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Bioaccumulation patterns of heavy metals in pelagic and demersal fishes of the gulf of kachchh

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Abstract
This study assessed the bioaccumulation of four heavy metals-Lead (Pb), Cadmium (Cd), Nickel (Ni), and Chromium (Cr) in sixteen commercially important marine fish species with varied feeding habits collected from four coastal locations (Kandla, Mundra, Mandvi, and Pingleshwar) in the Gulf of Kachchh, India. Muscle, gill, liver, and intestine tissues were analyzed for heavy metal concentrations. Results revealed that Pb and Cd were detected in all fish tissues across all locations, with concentrations exceeding permissible limits for human consumption (0.5 mg/kg). The highest concentrations were observed in samples from Kandla, followed by Mundra, Pingleshwar, and Mandvi coasts. Concentrations of Pb ranged from 3.6±2.47 to 60.3±0.26 mg/kg, while Cd ranged from 0.409±1.15 to 4.950±1.28 mg/kg. Ni and Cr were below detectable levels in most samples except those from Mandvi coast. The rate of bioaccumulation was higher in muscle and gills compared to liver and intestine.

Keywords: Bioaccumulation, heavy metals, gulf of kachchh, detection level, lead, cadmium, fish tissues, human health risk

Introduction
Marine organisms have been extensively used in marine pollution monitoring programs as the pollutants like heavy metals and the non-degradable chemicals tend to accumulate in the tissues [1]. The heavy metals in water and sediments are absorbed into the organs at different levels [2]. The heavy metal concentration in the fishes and shell fishes have been monitored in the marine environment due to their ability to accumulate in the organs through the body surface, gills and food [3]. The heavy metals in the tissues because of their high propensity to infiltrate and biomagnified along the food chain [4] particularly Cu, Zn, Mn, Fe, Co, Cr, Ni, Pb, Cd and Hg, leading to severe risk for animal and human health. The risk of bioaccumulation to humans have been reported through consumption of fishery products which have higher levels of these heavy metals in the muscle [5] causing renal, cardiovascular, nervous, bone diseases and immunity issues [6, 7]. Along the Gulf of Kachchh coast in Gujarat, many large, medium, and small industries are located which are the sources of various pollutants particularly heavy metals. In the Gulf of Kachchh, the spatial distribution of heavy metals and their potential ecological risks have been pointed out.[8] The mean concentrations were in the following order: Cr>Cu>Ni>Pb>Cd>Hg and hence it is imperative to investigate the bioaccumulation rate of these metals in fishes caught from this region. In this context a study was undertaken on the bioaccumulation of four heavy metals such as Lead, Cadmium, Chromium and Nickel in the organs of the commercially important fishes with varied feeding habits to understand the potential health risk to the regular consumers.

Materials and Methods
Study Area and Sampling Procedure
The study was conducted on 10 pelagic and 6 demersal fish samples collected from the catch brought by the fishermen at Kandla, Mundra, Mandvi, and Pingleshwar coasts located in the Gulf of Kachchh (GoK) (Figure 1). The fishes were immediately preserved in iceboxes for transportation to the laboratory and stored at -20°C. The fishes were identified and classified based on the keys from published literature and FishBase [9].

The fishes were thawed and washed with distilled water and quickly measured the total length and weight before dissecting with sterile instruments. The liver, stomach, intestine, gills, and muscle tissue were separated under sterile conditions and followed the standard procedures for tissue digestion and further analysis [10]. The samples were analyzed by Atomic Absorption Spectrophotometer (Perkin Elmer Analyst 400) using air-Acetylene flame with a digital readout system, and results were expressed as mg/kg.

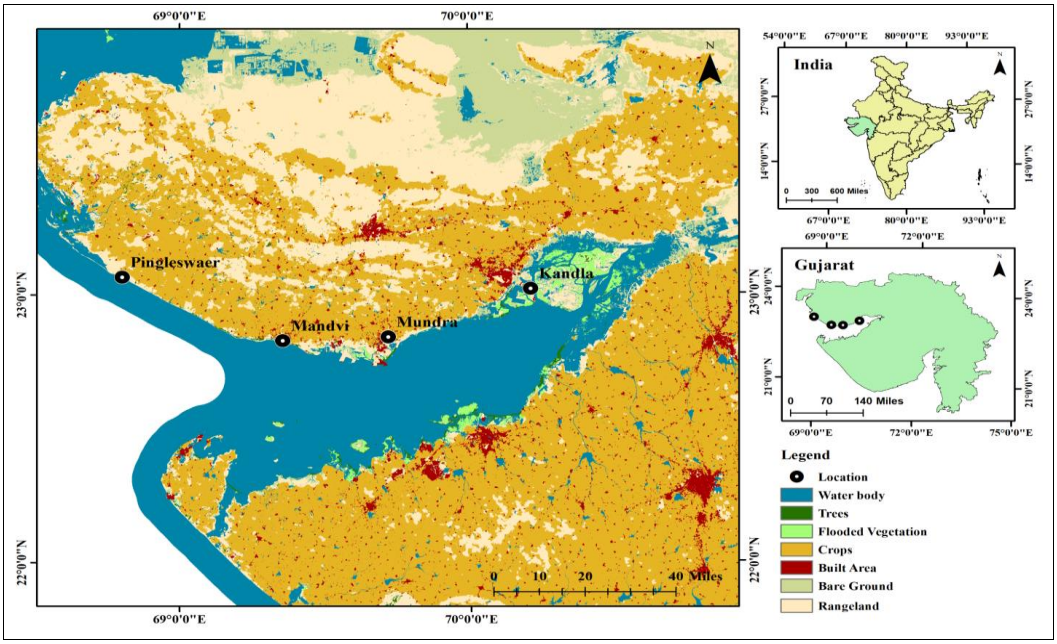

Fig 1: Sample collection locations in the Gulf of Kachchh

Statistical analysis

The data were analysed using Microsoft Excel (2010) and R software (v2024.4.2.764). To explore the rate of accumulation of the selected heavy metals on the different organs of fishes falling under different trophic levels the tissues were treated separately. The Principal Component Analysis (PCA) was conducted using R software to reveal the interrelationships between the concentration of heavy metals in the different tissues of the fishes.

Results and Discussion

Sixteen fish species with varied feeding habits were selected from catches brought by fishermen. The heavy metals lead (Pb) and cadmium (Cd) were detected in the muscle, gill, liver, and intestine of the fishes (Table 1). The metals nickel (Ni) and chromium (Cr) were below detectable concentrations in the fishes except those collected from the Mandvi coast. There were noticeable differences in the concentration of Pb, Cd, and Ni in fish tissues. The accumulation of heavy metals was comparatively lower in fishes collected from the Mandvi coast, while higher values of Pb and Cd were observed in the tissues of fishes obtained from Kandla.

The results showed varied rates of accumulation of Pb and Cd in different organs of the fishes. The concentration of metals was higher in the muscle and gill than in the liver and intestine. The overall mean accumulation (pooled) of heavy metals (Table 1) in the tissues revealed high concentrations of lead ranging between 3.6±2.47 and 60.3±0.26 mg/kg, while the permissible limit is 0.5 mg/kg (FAO/WHO) [11]. The mean Pb concentration varied from 2.38 mg/kg in the gills to 51.07 mg/kg (Table 2). The Cd in the tissues ranged between 0.409±1.15 and 4.950±1.28 mg/kg, which were higher than the permissible limit of 0.5 mg/kg (FAO/WHO) [12, 13].

Heavy metals can accumulate in marine organisms through various pathways, including respiration, absorption, and ingestion, and can be biomagnified through the food chain, ultimately being assimilated by human consumers, resulting in health risks. Cadmium (Cd) in humans is well known to cause chronic toxicity that affects renal function, tumors, and hepatic dysfunction [14]. Bioaccumulation of metals results from interactions between physiological, chemical, and environmental factors.

The cumulative mean concentration of heavy metals was comparatively lower in the tissues of fishes analyzed from the Mandvi coast. The rate of accumulation of the detected heavy metals was maximum at Kandla, followed by Mundra, Pingleshwar, and Mandvi (Table 1). The highest concentration of Pb in muscle tissue was found in *Acanthopagrus arabicus* (60.300±0.68 mg/kg) and Cd in *Mugil parsia* (4.950±1.28 mg/kg) (Table 1). The *Cynoglossus* sp. from Pingleshwar coast showed concentrations of 42.35 mg/kg (Pb) and 2.63 mg/kg (Cd) in the muscle.

The mean concentration of Lead in the muscle of the analyzed fishes was 26.75 mg/kg, which is higher than the mean value for Cadmium (2.3185 mg/kg). The observed values from all the fishes were above the admissible limit (0.5 mg/kg) specified by FAO and FAO/WHO [12, 11]. In aquatic ecosystems, Lead can be easily absorbed into a fish's bloodstream through the skin and gill surfaces and accumulate in the viscera, subsequently entering the human body via consumption [15]. The concentration of Lead was very high in all the species analyzed, particularly *Acanthopagrus arabicus* (60.300±0.68 mg/kg) and *Thryssa purava* (60.100±0.52 mg/kg) from Kandla and *Epinephelus coioides* (50.9±1.85) from Mundra coast. In general, the fishes caught from Kandla showed the highest concentration of Lead in the tissues.

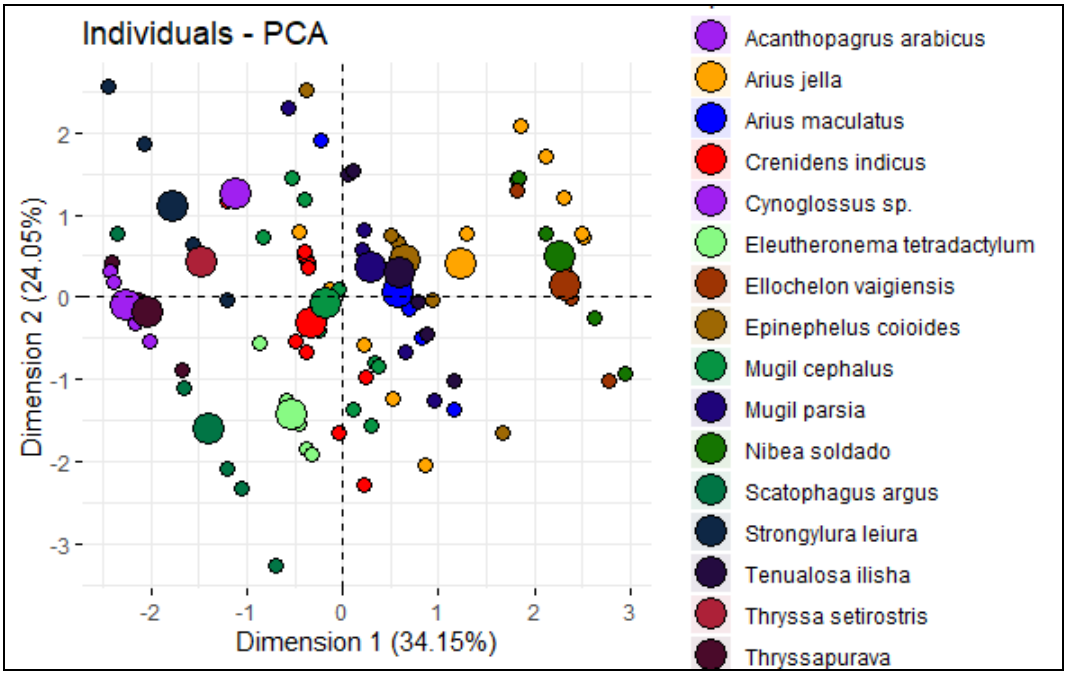

Fig 2: PCA analysis of the heavy metal concentration

The Principal Component Analysis (PCA) reveals that Dimension 1 explains 34.15% of the total variance in the data, capturing the largest proportion of variability among the species, while Dimension 2 adds 24.05% of the variance, representing the second-largest source of variability in the dataset (Figure 2). Together, Dimensions 1 and 2 account for 58.20% of the total variance, making the plot a robust summary of the dataset.

The permissible level of Cadmium in food products is 0.5 mg/kg (FAO, 1984) [21], and in the present study, it ranged between 0.409±1.15 mg/kg and 4.950±1.28 mg/kg in the fish tissues, indicating the potential health risk to consumers. The fishes obtained from Mundra and Kandla showed high values of Cadmium. Among the species, the maximum concentration was found in the muscle of *Thryssa purava* (3.50±1.06 mg/kg) from Kandla and in *Mugil parsia* (4.95±1.28 mg/kg) from Mundra and *Strongylura leiura* (4.90±1.28) from Mandvi coast. This non-essential metal gets deposited in the organs through regular consumption of bioaccumulated fish food as it has been considered carcinogenic for humans [16, 17].

This PCA plot provides a concise visualization of the multivariate relationships among 16 species. Each species is represented by a distinct color, as indicated in the legend on the right. Clusters and separations highlight the pattern of accumulation of heavy metals in different organs. Species on opposite ends of this axis differ significantly in the variables contributing to this dimension. It has been reported that fish liver accumulates the highest levels of metals [18] and is an exceptional indicator to determine inactivation and storage of them. [19] The fishes examined in this study from Kandla and Mundra showed high values of Cadmium, which varied depending on feeding habits [20], ecological needs, metabolism, [21] age, size of the fish [22] and their habitats [23]. It has been noted that gills are the primary target of metal toxicity as they have a higher affinity towards metal accumulation than the muscle [24, 10].

The PCA presents the variability in two dimensions: Dimension 1 and Dimension 2. For example, the fishes *Arius jella* (yellow), *Ellochelon vaigiensis* (dark brown), and *Thryssa purava* (dark maroon) are visually distinguishable and varied with respect to the accumulation of heavy metals. The clusters represent similarity in the rate of accumulation of heavy metals in different organs of the fishes. Species such as *Arius jella* (yellow) and *Scatophagus argus* (dark green) are grouped on the negative side of Dimension 1. This suggests that they share similar ecological traits or environmental preferences. Species like *Epinephelus coioides* (orange) and *Thryssa purava* (dark maroon) are located on the positive side, indicating contrasting traits or adaptations. Species like *Mugil cephalus* (light green) and *Ellochelon vaigiensis* (dark brown) (Mugilidae) occupy the upper quadrants, suggesting a strong association with variables contributing positively to this dimension. The fishes *Arius spp.* and *Tenualosa ilisha* collected from Mandvi coast showed Nickel in the muscle. The concentration was 2.85 mg/kg and 1.89 mg/kg in *Arius jella* and *T. ilisha* respectively. Nickel (Ni) is a necessary metal that occurs in trace amounts in the environment; however, no evidence of Ni deficiency has been reported in humans. The concentration of Ni in the above fishes collected from Mandvi was above the permissible limit (0.5-0.6 mg/kg) set by WHO (1995) [13] for food and drinking water.

Often, fish and other seafood represent one of the main sources of exposure to metals in the general population. Foods that contain toxic metals above the permitted levels are considered to be harmful to human health and are banned for trade by many national and international regulations [25]. The average value of Pb concentration in all the fishes was around 8.31 mg/kg, and it was several orders higher than the permissible limit recommended for human consumption according to FAO and WHO (Kaleshkumar *et al*., 2017) [26]. From the east and west coast of India, reports are available on the metals Nickel, Lead, and Cadmium values in edible fishes above the maximum permissible limits set by National and International food regulatory agencies (Zodape, 2014; Siji Thomas and Mohaideen, 2014) [27, 28]. Very high Cd concentrations (up to 195 µg g^{-1}) in the liver of some fishes from southern Oman is presumed to be due to the upwelling process in the region, which brings mixing of the bottom water with the surface layer (de Mora *et al*., 2004) [24]. According to EU (European Union 2006) [29], the maximum

tolerable limit (MTL) of Pb in fish is 0.5 mg/kg, whereas Cd and Mercury (Hg) are 0.05-0.3 and 0.5-1.0 mg/kg wet weight, respectively, depending on the type of fish.

Fishes accumulate heavy metals in the order gill > liver > muscle (Agbugui and Abe, 2022) [30] and also in the digestive tract (Milanov *et al*., 2016) [31]. Since muscle is the edible part, high concentrations of heavy metals may cause health risks to consumers (Mahmood and Alkhafaji, 2016) [32]. Studies have demonstrated that feeding habits have a great influence on the accumulation of heavy metals, and the rate varies with species. Accordingly, omnivorous fish were detected with elevated concentrations of Cd and Ni, whereas carnivorous fish had the highest concentration of Pb. Similarly, concentrations of Cd and Pb in fish tissues were positively correlated with body weight and also show a considerable degree of species specificity (Mitra *et al*., 2011) [33]. In the present study, carnivorous species such as *Acanthopagrus arabicus*, *Nibea soldado*, *Epinephelus coioides*, *Crenidens indicus*, and the zooplanktivores *Thryssa purava* and *Thryssa setirostris* showed high values of Lead in the muscles and gills compared to omnivores.

The three metals *viz*., lead, cadmium, and mercury have been included in the regulations of the European Union for hazardous metals (EC, 2001) [34], while the USFDA has included three additional metals chromium, arsenic, and nickel in the list (USFDA, 1993) [35]. The concentration of lead in the fishes was much higher than the permissible level set by the Indian Standards Institution for fishes intended for human consumption. Experts have increased the safe level, known as the Tolerable Daily Intake (TDI), from 2.8 micrograms per kilogram of body weight to 13 micrograms per kilogram of body weight. The findings point out the need for continuous monitoring of bioaccumulation of heavy metals in the Gulf of Kachchh and to avoid possible health risks through continuous consumption of the fishes inhabiting this region.

Table 1: Heavy metal concentration in the tissues of the selected fishes

Place	S. No	Species	Organs	Sample	Metal concentration (mg/kg)			
				(g)	Lead	Cadmium	Nickel	Chromium
Kandla	1	*Acanthopagrus arabicus*	Muscle	1	60.300±0.26	2.500±0.34	BDL	BDL
			Gill	1.2	52.833±1	2.417±1.08	BDL	BDL
			Liver	1	58.850±0.45	2.750±1.32	BDL	BDL
			intestine	1.1	57.045±0.92	2.636±1.22	BDL	BDL
	2	*Thryssa purava*	Muscle	1.4	45.000±0.39	3.500±1.06	BDL	BDL
			Gill	1	60.100±0.52	2.300±1.23	BDL	BDL
			Liver	1	50.900±10.95	1.300±0.74	BDL	BDL
			Intestine	1.5	40.400±0.32	2.967±0.99	BDL	BDL
	3	*Nibea soldado*	Muscle	1.2	46.583±1.25	2.667±0.16	BDL	BDL
			Gill	1.2	49.000±0.29	2.583±1.1	BDL	BDL
			Liver	1.6	34.156±0.63	0.844±0.27	BDL	BDL
			Intestine	1.1	48.773±0.88	0.909±0.85	BDL	BDL
			Stomach	1.6	36.375±0.07	1.969±0.78	BDL	BDL
	4	*Mugil cephalus*	Muscle	1.1	51.000±0.97	2.636±1.82	BDL	BDL
			Gill	1.1	51.273±0.73	2.636±0.37	BDL	BDL
			Liver	1.1	44.864±0.27	0.727±0.42	BDL	BDL
			Intestine	1.6	33.156±1.07	1.563±0.61	BDL	BDL
			Stomach	1.6	39.906±1.79	1.844±0.34	BDL	BDL
	5	*Arius jella*	Muscle	1.2	44.917±0.96	3.292±1.85	BDL	BDL
			Gill	1.3	42.192±1.44	2.656±1.92	BDL	BDL
			Liver	1.4	35.643±0.57	2.107±0.29	BDL	BDL
			Intestine	1.6	31.875±0.29	0.688±0.94	BDL	BDL
			Stomach	1.6	31.719±0.68	2.094±1.38	BDL	BDL
Mundra	6	*Mugil parsia*	Muscle	1.3	13.192±1.18	1.385±1.22	BDL	BDL
			Gill	1.1	10.955±4.01	2.500±1.77	BDL	BDL
			Liver	1	13.900±1.21	4.950±1.28	BDL	BDL
			Intestine	1	3.600±2.47	2.500±0.61	BDL	BDL
			Stomach	1.5	5.867±1.34	1.567±0.99	BDL	BDL
	7	*Scatophagus argus*	Muscle	1.4	16.143±0.4	1.000±1.3	BDL	BDL
			Gill	1.2	24.250±0.64	1.958±1.03	BDL	BDL
			Liver	1	19.850±1.34	4.200±1.84	BDL	BDL
			Intestine	1.7	14.500±0.3	1.029±1.14	BDL	BDL
			Stomach	1.4	20.429±0.5	1.357±1.84	BDL	BDL
	8	*Arius maculatus*	Muscle	1.2	27.583±0.65	2.167±0.6	BDL	BDL
			Gill	1.4	23.393±1.21	2.464±0.16	BDL	BDL
			Liver	1.3	25.692±0.89	1.308±0.56	BDL	BDL
			Intestine	1	31.850±1.02	3.800±2.01	BDL	BDL
			Stomach	1.7	22.912±0.37	1.971±0.11	BDL	BDL
	9	*Epinephelus coioides*	Muscle	1.4	35.250±0.15	3.750±2.16	BDL	BDL
			Gill	1	50.900±1.85	4.450±0.62	BDL	BDL
			Liver	1.3	31.154±0.16	1.885±0.96	BDL	BDL
			Intestine	1.3	33.769±0.32	3.077±0.65	BDL	BDL
			Stomach	1.7	22.176±0.29	1.324±0.39	BDL	BDL
		Crenidens indicus	Muscle	1.2	39.250±0.94	2.833±1.48	BDL	BDL
			Gill	1.2	36.875±0.35	0.542±0.86	BDL	BDL

	10		Liver	1.3	34.808±0.29	3.538±1.75	BDL	BDL
			Intestine	1.2	38.042±0.63	3.042±1.55	BDL	BDL
Mandvi	11	*Strongylura leiura*	Muscle	1.2	17.208±0.62	1.625±2.1	BDL	BDL
			Gill	1	28.300±0.89	4.900±1.28	BDL	BDL
			Liver	1	23.200±0.65	3.750±0.44	BDL	BDL
			Intestine	1.2	21.208±0.66	2.750±2.84	BDL	BDL
	12		Muscle	1	14.000±0.68	4.050±0.57	BDL	BDL
		Crenidens indicus	Gill	1.3	12.731±0.91	0.769±1.25	BDL	BDL
			Liver	1.3	10.962±0.2	2.731±0.72	BDL	BDL
			Intestine	1.6	8.250±2.4	1.375±2.15	BDL	BDL
			Stomach	1.1	9.591±0.75	1.455±2.92	BDL	BDL
	13		Muscle	1.1	4.955±2.38	1.273±1.52	BDL	BDL
			Gill	1	11.350±1.35	3.000±1.12	2.85	BDL
		Arius jella	Liver	1.2	4.417±2.46	2.625±0.84	BDL	BDL
			Intestine	1	4.100±1.18	2.450±1.31	1.7	BDL
			Stomach	1.1	5.091±0.94	1.318±1.24	1.818	BDL
			Muscle	1.2	19.667±1.3	1.042±0.31	BDL	BDL
			Gill	1.1	20.318±1.25	3.864±0.75	BDL	BDL
	14	*Tenualosa ilisha*	Liver	1.1	15.091±0.06	4.000±0.28	BDL	BDL
			Intestine	1.4	14.571±0.82	1.179±1.2	1.893	BDL
			Stomach	1.1	16.045±1.61	1.227±0.53	BDL	BDL
	15		Muscle	1.1	20.727±0.51	2.409±0.22	BDL	BDL
			Gill	1.4	17.321±0.92	1.964±1.32	BDL	BDL
		Ellochelon vaigiensis	Liver	1.4	16.143±0.89	1.821±1.59	BDL	BDL
			Intestine	1.3	11.731±0.61	1.962±1.49	BDL	BDL
			Stomach	1.6	14.844±0.96	1.188±0.79	BDL	BDL
	16	*Mugil cephalus*	Muscle	1.4	9.393±1.7	2.000±0.39	BDL	BDL
			Gill	1.1	14.909±1.37	0.409±1.15	BDL	BDL
			Liver	1	9.650±1.86	3.850±2.22	BDL	BDL
			Intestine	1.3	11.846±1.14	3.077±2.92	BDL	BDL
			Stomach	1.1	6.682±0.5	3.955±3.27	BDL	BDL
	17	*Eleutheronema tetradactylum*	Muscle	1.6	12.813±0.22	1.750±1.02	BDL	BDL
			Gill	1.4	14.286±0.73	1.786±1.24	BDL	BDL
			Liver	1.4	13.321±0.77	3.000±1.25	BDL	BDL
			Intestine	1.6	15.563±0.82	1.813±1.43	BDL	BDL
Pingleshwar	18	*Thryssa setirostris*	Muscle	1.1	43.136±0.89	2.364±0.69	BDL	BDL
	19	*Cynoglossus* sp.	Muscle	1	46.700±0.62	2.950±0.9	BDL	BDL

Table 2: The mean concentration of heavy metals in all fishes from the four stations

Station	Tissue	Mean concentration of heavy metals (mg/Kg)			
		Lead	Cadmium	Nickel	Chromium
Kandla	Muscle	49.56	2.91	0	0
	Gill	51.07	2.51	0	0
	Liver	44.88	2.36	0	0
	Intestine	42.24	1.75	0	0
	Stomach	36.00	1.96	0	0
Mundra	Muscle	26.28	2.22	0	0
	Gill	29.27	2.38	0	0
	Liver	25.08	3.17	0	0
	Intestine	24.35	2.68	0	0
	Stomach	17.84	1.55	0	0
Mandvi	Muscle	14.10	2.21	0	0
	Gill	2.38	2.38	2.85	0
	Liver	13.25	3.11	0	0
	Intestine	12.46	2.08	1.79	0
	Stomach	10.45	1.82	1.81	0
Pingleshwar	Muscle	42.35	2.62	0	0

Conclusion

Totally sixteen commercially important fishes collected from Kandla, Mundra, Mandvi, and Pingleshwar were selected for the analysis of Pb, Cd, Ni, and Cr accumulation in the tissues. The two heavy metals, Pb and Cd were found in the tissues of all the fishes analyzed and the level was higher than the permissible limit specified by FAO and WHO. The study acclaims the necessity to have detailed investigations of heavy metal monitoring in the environment and the bio resources in the Gulf of Kachchh to address the safety of the fish consumers.

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Conflict of interest
The authors declare that there is no conflict of interests.

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