bioindicator organisms belonging to different kingdoms and ecosystems from around the world. Many of them can detect multiple heavy metals. Finally, these bioindicators have been compared to ascertain their suitability under different conditions. Thus, this review covers a wide variety of bioindicators and mentions the bioindication processes taking place in these organisms under varied environmental conditions.

2.BIOINDICATORS OF COMMON HEAVY METAL POLLUTANTS

2.1 Bioindicators of Copper

Compounds containing copper (Cu) metal have been widely used in the agricultural fields and systems in the form of ingredients of fertilizers and fungicides [3] and has thus resulted in the accumulation of the metal in soils. This accumulation of copper results in the generation of various types of stresses posing on the environment which leads to display of specific injury symptoms or shifts in the community composition in various communities of organisms [4]. Research has found that certain organisms can be used as a bioindicator to detect the levels of copper in the environment in which they are found.

In a metal smelting plant in the Plateau state of Bukuru, study have found that mango plant can be efficiently used as a bioindicator for detecting copper metal (27μg/g) [5]. The study at Usmanu Danfodiyo University, Sokoto revealed that the concentration of copper metal (30.41 μg/g) was higher close to the road than away from the road edges in *Acacia nilotica* [6]. Arthropods such as spider and Rambur's forktail found in Legnica, Western Poland and Hawr Al Azim wetlands, respectively, confirmed that the smelters present in their areas indicated high air pollution and high copper (112.45 µg/g) and lead concentrations in the environment [7] respectively. In Cairo, Egypt, it was observed that the fungi *Aspergillus flavus*, in the presence of copper metal (32.1 µg/g) could decolourize the textile wastewater [8]. In the Point Lomo kelp forest, San Diego, studies have found that giant kelp by bioaccumulating copper (100 µg/g) and zinc from the water column can respond to heavy rainfall and storm events resulting to which the tissue concentration of these metals increases [9].

Table 1 denotes the commonly used bioindicators found in the different parts of the world for the detection of copper metal. From the bioaccumulation values for various bioindicators as seen in table 2, it can be concluded that among animals, arthropods are good bioindicators of copper while among plants giant kelp can be very useful to detect surrounding copper concentration in µg/g.

Table-1: Commonly used bioindicators for copper detection

Category	Bioindicator	Location of Study	References
Plants	Mango *(Mangifera indica)*	Plateau state	[5]
	Gum Arabic Tree *(Acacia nitolica)*	UDU, Sokota	[19]
Arthropoda	Spider *(Agelena labyrinthica)*	Legnica, western Poland	[20]
	Rambur's forktail *(Ischnura ramburii)*	Hawr Al Azim wetlands.	[21]
	Mediterranean green crab (*Carcinus aestuari*)	Narta Lagoon, Albania	[22]
Fungi	*Aspergillus flavus*	Cairo, Egypt	[8]
Algae	Giant Kelp *(Macrocystis pyrifera)*	Point Loma kelp forest	[23]
Parasite	Spiny headed worm *(Acanthocephalans)*	Antarctica	[24]
Fish	Nile tilapia (*Oreochromis niloticus*)	Nakivubo wetland, Uganda	[25]
	Huaiquil (*Micropogonias manni*)	Lake Budi, Chile	[26]

Table-2: Bioaccumulation levels of copper in different organisms

Species	Bioaccumulation (µg/g)	References
Mangifera indica	27	[5]
Acacia nitolica	30.41	[6]
Agalena labyrinthica	112.45	[7]
Ischnura ramburii	26	[7]
Aspergillus flavus	32.1	[8]
Macrocystis pyrifera	100	[9]
Acanthocephalans	50	[24]

2.2. Bioindicators of Mercury

Mercury (Hg) metal was found from the natural sources which include emissions from the geothermal and volcanic activity. It is also formed due to anthropogenic sources wherein the largest source is the combustion of coal and other fossil fuels including forest fires, waste disposal, metal, and cement production etc. [10]. MeHg poisoning have been observed in humans in various parts of the world [11]. Thus, mercury and its compounds can lead to harmful effects and present potential hazards to the environment even at very low concentrations.

Arthropod *Ligia italica*, found in the supralittoral zones of the Sicilian ecotones is observed to be a good bioindicator for detection of mercury pollution [12]. It was carried out in one of the most industrialized and affected region in Poland (Upper Silesia), which was a continuation of an investigation already going on the metal accumulation in the native and transplanted moss *Pleurozium schreberi* [13,14]. Waterbirds were useful as bioindicators of wetland heavy metal pollution, especially mercury since their presence in the environment influenced the survival and reproduction rate in them [15]. Selectivity of the heavy metal cations by algae *Cladophora sp.* was observed in various competitive adsorption studies. In the Acid Mine Drainage (AMD) waters, algae were found to be capable of being a good bioindicator of the mercury metal as well as it is suitable for its removal [16]. Trace metal concentration of mercury in *Patella caerulea* was investigated to provide information on the pollution of the Ionian Sea (Mediterranean Sea- Italy) [17]. According to various research studies, it is concluded that the carnivorous (piscivorous) fishes are the most common bioindicator for the detection of mercury metal accumulation in the environment. Earthworms can also be used as a viable alternative bioindicator for the detection of mercury due to its ability to accumulate heavy metals from the polluted salts and other media [18].

Table 3 denotes the commonly used bioindicators found in the different parts of the world for the detection of mercury metal.

Table-3: Commonly used bioindicators for mercury detection

Category	Bioindicator	Location of Study	References
Arthropoda	Rock Lice *(Ligia italica)*	Sicilian ecotone	[12]
Moss	Red stemmed feather moss (*Pleurozium schreberi)*	Poland (Upper Silesia)	[28]
Birds	Waterbirds	Wetlands	[29]
	Penguin (*Spheniscidae sp*)	Kerguelen Islands, southern Indian Ocean	[30]
Algae	Green algae (Cladophora sp)	Acid Mine Drainage (AMD) waters, South Africa	[31]
Mollusca	Mediterranean Limpet (Patella caerulea)	Mediterranean area	[17]
Annelids	Manure Worm *(Eisenia foetida)*	Cachoeira do piriá, Brazil	[18]
Fish	Red-eyed piranha *(Serrasalmus rhombeus)*	Tapajós River	[18]
	European perch *(Perca fluviatilis)* Common roach *(Rutilus rutilus)*	Pluszne Lake, Poland	[32]
Reptiles	Watersnakes *(Nerodia taxispilota)*	Savannah River,United States	[33]

2.3 Bioindicators of Cadmium

Cadmium (Cd) is a heavy metal that is becoming increasingly prevalent in our environment as a result of industrial production and usage [34]. The findings of research on marine bivalve (*Ruditapes decussates*) specimens show that metallothionein (MT) synthesis responds to modest changes in metal concentrations [35].

Another study carried out in *Cerastoderma glaucum* showed significant fluctuations in the MTLP concentrations [36]. Among 12 common species of hydrophytes chosen, roots and shoots of *Mentha aquatica* was found to be the most promising single indicator of the pollution of heavy metals like Ni, Cd and Cr.

This study carried out in water-scarce and budget-limited countries like Lebanon has various ethnobotanical uses [37]. *Medicago sativa* cultivated in various heavy metal concentrations displayed reductions in chlorophyll content, increased liquid peroxidation, increased glutathione reductase activity. The plant development slowed dramatically as the metal concentrations grew [38].

Due to their high cation exchange capacity and long deciduous periods, mosses are useful plants for scanning heavy metal deposition according to a study conducted in Serbia [39].

Barley seeds grown in varying levels of cadmium displayed a lower root growth at higher levels of metal concentration [40]. The sea urchin embryo (*Paracentrotus lividus*) is a major invertebrate that has been used as a bioindicator of heavy metal contamination and a model organism in developmental biology through altered levels of HSP70. Another species of sea urchin, (*Anthocidaris crassispina*), is also an important model to study cadmium induced stress. Reduced sperm motility and fertilization and increased egg size can be observed at high cadmium levels [41].

Flying foxes (*Pteropus poliocephalus*) can also serve as potential bioindicators for environmental metal exposure through tissue, urine, and fur samples. Specimen samples collected from the Sydney basin, Australia, were used to determine cadmium, arsenic, and various other trace metals [42].

Table 4 showcases the findings from research aimed at examining the correlation between the bioaccumulation of cadmium in organisms and the levels of cadmium in their surroundings.

Table-4: Commonly used bioindicators for cadmium detection

Category	Bioindicators	Location of Study	References
Mollusc	Grooved carpet shell *(Ruditapes decussatus)*	Tunisia	[35]

	Lagoon cockle *(Cerastoderma glaucum)*	Gulf of Gabès, Tunisia	[50, 36]
Fish	Acanthocephalans	Baía and Paraná rivers, Brazil	[51]
Lamiaceae	Water mint *(Mentha aquatica)*	Lebanon	[37]
Fabaceae	Alfalfa *(Medicago sativa)*	Not Mentioned	[38]
Bryophyta	*Brachythecium sp.*, Hypnum moss (*Hypnum cupressiforme),*	Obrenovac (Serbia)	[39]
Echinodermata	Common Sea Urchin *(Paracentrotus lividus)*	Mediterranean Sea and eastern Atlantic Ocean.	[41]
	Purple Sea Urchin *(Anthocidaris crassispina)*	Tropical and subtropical coastal waters	[41]
Arthropoda	Antlion *(Myrmeleontidae)*	Near Local Steel Factories	[52]
Chordata	Grey headed flying fox *(Pteropus poliocephalus)* and Black headed flying fox (*Pteropus alecto)*	Sydney basin, Australia	[42]
Magnoliophyta	Seedlings of Barley *(Hordeum vulgare*)	In Laboratory Experiment	[40]
Nematoda	Parasitic Roundworm Larvae *(Hysterothylacium sp.*)	Sea of Oman	[53]

2.4 Bioindicators of Arsenic

Arsenic (As) is recognised to be toxic to both plants and mammals due to its affinity for protein, lipids, and other biological components. Specimens of testate lobose amoeba collected from 59 lakes in Canada displayed various assemblages. The specific spatial pattern obtained suggests the presence of industrially derived arsenic [43].

Because they are at low trophic levels and act as the trophic web's entrance doorway, molluscs have been regularly used to predict environmental risk. The freshwater snail (*Pomacea canaliculate*) selectively accumulates metal contaminants at high levels in the kidney, and symbiotic corpuscles. In arsenic-exposed apple snails, preferential accumulations in the digestive gland were 9 and 276 times larger than in nonexposed snails [44]. Ant colonies belonging to different microhabitats may have diverse responses to environmental effects because they are exposed to different habitat conditions and resource availability [45].

Aquatic bryophytes have also been used to study the heavy metal contamination of certain areas. The amount of arsenic in the biotope is reflected in the amount of arsenic in the investigated aquatic bryophytes. Water analyses are less reliable than these plants in determining the presence of arsenic [46]. Some plants can act as bioindicators with respect to their absorption spectrum. A study on Vallisneria gigantean and *Azolla filiculoides* showed an increase in absorption in the 400 to 500 nm region. There was an additional increase in the 530 nm region for *Azolla filiculoides*. As a protective reaction to arsenic activity, this shows an increase in flavonoid production [47]. The basis of another study was the nodule bacteria of the genus *Trifolium L.* genus as bioindicators. Lower clover nodule bacteria colonies were formed in soil that had a higher metal concentration [48].

Blood and excrement samples from birds are used to detect internal metal concentrations. Clear relationships between As, Cd, and Pb were observed in liver and blood. This proved that blood can be used as a beneficial tool to determine heavy metal concentrations [49]. The findings of investigations that sought to establish the connection between the bioaccumulation of arsenic in various organisms and the arsenic concentrations in their environment are demonstrated in Table 5.

Table-5: Commonly used bioindicators for arsenic detection

Category	Bioindicators	Location of Study	References
Amoeba	Testate lobose amoebae *(Lacustrine arcellinina)*	Yellowknife, Northwest Territories, Canada	[43]
Arthropoda	Arboreal and epigaeic ants	Nova Lima, Minas Gerais, Brasil	[45]
Bryophyta	Aquatic bryophyte-like Pale Liverwort (*Chiloscyphus pallescens*)	Sudetes Mts., Poland; and east Sudetic Rychlebske Mts. and Jesenik Mts. (Czech Republic).	[46]

	Brachythecium sp., Hypnum moss (*Hypnum cupressiforme*), Silvergreen Bryum moss (*Bryum argenteum*)	Obrenovac (Serbia)	[54]
Pteridophyta	Eelgrass (Vallisneria gigantean) and Water Fern (Azolla filiculoides)	Latin America	[47]
Mollusc	Channeled apple snail or golden apple snail *(Pomacea canaliculata)*	Laboratory	[44]
Birds	Migratory birds like European pied flycatcher (*Ficedula hypoleuca*)	Saharan Africa or UK	[49]
Fabaceae	Nodule bacteria of red clover (*Trifolium praténse)*, Alsike clover (*Trifolium hibridum)*,	North Caucasus Research Institute of the Vladikavkaz Scientific Center of the Russian Academy of Sciences.	[48]

2.5 Bioindicators of Zinc

Metal compounds are researched in green algae (Ulva rigida), mussels (*Mytilus galloprovincialis*), and molluscs (*Tapes philippinarum*), three species found in marine biological systems. The elements under consideration are Hg, Cu, Pb, As, Zn, Ni, and Cr. Zinc exhibits a standard deviation of 6% [55]. Concentrations of persistent organochlorines (OCs) such as polychlorinated biphenyl (PCBs), dichloro diphenyl trichloroethane (DDTs), chlordanes (CHLs), and HCHs found in the liver of bluefin fish (*Thunnus thynnus*) are gathered. The amounts of PCBs, DDT, and CHL in bluefin fish increased considerably with body length (30–190 cm).

There was a significance of dietary intake of PCBs, DDTs, and CHLs in comparison to gill entry. The straight relapse condition obtained from the plot of fixations and body length was used to determine the Body-Length Standardized Qualities (BLNV) of PCBs, DDTs, and CHLs fixations in bluefin fish.

The BLNV demonstrated the current condition of PCB, DDT, and CHL contamination in water. These findings suggest that bluefin tuna is a suitable bioindicator for assessing OC pollution in the wild ocean biological system [56].

ICP-MS (Inductively Coupled Plasma-Mass Spectrometry) was used to analyse heavy metal fixations in ocean water and accumulation in the tissues of *Haliclona tenuiramosa*. Sponges living near the shore amassed more concentrations of heavy metals ranging from 2 to multiple times higher fixation than that observed further away from the shore.

The fixation levels in water and bioaccumulation in tissues was observed in certain fish. The current findings suggested that a more complete examination of the concentration of heavy metals in *Haliclona tenuiramosa* from the surroundings is required to aid in a better resolution of the problem [57].

Algae, bivalves, Cnidaria, Nematoda, amphipoda, and fish are some of the bioindicators used to detect zinc (Zn) as denoted in table 6.

Table-6: Commonly used bioindicators for zinc detection

Category	Environment	Bioindicator	References
Algae	Tropical waters	Green Algae, Sea lettuce *(Ulva lactuca)*, Brown algae (*Lobophora variegate)*	[61,62]
Bivalves	Coast of Arabian Gulf of Mauritania	Venus verrucos, Blue mussel *(Mytilus edulis), Crassostrea gigas, Crassostrea virginica, Crassostrea corteziensis,*	[55]
Cnidaria	Water Column Sessile Estuarine Sediments	Jelly fish *(Aurelia aurita)*, Snakeslocks, Anemone *(Anemona viridis)*, Starlet sea anemone *(Nematostella vectensis).*	[63]
Nematoda	Sea Water	Turbot *(Scophthalmus maximus)*, Gilt-head (sea) bream (*Sparus aurata), Trachus trachus, American* alligator (*Alligator mississippiensis).*	[61]

Amphipoda	Mediterranean coast, marine and estuarine sediments	*Corophium volutator, Echinogammarus pirloti, Gammarus salinus, Artemia salina, Ostracoda cypris sp., Cyprideis torosa, Leptocythere psammophila,*	[56]
Fish	Mediterranean	Nile tilapia *(Oreochromis niloticus),* Red Mullet *(Mullus barbatus),* Brown Comber *(Serranus hepatus),*	[57]

2.6 Bioindicators for Lead

Sunflowers, lichens, trees, birds, honeybees, aquatic animals, insects, and annelids are some of the bioindicators utilised in lead (Pb) detection as mentioned in table 7. Insects can be used as natural bioindicators of contamination. One of the most adaptable and effective bioindicators is honeybees. Deformations in hatchlings from a few genera in the Chironomidae family (e.g., *Procladius*, *Chironomus*, and *Cryptochironomus*) have been seen in numerous studies, and the results show that the anomalies are strongly linked to dirty silt. Honeybees, on the whole, have a better lattice for detecting metal contamination than honey. The higher amounts of each of the three metals in honeybees in rural areas may indicate that these metals are diffused in the air and do not seep into or store on the natural parts visited by honeybees, implying that they are not ingested. Thus, it was concluded that honeybees can be used to detect metal pollution. Live honeybees are better than dead honeybees at detecting [58]. Gerridae are used to find varied iron and manganese concentrations, however it appears that it is less appropriate for nickel and lead collection [59]. Wasps are used for lead biomonitoring since their bulk larval excrement can accumulate to many times the size of the adult body.

Metal accumulation in plants can also be influenced by soil particles. Ficus leaves have the potential to screen for heavy metal contamination in urban areas. Lead fixations in Ficus leaves remained fundamentally higher across the polluted areas. Vehicles are the principal source of lead pollution in plants, as seen by the positive link between lead fixation and thickness [60].

Chart 1 denotes the bioaccumulation levels of Lead and Zinc in different organisms measured in µg/g. The graph shows a comparative analysis using the same bioindicator for both the metals Lead and Zinc. It is inferred that the bioaccumulation has been the greater for Zinc in all the 5 cases It is seen that the maximum bioaccumulation happened in *Corallina officinalis*/tafsout with a highest in Zinc. A minimum has occurred in *Ulva lactuca*/Ouled saleh for lead.

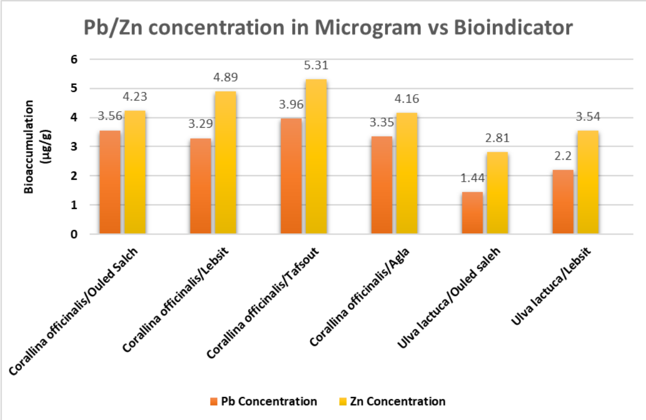

Chart-1: Comparison of bioaccumulation levels in Lead and Zinc in various species. [68]

Table-7: Commonly used bioindicators for lead detection

Category	Environment	Bioindicator	References
Flowers	Land terrain	Sunflower *(Helianthus)*	[64]
Lichens	Fog Belts	Script lichen *(Graphis scripta)*	[65]
Trees	Landy terrain	Sacred fig (*Ficus religiosa*)	[60]
Birds	Urban Terrain	House sparrows (*Passer domesticus*)	[66]
Honeybees	Mediterranean area	Western honeybee *(Apis mellifera L),* Italian bee *(Apis mellifera ligustica spinola),*	[58]
Aquatic Animals	Marine	Starfish *(Asteroidea)*	[67]
Insects	Forest terrain	Warps *(Polistes)*	[59]
Annelida	Sediments, Coasts	Arenicola Marina *(Hediste diversicolor)* Hediste diversicolor	[61]

2.7 Bioindicators of Chromium

Chromium (Cr) is the seventh most prevalent element on the planet. Chromium is a toxic heavy metal which usually occurs as either of two ionic forms - Cr (VI) and

Cr (III). Chromium interferes with several metabolic processes, modifies the activities of antioxidants and enzymes like ribonuclease and causes oxidative damage to biomolecules. It is also toxic to plants and results in reduced growth, foliar chlorosis, stunting, and plant mortality [69,70]. Exposure to Cr (VI) has been related to nasal mucosa damage, allergic contact dermatitis, renal, gastrointestinal, and cardiovascular effects, haematological effects, and liver necrosis. The Cr (III) valence states are also reactive and soluble, causing damage to DNA, proteins, and lipids [71].

The usual practice of bioindication is to test the collected samples of biomass for chromium ions. In an early statistical evaluation, a dozen common hydrophytes were compared from two different locations in the Bekaa valley, Lebanon. The concept of bioconcentration factor (BCF) served as a numerical bioindicator. After a period of 21 days, 9 out of 12 plants showed a chromium accumulation suitable for use as bioindicators [37]. A recent experiment was carried out by Perillo et al. [72] to determine the amounts of chromium and other heavy metals in the hair of Holstein dairy cows. The main advantage of this method is that it is bloodless and simple to obtain hair samples and analyse them. All six examined herds showed a similar concentration of chromium except one which had almost double. This revealed that excess fertilizers were being used in the province of Ragusa, Italy which may have been subsequently reduced [72].

The hydrophytes in table 8 were also found to be suitable bioindicators for chromium. Leaf or stem samples were extracted and tested for chromium ions. These hydrophytes exhibited all required properties of bioindicators and some also displayed linear relationships between the for the presence of chromium ions with high positive correlation coefficients between chromium accumulation and the amount present in the soils and environment.

Table-8: Commonly used bioindicators for Chromium detection

Category	Bioindicator	Environment	Reference
Cyano-bacteria	*Oscillatoria tenuis*	Tannery effluent	[75]
	Phormedium bohneri		[76]
Plant	Common water hyacinth (*Eichhornia crassipes*)	Coal mine effluent	[75]
	Water Butterfly Wing *(Salvinia natans)*	Electroplating effluent	[77]
	Eastern Mosquito Fern (*Azolla caroliniana*)	Fly ash effluent	[78]

2.8 Bioindicators of Manganese

Manganese (Mn) is an essential micromineral for both plants and animals. It has various functions such as being an important part of the enzymatic systems while also being involved in the synthesis of vitamin B1 and insulin. It is also a critical electron transporter in photosystem II. Exposure to Mn (II), Mn (III), or Mn (IV) ions has been shown in animals and people to have negative neurological consequences. Manganese poisoning can cause manganism, a long-term neurological condition characterised by tremors, difficulty walking, and facial muscle spasms. It has also been associated with Parkinson's disease and other cognitive disorders [73].

In a study which observed the seasonal variations of manganese in the environment, Catsiki et al. [56] made use of *Mytilus galloprovincialis*, as an estuarine bioindicator near the Thermaikos gulf in Greece. Seasonal variations in Manganese were lowest during the summer season and highest at the start of spring. It was concluded that mussels bioaccumulate less during warm periods than during the winter based on their reproductive cycles [56]. In another study by Demirezen et al. [74] five aquatic hydrophytic species *Phragmites australis, Ranunculus sphaerosphermus, Typha angustifolia, Potamogeton pectinatus*, and *Groenlandia densa* were found to be suitable indicators of manganese contamination after being tested for relations between their indicator value and actual degree of contamination.

Table 9 illustrates the results of studies to determine the relation between the bioaccumulation of manganese in a variety of organisms and the actual concentrations of manganese in their environment. The dual properties of high bioaccumulation along with a linear relation between metal concentration in the plant and the soil as illustrated in chart 2. These make *Typha angustifolia* the most suitable bioindicator while *Groenlandia densa* can also be used due to its linear bioaccumulation.

Table-9: Commonly used bioindicators for Manganese detection

Category	Bioindicator	Environment	Reference
Algae	*Antithamnion cruciatum*	Black Sea coast of Samsun in Turkey	[79]
	Corallina panizzoi		
Insects	Waterstriders (*Gerris argentatus*)	Iron and steel factory	[52]
	Dragonfly larvae (*Odonata*)		
Plants	Paper flower (*Bougainvillea glabra*)	Industrial Zone	[80]
		Residential Zone	

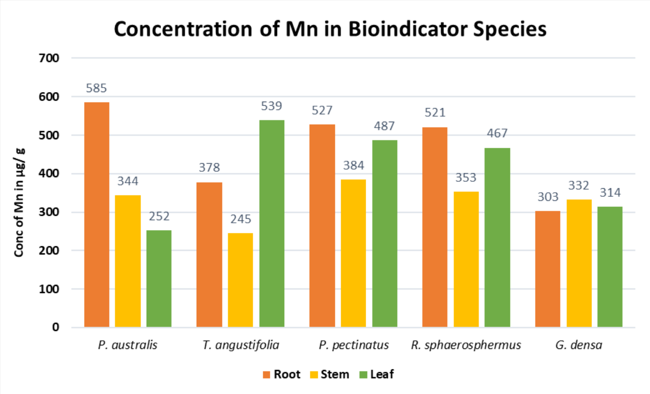

Chart-2: Comparison of bioaccumulation levels in Manganese in various species. [74]

2.9 Bioindicators of Iron

Iron (Fe) is a heavy metal which is necessary for the proper growth of those organisms. In certain regions due to the presence of factories, especially steel factories and coal mines the iron content in the soil and surroundings increase above safety levels which becomes detrimental for organisms living in such environments. The iron content in the environment needs to be detected and certain organisms which live in such environment can be used as bioindicators. Experiments conducted in different places across the globe has led to discovery of many such bioindicators.

In a Brazil pelletizing factory two plants Surinam cherry (*Eugenia uniflora*) and *Clusia hilariana* showed necrosis, chlorosis, purple spots on leaves [81]. In the Thermaikos Gulf, Greece, neptune grass (*Cymodocea nodosa*) iron content in leaves was analysed by Malea and Haritonidis [82]. In Africa, African catfish (*Clarias gariepinus*) show suppressed growth, high concentration of malondialdehyde in liver [83]. In Australia, mussels show reduction in responsiveness to ambient iron concentration changes [84,85]. In Malaysia, clams show hemochromatosis as an indication of iron accumulation [86]. In RSA and CI channel, Argentina Chinese Hat Snail (*Bostrycapulus odites*) show thicker shells with microstructure malformations due to excess iron accumulation [87]. In Iran, reaction of honeybees to changes in environmental iron content is depicted in the iron levels in their bodies [88].

Table 10 shows the bioaccumulation values in different organisms under study in a tabular form. From this table we can infer which bioindicator is most effective. Since *Clarius gariepinus* shows a maximum of 6000 µg/g it is the most effective bioindicator out of the lot. The effectiveness of the bioindicator is proportional to its bioaccumulation capability. Finally, the bioaccumulation levels also give a brief idea about the iron content in the environment cross different parts of the world where these potential bioindicators are found as shown in table 11.

Table-10: Commonly used bioindicators for Iron detection

Category	Bioindicator	Environment	References
Plants	Surinam Cherry (*Eugenia uniflora)*	Brazil- pelletizing factory	[81]
	Clusia hilariana	Brazil- pelletizing factory	
	Neptune Grass (*Cymodocea nodosa)*	Seawater and in sediment from the Thermaikos Gulf (Greece)	[82]
Fishes	*African catfish (Clarias gariepinus)*	Aquatic ecosystems of Africa and Middle East.	[83]
Bivalves (Oysters, molluscs)	Flood plain Mussel (*Velesunio ambiguous)*	River Murray, South Australia	[85]
	Sydney Rock Oyster (*Saccostrea glomerata)*	Sea Ports of New South-Wales (NSW), Australia	[84]
	Mangrove clam (*Polymesoda expansa)*	Aquatic habitat of Kuala Kemaman, Terengganu, Malaysia	[86]
	Chinese hat snail (*Bostrycapulus odites)*	Ría San Antonio channel (RSA) and Canal del Indio channel (CI), Argentina	[87]
Insects	Honeybee *(Apis mellifera)*	Markazi Province, Iran: varying degrees of anthropogenic impact	[88]

Table-11: Bioaccumulation levels of iron in different organisms

Species	Bioaccumulation (µg/g)	References
Eugenia uniflora	895	[81]
Clusia hilariana	596	
Cymodocea nodosa	2466	[82]
Clarias gariepinus	6000	[83]
Velesunio ambiguous	5574	[85]

Saccostrea glomerata	790	[84]
Polymesoda expansa	7	[86]
Anadara granosa	3	
Apis mellifera	1695	[88]

2.10 Bioindicators of Cobalt and Nickel

Cobalt (Co) and nickel (Ni) are trace elements that are required in trace amounts by plants and animals to grow normally. The MPA (Maximal Permissible Addition) of cobalt in soil is 24 µg/g while that of nickel is 2.6 µg/g [89]. Mosses (*Bryum argenteum, Bryum capillare*) are employed in Serbia as cobalt bioindicators using atomic absorbance spectrophotometer principle [39]. In Nigeria earthworms (*Hyperiodrilus africanus*) are used as bioindicators as they show changes in alimentary tract [90]. Plants like Gum Arabic tree (*Acacia nilotica*) are also used in Nigeria for bioindication where tree barks are analysed by AAS [19].

Freshwater silver catfish (*Chryshchythys nigrogitatus*) are used in Nigeria as cobalt accumulation in liver and gills can be detected by AAS [91]. In Santos Bay, Brazil, Madamango sea catfish (*Cathorops spixii*) show altered growth rate, reproductive phases, cellular mutations and even death due to cobalt accumulation [92]. Hydrophytes are used as bioindicators of Nickel in Mediterranean region by inductively coupled plasma mass spectrometry analysis of roots and shoots [37]. In Baghdad, molluscs (*Bellamya bengalensis, Physella acuta*) are used as bioindicators as Ni accumulation affects growth, feeding, reproduction, physiological activity and maturity [93]. In estuaries of Australia, microalgae (*Catenella nipae*) epiphytes grow on aerial roots of mangroves as bioindicator of Ni [94]. In Egypt, *Bougainvillea glabra* is used since Ni accumulation causes increase in flavonoid and phenolic content analysed by AAS [80]. In Serbia, Pygmy iris (*Iris pumila*) was found to have a considerable block effect on nickel concentration in its leaves [95]. Table 12 gives an insight on the potential indicators of nickel and cobalt found in different regions of the world.

Table-12: Commonly used bioindicators for Cobalt and Nickel detection

Category	Bioindicator	Environment	References
Cobalt			
Mosses	Silvergreen byrum moss (*Bryum argenteum*), Bryum moss (*Bryum capillare*)	County of Obrenovac (Serbia)	[39]
Earthworms	Earthorm (*Hyperiodrilus africanus*)	Lafarge, WAPCO Cement Factory, Ewekoro, Nigeria	[90]
Plants	Gum Arabic Tree(*Acacia nilotica*)	Usmanu Danfodiyo University, Sokoto - Nigeria	[19]
Fish	Fresh-water silver catfish (*Chryshchythys nigrogitatus*)	Cross River, south-eastern part of Nigeria	[91]
	Madamango sea catfish (*Cathorops spixii*)	Santos Bay, Brazil	[92]
Nickel			
Hydrophytes	*Nasturtium officinale, Cardamine uliginosa, Mentha longifolia, M. aquatica, M. sylvestris*	Aquatic ecosystem in Mediterranean (Lebanon)	[37]
Molluscs	Freshwater snail (*Bellamya bengalensis*), Bladder snail (*Physella acuta*)	Tigris river, Baghdad	[93]
Algae	Nipae palm (*Catenella nipae*)	Estuaries in the vicinity of Sydney, Australia.	[94]
Plants	*Paper flower (Bougainvillea glabra)*	Sadat City, Western Nile Delta, Egypt	[80]
	Pygmy iris (*Iris pumila*)	Belgrade, Serbia	[95]

3. COMPARATIVE STUDY OF BIOINDICATOR ORGANISMS

3.1 Plants

Organisms like micro and macroalgae, lichens, mosses, tree bark, fungi and leaves of higher plants have shown to detect the accumulation, deposition of metal and distribution of the metal pollution in water, soil and air. This accumulation and distribution of metal pollution depends upon the levels of the metals in the soil, water and air, the element species and the bioavailability, pH, vegetation period, cation exchange capacity and multiple other factors.

Some algae like *Macrocystis pyrifera*, *Cladophora sp., Ulva lactuca, Lobophora variegate, Antithamnion cruciatum, Catenella nipae* are used as standard bioindicators which represent the primary producers. For instance, the presence of algae *Fucus vesiculosus* is observed to show heavy metal pollution in marine environment whereas the presence of *Klebsormidium* dominated algal mats are found to be good indicators of high concentration of iron in water.

Mosses such as *Pleurozium schreberi, Bryum argenteum*, *Bryum capillare* are useful plants for scanning heavy metal deposition. They can accumulate large amounts of heavy metals without any significant damage due to their deciduous periods and their high cation exchange capacity.

Higher plants have also been used as bioindicators in areas with significant amount of pollution in the detection of heavy metals like Cd, Zn, Pb, As, Cu, Hg etc. In higher plants, distribution of heavy metals is found to be unequal with the maximum found to be in the tree bark. After the tree bark, heavy metals are accumulated in the roots, then leaves and finally in the fruits.

Hydrophytes are used as bioindicators of nickel in Mediterranean region as nickel gets accumulated in roots and shoots. They are also very useful for monitoring environmental pollution at the interface between aquatic and terrestrial ecosystems which is where heavy metals such as chromium from industries usually ends up.

3.2 Terrestrial Animals

Different categories of animals have been used as bioindicators based on certain characteristics they show in response to accumulation of heavy metals in their systems. Most commonly found bioindicators include insects, earthworms, birds and even higher animals.

Insects like spiders, bees, ants, wraps and flies are used for bioindication of different heavy metals like Cu, Hg, Cd, As, Zn, Pb, Mn and Fe. Heavy metal accumulation in their systems change their responsiveness and growth and the bioaccumulation can be analyzed using analytical methods.

Different types of worms like roundworms and earthworms are also used as bioindicators as they show changes in their alimentary tract and responsiveness to environmental change on accumulation of metals like Hg, Cd, Zn, and Co.

Birds like sparrows, waterbirds, flycatchers are also used as bioindicators as they show detrimental effects in growth and reproduction on accumulation of Hg, As and Pb.

Chordates like flying fox (*Pteropus poliocephalus* and *Pteropus alecto*) are also used for bioindication of cadmium. Blood proved to be an indispensable test sample for determination of heavy metal concentration. Cows were also used for bioindication of Cr and some other heavy metals based on assessment of hair samples.

3.3 Aquatic animals

In the aquatic environment different varieties fishes like piranha, mullets, combers, huaiquils, perchs, roaches, catfishes and acanthocephalans are used for bioindication as metals like Hg, Cd, Zn, Fe and Co get accumulated in liver and gills and can be analyzed to obtain levels of those metals.

Other aquatic animals like water snakes, crabs, snails, starfishes, limpets, shellfishes, sea urchins, bivalves, amphipods, oysters, mussels, jelly fish, sea anemones are also used for bioindication of Hg, Cd, As, Zn, Fe and Ni. They show changes in their shells, larval development, growth and other physiological factors which can be used for indication.

Heavy metal deposition in aquatic creatures around a gold mining location in Thailand is shown in one case study. Three different fish species *Rasbora torneiri*, *Brachydanio albolineata* and *Systomus rubripinnis* accumulate metals like Fe, Zn, Cr, Mn, Ni, As etc. This bioaccumulation level in turn provides for the bioindication in these fishes [96].

Another case study in the Gulfs of Oman and Persian demonstrated the use of marine species such as ghost shrimps, barnacles, polychaetes, and bivalves for the bioindication of metals such as Pb and Cd. The bioaccumulation levels in these animals made them applicable for further monitoring programs that could help detect the level of these heavy metal pollutions [97].

3.4 Microorganisms

Amoeba is used as a bioindicator for Arsenic as different concentrations leads to formation of assemblages which can be studied to determine levels of *arsenic* in the environment.

Cyanobacteria like *Oscillatoria tenuis* and *Phormedium bohneri* are used as bioindicators of chromium and the bioaccumulation levels were determined. Thus, we observe each category has a wide variety of organisms which can be used as bioindicators in their natural habitat.

Responses of certain microorganisms to heavy metal pollution make them ecologically significant. Certain bacteria show resistance to heavy metals and this property is due to resistant genes in their plasmids. Thus, they can serve as useful bioindicators [98].

4.DISCUSSION

All of the research in the last few decades has been focused on finding bioindicators such as microbes, plants, and animals that collect harmful metals. Bioindicators are useful for defining natural environment characteristics as well as detecting and assessing human impacts. Because bioindicators are particularly sensitive to contaminants in their environment, if pollution is present, the organism may change its morphology, physiology, or behavior, or even perish. Heavy metal bioindicators include a variety of microorganisms, plant, and animal species. In areas where mosses are absent, higher plants can be used as bioindicators to detect air pollution.

The use of flora for heavy metal contamination bioindication isn't always done. Some higher plant responses to heavy metals as bioindicators of soil contamination have that potential. Insects and animals from the Arthropoda class, such as spiders, honey bees, ants, worms, and flies, are used to detect Cu, Hg, Cd, As, An, Pb, Mn, and Fe. On accumulation of metals such as Hg, Cd, Zn, and Co, many nematodes display alterations in their alimentary tract and reactivity to environmental change.

Aves have also been employed as bioindicators because they have negative impacts on Hg, As, and Pb accumulation during growth and reproduction. Chordates such as the flying fox are also utilized for cadmium bioindication. Hg, Cd, Zn, Fe, and Co are detected using organisms from the Pisces class because they accumulate in the liver and gills and may be examined to determine amounts of those metals. Microbes also have physiological and structural reactions. Lichens serve as good pollution bioindicators.

Heavy metals are thus discharged into the air, surface water, and soil, and consequently into groundwater and crops; once in the environment, they do not dissipate, but rather accumulate in soils, sediments, and biomass. Metal content in bioindicators is influenced not only by metal concentrations in air, water, soil, and sediment, but also by environmental factors and biological factors in the organisms. As a result, the impact of these factors in this complex ecosystem must be monitored. All of the research in the last few decades has been focused on finding bioindicators such as microbes, plants, and animals that collect harmful metals.

5. CONCLUSION

All heavy metals even though naturally present in the environment can cause toxicity to organisms if their concentration rise above safety levels. These metals get accumulated in the systems of these organisms due to the lack of metabolism mechanisms. After certain concentrations they show notable changes in their physiological characteristics. On proper monitoring, these characteristics can be analysed to determine the concentration of these metals in the surrounding environment.

As a result, species that live natively in those ecosystems can be employed as bioindicators for heavy metals like Cu, Cr, Fe, As, Zn, Hg, Cd, Pb, Mn, Co, and Ni. We can take further actions to lower heavy metal concentrations in certain places based on the levels determined from these creatures. This in turn prevents irreversible damage to the ecosystem and humans which would have occurred due to excessive contamination by heavy metals. This also leads to economic growth and social development in those regions by improving the environment.

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