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Quantitative trace level heavy metals (Cd, Cr and Pb) toxicity analysis in various fish species muscles by ICP-OES: A continuous threat for human consumption

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Key words: Heavy metal toxicity, Cd, Cr, Pb, fish muscle, Punjab regions, ICP-OES

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Abstract

The quantitative analysis of toxic heavy metals: Cadmium (Cd), Chromium (Cr) and lead (Pb) up to trace levels was performed in the fish muscles by ICP-OES following protocols of AOAC. The objective of this research basically included metal toxicity comparative evaluation levels from three chosen target sources namely River Ravi, Government fish ponds and Privately established fish farms distributed all over the Punjab, Pakistan. A total number of 300 fish samples comprising of thirteen different fish species were collected from twenty-one different sites of Punjab regions during 2011-2013. The heavy metals were found present in varying degrees and extents in some of the fish species. The results of the study interpreted that the heavy metals contamination in river fish were mostly at high concentration levels and above the maximum permissible limits of FAO (1983)/WHO guidelines (1985, 1989) which suggested high health risk to the human body through their frequent consumption. However, the situation was vice versa and was found within safe limits in case of Government fish ponds and mostly Private fish farms.

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Introduction

The consumption of fish and its products have quite increased worldwide during previous years following their high health frame performances, better cardiovascular disease control outcomes and health benefits received enhancing its overall utilization graph values (Cahu *et. al.,* 2004). Fish are the significant component of the human nourishment due to its small degree of saturated fat, high level of protein content and adequate omega fatty acids which as a whole support good health conditions, therefore, various research aspects have been under taken worldwide on the poisonous degrees of contaminations received in various fish species through introduction of heavy metals (Sivaperumal *et. al.,* 2007; Raychaudhuri *et. al.,* 2008; Raouf *et. al.,* 2009; Yilmaz *et. al.,* 2009; Bhattacharyya *et. al.,*2010). Since the Human industrial activities have increasingly intervened now-a-days resulting in the introduction of highly toxic compounds into the aquatic environment (Udonsen, 1998), therefore the naturally existing aquatic environments have been highly polluted and contaminated through the release of heavy metals from industrial, domestic and miscellaneous man-made activities (Conacher *et. al.,* 1993).

Although the trace quantities of these heavy metals may not cause any health problem but their higher concentration may lead to their bioaccumulation in various parts of fish, water and vegetables, which has been confirmed by a lot of multifarious research carried out in this field (Iqbal and Kataria, 2006; Kachenko and Singh, 2006; Pandey *et al.,* 2006; Sumant Rao *et al.,* 2008; Amiya *et al.,* 2012; Jacob and Kakulu, 2012; Opaluwa *et al.,* 2012).

Cadmium (Cd), Chromium (Cr) and lead (Pb) comes under the category of toxic heavy metals and their evaluation is an essential part of studies because of their vital role in environmental phenomena. These concentrates in fish bodies making them polluted which after consumption ultimately results in the appearance of diversified diseases in humans beings.

The presence of these metals in the aquatic systems can not only risk the aquatic biota but also can badly affect the fish consumptions through humans (Forstner and Wittmann, 1983).

The fish accumulates toxic/heavy metals directly from aquatic environment and food, and the contamination ratio can cross hundreds to thousands times higher concentrations as compared to be measured in water i.e., its living environment, the bed /sediments and its natural food (Labonne *et al.,*2001; Goodwin *et al.,*2003; Osman *et al.,*2007).

They have become serious threats for human consumption because of the acute toxicity, bioaccumulation and high persistence values of heavy metals contained in their bodies (Eisler, 1988).

Fish have been widely used worldwide as a bio-indicator of pollution by heavy metals.

The muscle tissue of fish is the main edible part of the fish, hence, it was the target tissue for this research analysis.

The main objective of this study is to comparatively analyze the presence and level/concentration of Cd, Cr and Pb in the muscle tissues of fish from government farms with those from private established fish farms, from River Raviand then finally to quantify this research in terms of heavy metals toxicity levels.

Materials and methods

The present research was carried out at Chemistry Section, Fish Quality Control Labs, Fisheries Research & Training Institute (FR&TI) situated in Lahore, Pakistan.

Study Area/Sites

The Fish samples have been collected from twenty-one selected regions located in Punjab province of Pakistan as described in Table 1. These regions can be well understood through the map of Punjab, Pakistan (Fig 1).

Fig. 1. Map of Punjab showing various districts under study.

Thirteen different fish species were collected from these regions from natural sources i.e., River Ravi, from Government and private fish farms/ponds as indicated in Table 2.

Sample Collection Procedure

The fish samples were freshly collected from different sources; weight and length was recorded on site location; then were transferred to separate labeled polythene bags; placed in ice containers and immediately shifted to the ICP sample preparation lab and placed in deep freezer at-20^{o}Cwithout any delay.

Technical Section Reference and Procedural Flow Sheet

The FAO Manual of Methods for Analysis of Metals in Fish (1983) was followed for digestion of fish samples. The AOAC, Official methods of analysis 990.08 was being followed for ICP-OES instrumental metal analysis conditions, requirements and analysis purposes. The flow sheet for description of the procedure adopted for fish samples digestion is shown in Fig 2.

Reagents and Preparation of Standard Solutions

The reagents/chemicals being used included Nitric acid, Hydrogen peroxide, Cadmium chloride, Chromium chloride and Lead nitrate. All chemicals of Analytical reagent grade with 99.999% purity were used for analysis. Deionized water was utilized for preparation of standards and dilutions from aforementioned reagents.

Machinery and Equipment

The machinery and equipment included Inductively Coupled Plasma-Optical Emission Spectrometer (PerkinElmer Optima 7000 DV) with all its accessories, Analytical balance (Ohaus Pioneer PA224 Brand), Hot plate (PCSIR, Lahore Brand) and pH Meter (Jenway 3505 Brand).

Sample analysis method

The ICP-OES was turned on with full laboratory protocols. The blank solution was used to set the zero emission and was again analyzed during sample analysis for blank emission reading. The calibration curves of metals under consideration were constructed using a series of standard solutions at their specific emission wavelengths; Cadmium at 228.802 nm, Chromium at 267.716 nm and Lead at 217.000 nm, respectively.

The calibration curves thus developed with Correction Coefficient of Calibration above 0.9970 were accepted and selected while all below 0.9970 were rejected and all the calibration procedure was again repeated. The fish samples were then analyzed by ICP-OES, their emissions recorded and the concentrations were obtained from the respective calibration line for each element. The final concentration of each sample was obtained by subtracting the concentration in the test portion from blank emission reading, then multiplying with the dilution factor and finally dividing by the weight of fish in grams.

Statistical analysis

The heavy metals from different fish species and different sampling sites were compared statistically following Steel *et al.*, 1996. The SPSS software 22 was used for statistical analysis. The figures having comparative bar graphs were established with the help of Microsoft Excel 2016.

Results and discussion

Fish samples collection data

In the present study, a total of 300 fish samples were collected from twenty-one different sites of Punjab, Pakistan. From these sites, mainly a good variety of various fish sample species were collected from government fish ponds, private fish farms and River Ravi. A total of 142 fish samples were collected from Government fish ponds for analysis purposes, their break up detail is being described in Table 3 and Fig 3;while110 fish samples were collected from private fish farms as described in Table 4 and Fig. 3. Furthermore, 48fish samples were collected from River Ravi for analysis purposes; their break up detail has been mentioned in Table5 and Fig 3.

Fish Weight and Length data

The Weight and Length of all fish samples, collected from Government fish ponds, Private fish farms and River Ravi, were taken immediately on site before bringing to the laboratory and before freezing the samples. These are well described in Table6,7,8, respectively.

Table 1. Selected sampling regions of Punjab.

Sr. No.	Divisions	Districts	Locations / Sites	Allocated Site No.
1.	Bahawalpur	Bahawalpur	Bahawalpur	S -1
2.	Gujranwala	Gujranwala	Chhenawan	S -2
3.	D.G. Khan	D.G. Khan	D.G. Khan	S -3
4.	Faisalabad	Faisalabad	Faisalabad	S -4
5.	Lahore	Sheikupura	Farooqabad	S -5
6.	Gujranwala	Gujranwala	Gujranwala	S -6
7.	Faisalabad	Jhang	Jhang	S -7
8.	Lahore	Kasur	Kasur	S -8
9.	Multan	Khanewal	Khanewal	S -9
10.	Gujranwala	Sialkot	Kotliarian	S -10
11.	Lahore	Lahore	Lahore	S -11
12.	Multan	Khanewal	Mianchannu	S -12
13.	Multan	Multan	Multan	S -13
14.	D.G. Khan	Muzafargarh	Muzafargarh	S -14
15.	Lahore	Nankana Sahib	Nankana Sahib	S -15
16.	Sahiwal	Pakpattan	Pakpattan	S -16
17.	Faisalabad	T. T. Singh	Pirmahal	S -17
18.	Lahore	Sheikhupura	Sheikhupura	S -18
19.	Gujranwala	Sialkot	Sialkot	S -19
20.	Bahawalpur	Vehari	Vehari	S -20
21.	Rawalpindi	Rawalpindi	Rawalpindi	S -21

It is evident from all above tables that there is a marked difference in the observed fish weight from all three sites; especially the fish collected from Government ponds were having higher weight as compared to the other two sites while fish from River Ravi were having lowest weight among the total three sites under consideration.

Permissible heavy metals limits

Lead **(**Pb) and Cadium (Cd) are characterized as heavy, toxic and non-essential group of elements which have no role in biochemical functions of human body. However, Cr is an essential metal but becomes toxic at higher concentrations (Wagner and Boman, 2003).

Table 2. Types of fish species collected.

Sr. No.	Fish species		Allocated Fish No.
	Common names	Scientific names	
1	Rohu	Labeo rohita	F -1
2	Mrigal	Cirrhinus *mrigala*	F -2
3	Thaila	*Catla catla*	F -3
4	Grass carp	Ctenopharyngodon idella	F -4
5	Silver carp	*Hypophthalmichthys molitrix*	F -5
6	Common carp	Cyprinus carpio	F -6
7	Tilapia	Oreochromis mossambica	F -7
8	Big head	Hypophihal michthys	F -8
9	Daula	Channa punctatus	F -9
10	Pari	Notopterus notopterus	F -10
11	Mullee	Wallago attu	F -11
12	Khagga	Rita rita	F -12
13	Singarhi	Sperata sarwari	F -13

The Permissible Limits of these Heavy Metal Concentrations in Fish are shown in Table 9 after FAO (1983)/WHO guidelines (1985, 1989).

The observed levels of heavy metal concentrations were compared with available certified safety guidelines proposed by FAO/WHO.

Heavy metals Concentration in fish samples from Government fish ponds

Table 10 and Fig 4mentions the analyzed concentrations of all heavy metals under consideration in fish samples collected from various Government fish ponds.

Table 3. Break-up detail of number of fish samples collected from Government fish ponds.

		Allocated Sample No.							
Sr. No.	Allocated Location No.	F-1	F-2	F-3	F-4	F-5	F-6	F-7	F-8
1.	P-1	2	2	2	1	1	1	2	2
2.	P-2	5	2	2	2	2	2	2	2
3.	P-5	2	1	1	1	1	1	1	1
4.	P-7	1	1	1	1	1	2	-	-
5.	P-11	3	2	2	1	1	3	2	1
6.	P-12	4	1	2	2	2	1	-	-
7.	P-16	2	2	1	2	2	2	1	-
8.	P-17	2	1	1	2	2	1	-	1
9.	P-19	5	2	1	2	2	2	-	1
10.	P-20	2	2	2	2	2	1	1	1
11.	P-21	2	2	1	5	4	3	1	-
Total		29	18	16	21	20	19	10	9
Grand Total		142							

Table 4. Break-up detail of number of fish samples collected from Private fish farms.

		Allocated Sample No.					
Sr. No.	Allocated Location No.	F-1	F-2	F-3	F-4	F-5	F-6
1.	P-1	2	1	1	2	1	1
2.	P-3	4	2	1	2	1	1
3.	P-4	1	1	1	1	1	1
4.	P-6	4	3	3	1	2	1
5.	P-8	1	1	1	1	1	1
6.	P-9	1	1	2	2	2	1
7.	P-11	4	1	1	4	3	3
8.	P-13	4	1	3	1	1	1
9.	P-14	3	1	3	1	1	1
10.	P-15	2	4	1	1	2	1
11.	P-16	2	1	1	1	2	1
	Total	28	17	18	17	17	13
Grand Total					110		

It can be well understood from the analyzed data that the heavy metals Cd and Cr under consideration were not detected whereas Pb was detected within the desirable range in the fish samples under study thus proving it suitable for human consumption and nontoxic in respect of health concerns. This may be mainly attributed to the clean water supply systems available at the Hatcheries, Nursery Units and District Farms under Government command.

Table 5. Break-up detail of number of fish samples collected from River Ravi.

		Allocated Sample No.							
Sr. No.	Allocated Location No.	F-1	F-2	F-3	F-9	F-10	F-11	F-12	F-13
1.	P-11	6	5	4	3	1	3	3	3
2.	P-18	3	3	3	2	2	3	2	2
	Total	9	8	7	5	3	6	5	5
Grand Total						48			

Table 6. Weight and length of various fish species collected from Government ponds.

Allocated Fish Specie No.	Weight (in g)			Length (in cm)		
	Min.	Max.	Mean	Min.	Max.	Mean
F-1	251.0	1678.6	646.3	31.0	49.5	36.6
F-2	195.0	766.0	385.2	21.0	49.0	32.1
F-3	422.0	3522.1	1392.4	31.0	49.5	35.9
F-4	477.5	780.5	548.1	31.5	48.0	37.6
F-5	340.1	1803.2	861.4	31.0	55.0	43.4
F-6	200.0	521.1	354.9	25.0	35.0	29.5
F-7	120.3	125.0	122.5	20.0	25.0	22.5
F-8	450.2	2039.1	1554.4	30.2	54.0	34.0

The sources of water supply to the fish ponds is through well managed pipelines from either Government supply channels, tube wells or boring at sufficient recommended depth within earth.

Heavy metals Concentration in fish samples from Private fish farms

Table 11 and Fig 5reveals the concentrations of all heavy metals under consideration analyzed in fish samples collected from various Private fish farms.

Table 7. Weight and length of various fish species collected from Private farms.

Allocated Fish Specie No.	Weight (in g)			Length (in cm)		
	Min.	Max.	Mean	Min.	Max.	Mean
F-1	210.0	855.2	455.3	24.0	38.0	26.5
F-2	146.8	676.4	447.1	21.0	40.0	31.3
F-3	140.5	618.0	477.0	26.0	42.0	33.7
F-4	250.3	1423.1	561.1	30.7	26.0	39.0
F-5	234.9	952.7	554.8	30.0	46.0	37.3
F-6	132.1	371.0	224.1	20.0	30.0	25.0

The data reveals that the heavy metals Cadmium and Chromium were detected in some of the fish samples but mainly within range as per recommendations of FAO (1983)/WHO guidelines (1985, 1989). Only rare deviations from the suitable range in case of Cr and Pb were observed which were not considerable as a whole since the scenario was favorable on wide level. This leaded to the results that even private fish farming at Punjab level is providing people of Pakistan a healthy fish diet for consumption purposes not only fulfilling their food requirements and needs but also improving their health through high quality meat availability.

Heavy metals Concentration in fish samples from River Ravi

Table 12 and Fig 6 mentions the concentrations of all heavy metals under consideration, analyzed in fish samples collected from River Ravi.

It is evident from the data that the heavy metals under consideration were not only detected in the river fish but also were indicative of the high pollution rate and contamination.

Table 8. Weight and length of various fish species collected from River Ravi.

Allocated Fish Specie No.	Weight (in g)			Length (in cm)		
	Min.	Max.	Mean	Min.	Max.	Mean
F-1	170.0	258.3	198.6	22.0	25.6	23.4
F-2	132.3	450.0	288.2	18.0	32.2	24.6
F-3	120.1	380.3	245.7	16.0	30.2	23.3
F-9	77.0	250.0	163.5	16.0	20.1	18.1
F-10	158.3	400.6	235.1	22.2	30.0	24.3
F-11	170.1	700.5	459.5	20.0	42.7	37.0
F-12	220.5	396.8	295.3	20.3	33.0	28.4
F-13	85.0	350.2	110.0	27.0	33.0	27.0

Table 9. Permissible limits of heavy metals concentration in muscle tissues of fishes.

Sr. No.	Metals	FAO/WHO guidelines (µg g^{-1})
1.	Cd	1.0
2.	Cr	0.15 – 1.0
3.	Pb	2.0

Table 10. Concentration of heavy metals in various fish species collected from Government ponds.

Allocated Fish Specie No.	Cd (µg g^{-1})			Cr (µg g^{-1})			Pb (µg g^{-1})		
	Min.	Max.	Mean	Min.	Max.	Mean	Min.	Max.	Mean
F-1	N.D	N.D	N.D	N.D	N.D	N.D	0.01±.002	0.05±.004	0.04±.002
F-2	N.D	N.D	N.D	N.D	N.D	N.D	0.01±.001	0.03±.001	0.02±.001
F-3	N.D	N.D	N.D	N.D	N.D	N.D	0.09±.008	0.25±.012	0.12±.005
F-4	N.D	N.D	N.D	N.D	N.D	N.D	0.10±.007	0.15±.013	0.13±.006
F-5	N.D	N.D	N.D	N.D	N.D	N.D	0.03±.001	0.09±.007	0.05±.004
F-6	N.D	N.D	N.D	N.D	N.D	N.D	0.21±.011	0.30±.015	0.25±.015
F-7	N.D	N.D	N.D	N.D	N.D	N.D	0.15±.011	0.33±.020	0.22±.016
F-8	N.D	N.D	N.D	N.D	N.D	N.D	0.21±.008	0.34±.031	0.30±.022

Comparative Heavy metals Concentration data from all three sites

Fig 7 shows a comparative average concentration of pooled data of heavy metals from all the three compared target sites.

The higher value of Cd, Cr and Pb in river fish is due to the presence of heavy metal contents present in the water bodies which accumulates in the fish bodies.

The river fish were not found suitable for human consumption and proved to be highly toxic in respect of health concerns as per FAO (1983)/WHO guidelines (1985, 1989).

Table 11. Concentration of heavy metals in various fish species collected from Private farms.

Allocated Fish Specie No.	Cd (µg g^{-1})			Cr (µg g^{-1})			Pb (µg g^{-1})		
	Min.	Max.	Mean	Min.	Max.	Mean	Min.	Max.	Mean
F-1	N.D	N.D	N.D	N.D	N.D	N.D	0.11±.008	1.23±.086	0.55±.028
F-2	N.D	N.D	N.D	N.D	N.D	N.D	0.45±.025	0.95±.057	0.36±.014
F-3	0.30±.021	0.46±.027	0.35±.018	0.45±.030	1.36±.082	0.84±.040	0.29±.020	1.64±.114	0.65±.027
F-4	0.11±.009	1.06±.053	0.87±.053	0.34±.025	1.69±.085	0.47±.031	0.21±.011	1.25±.077	0.54±.090
F-5	0.28±.014	0.45±.029	0.33±.029	N.D	N.D	N.D	0.24±.013	0.45±.022	0.33±.011
F-6	N.D	N.D	N.D	0.32±.025	0.57±.023	0.39±.028	1.22±.085	2.59±.078	1.78±.112

Table 12. Concentration of heavy metals in various fish species collected from River Ravi.

Allocated Fish Specie No.	Cd (µg g^{-1})			Cr (µg g^{-1})			Pb (µg g^{-1})		
	Min.	Max.	Mean	Min.	Max.	Mean	Min.	Max.	Mean
F-1	0.21±.011	1.03±.041	0.63±.044	0.58±.035	3.74±.189	2.42±.145	0.87±.052	3.88±.232	1.09±.036
F-2	0.35±.025	4.50±.270	3.45±.181	0.79±.047	1.98±.099	1.02±.035	0.12±.008	2.56±.129	1.62±.081
F-3	0.11±.007	3.44±.170	2.88±.115	0.63±.039	2.57±.102	1.91±.115	0.33±.021	3.74±.221	2.60±.130
F-9	0.42±.020	1.77±.022	0.83±.041	0.85±.052	4.36±.251	2.53±.172	0.57±.399	4.11±.328	3.12±.121
F-10	0.19±.013	0.55±.115	0.23±.014	0.74±.050	2.01±.161	1.40±.060	0.45±.023	2.65±.079	1.35±.053
F-11	0.52±.042	1.65±.083	1.39±.111	1.32±.065	3.60±.216	2.03±.142	0.36±.024	4.25±.230	2.99±.149
F-12	0.64±.051	3.74±.223	2.13±.106	0.95±.055	3.33±.191	1.14±.060	1.84±.092	3.25±.162	2.75±.112
F-13	0.05±.004	1.31±.052	1.03±.045	1.00±.058	6.57±.525	5.11±.306	0.91±.033	3.08±.150	1.01±.037

Comparative Cd Concentration data from all three sites

Fig 8 is a comparative explanation of the Cd concentration in each of the fish species from three sites under consideration which indicates that higher concentrations above FAO levels were being shown by *Cirrhinus mrigala, Catla catla, Wallago attu and Rita rita* obtained from River Ravi fish samples only.

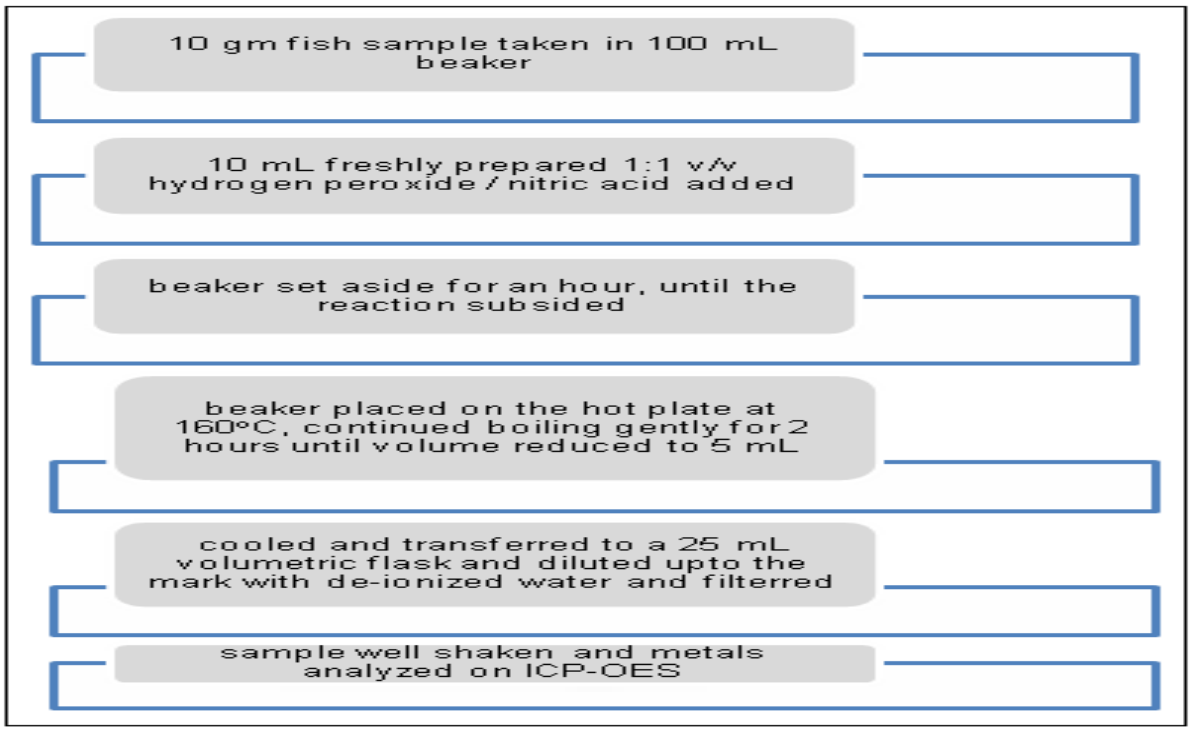

Fig. 2. Flow sheet diagram of adopted digestion procedure.

(G.P.-Government fish ponds, P.F.-Private fish farms, R.R.-River Ravi)

Fig. 3. Total number of each fish species collected from all three sites.

Comparative Cr Concentration data from all three sites

Fig 9 shows the comparison of Cr concentration in each of the fish species from three sites under consideration which indicates that the higher concentrations above FAO levels were found in *Catla catla, Channa punctatus, Notopterus notopterus, Wallago attu* and *Sperata sarwari* obtained from river Ravi fish samples only.

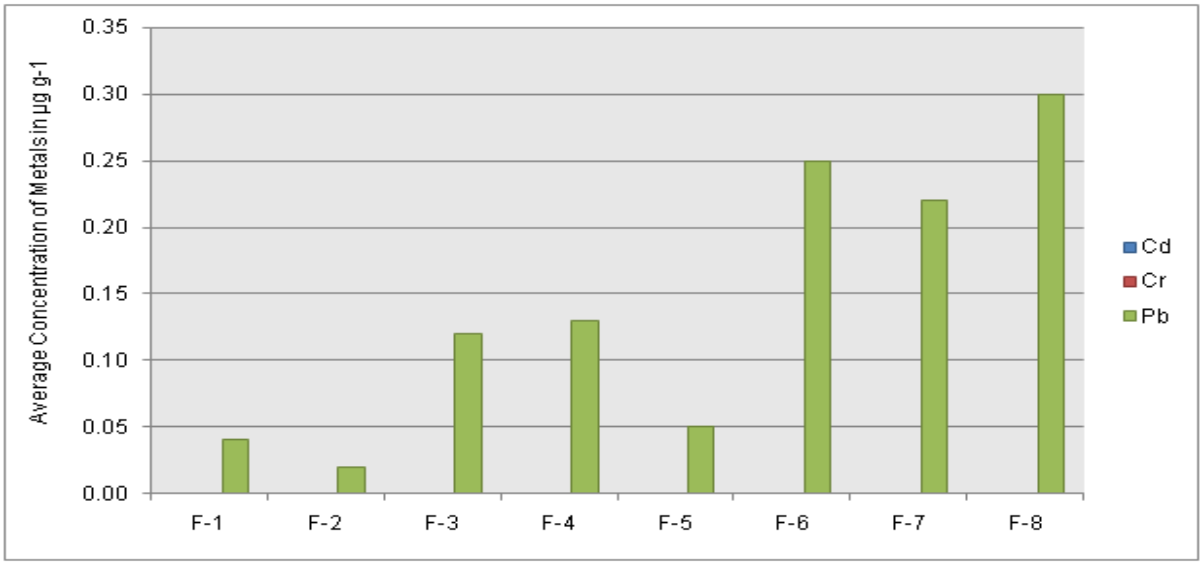

Fig. 4. Average concentration of heavy metals in various fish species collected from Government ponds.

Comparative Pb Concentration data from all three sites

Fig 10 explains the comparison of Pb concentration in each of the fish species from three sites under consideration which indicates that higher concentrations above FAO levels were being depicted by *Catla catla, Channa punctatus, Wallago attu* and *Rita rita* obtained from River Ravi fish samples only.

The results of the study interpreted that the heavy metals were found present in varying degrees and extents in some of the fish species muscles, however, the same were found either absent or within the suitable ranges in majority of the other fish species.

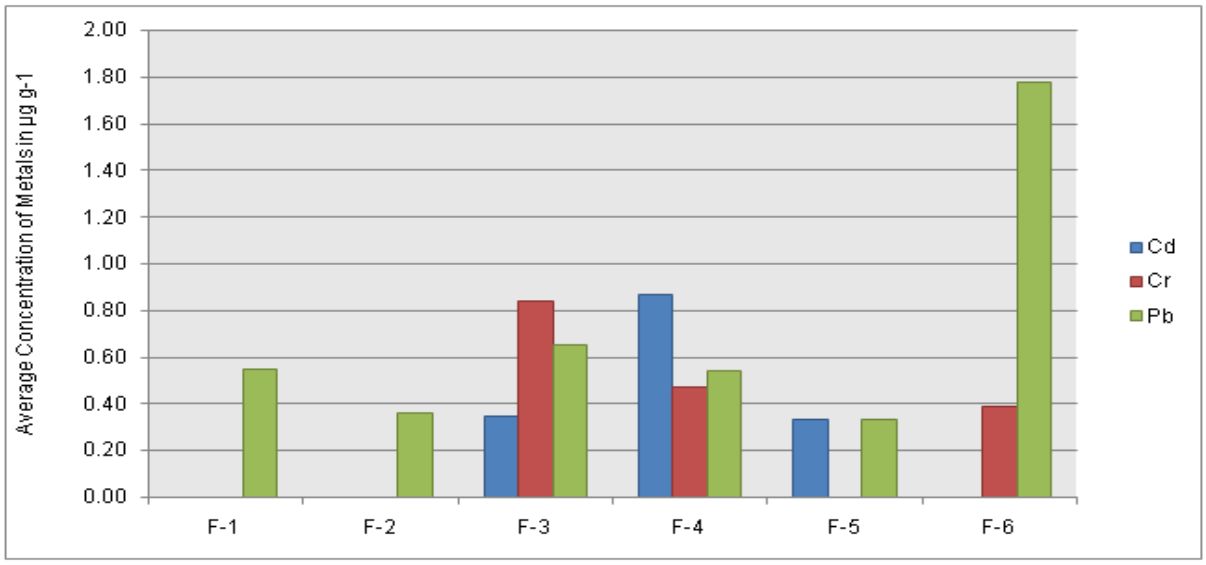

Fig. 5. Average concentration of heavy metals in various fish species collected from Private farms.

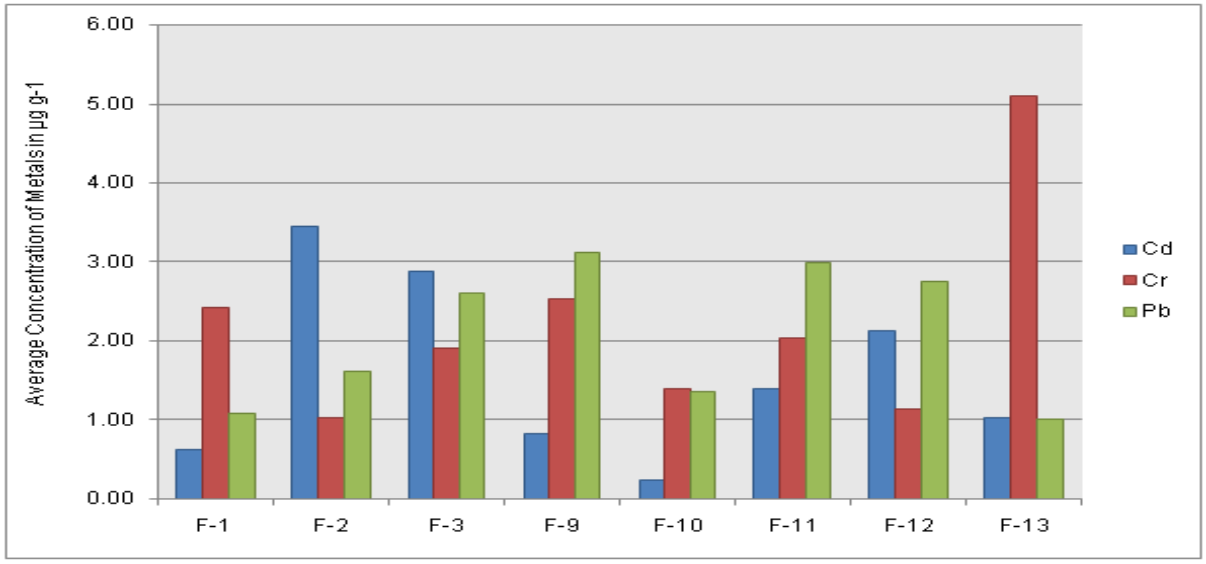

Fig. 6. Average concentration of heavy metals in various fish species collected from River Ravi.

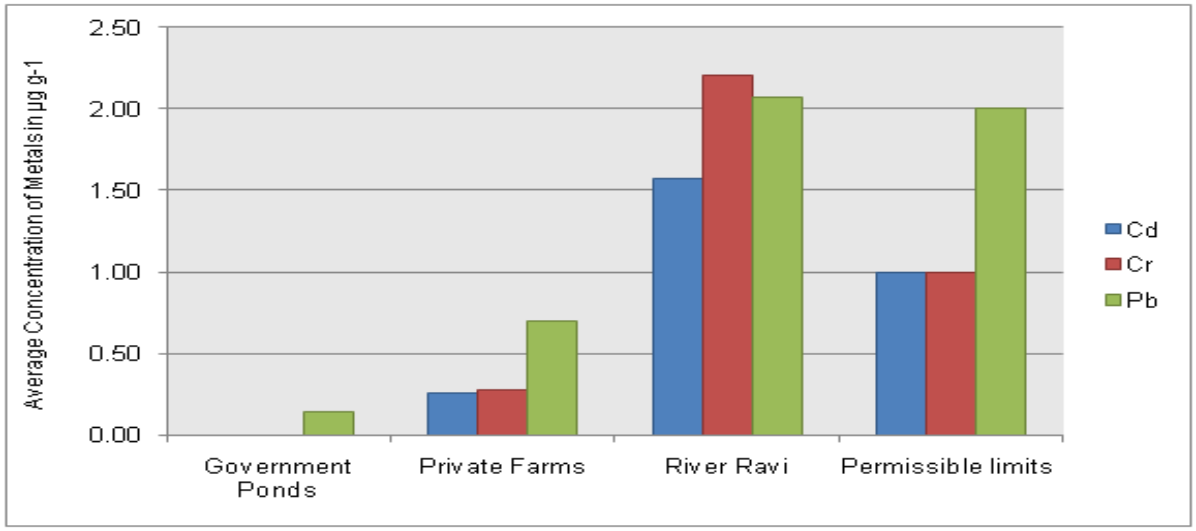

Fig. 7. Comparative average concentration of pooled data of heavy metals from all three targets.

The results depicted above also clearly indicates that the fish obtained from government as well as private fish farms were mostly with metal toxicity free levels. However, the out of range results and high concentrations of metals in river source may be mainly attributed to the discharge from Industries, Mills effluent discharge, tanning processing units, fertilizers run off, pesticides/fungicides/algaecide run off, Anthropogenic activities, etc coming into River Ravi, thus polluting the whole aquatic system and organisms making them unfit for human consumption.

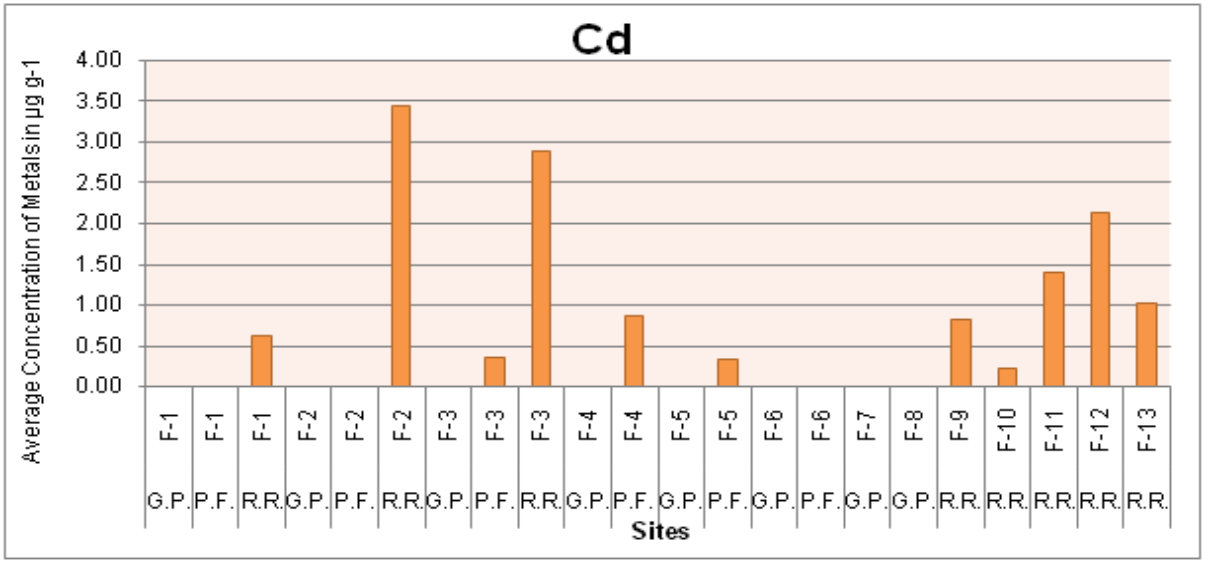

Fig. 8. All sites data comparison with each fish type for Cd metal concentration.

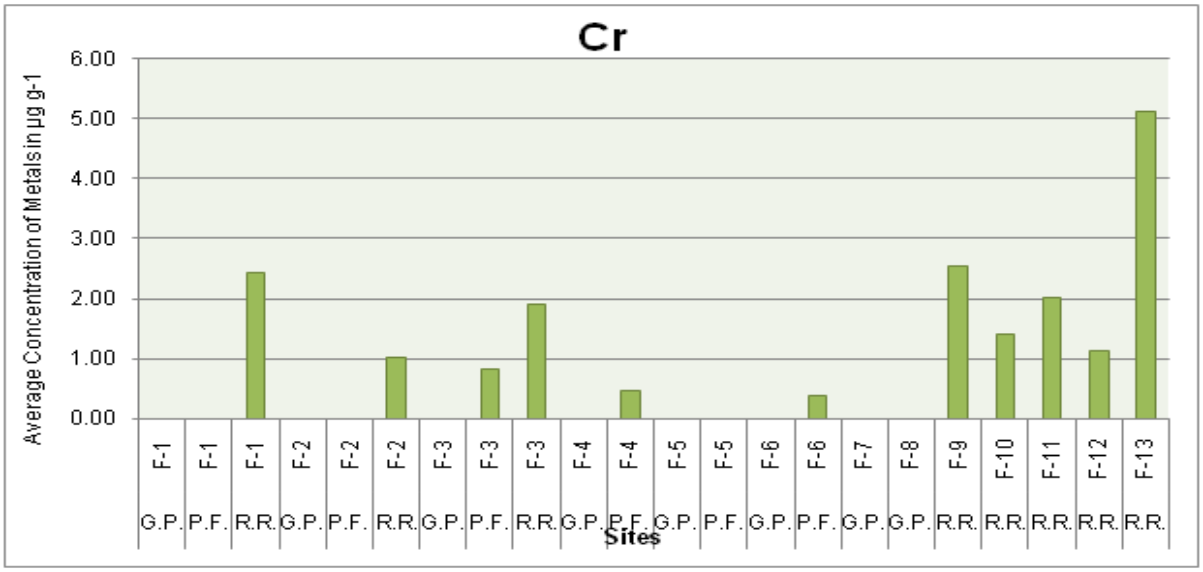

Fig. 9. All sites data comparison with each fish type for Cr metal concentration.

This is an alarming state of high pollution levels which should not be neglected and the remedial actions should be timely taken to control this aggravated situations of our river waters.

Similar studies have been conducted worldwide by many scientists showing high pollution levels due to heavy metal toxicity in various fish species making them risky for human consumption. A similar study was conducted by Uzairu *et. al.* (2009) who showed that the concentration of trace elements including Cd, Cr and Pb in fish from Kubanni River was found risky for human health while consuming fish livers as compared to fish muscles.

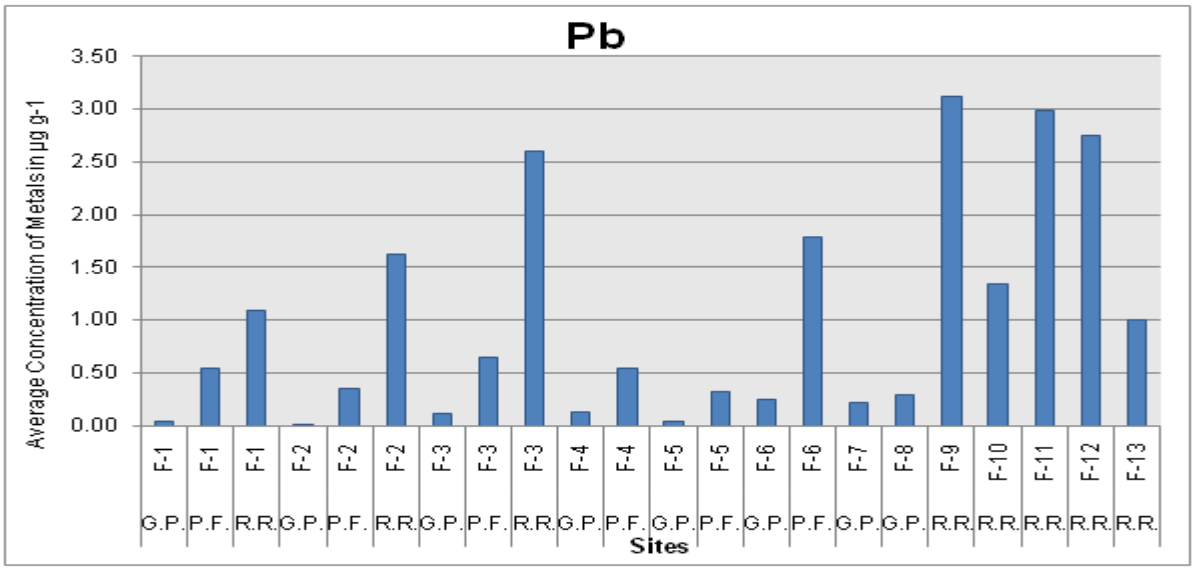

Fig. 10. All sites data comparison with each fish type for Pb metal concentration.

The results of Akoto (2014) were contrary with our studies which proved that there was no significant potential health risk involved with the consumption of lagoon fish. Özparlak *et. al* (2012) determined the concentrations of some heavy metals in muscle tissues of nine fish species from Beyşehir Lake, Turkey and showed that the Cr concentration exceeds the tolerance levels of FAO & WHO guidelines. Our studies are also in agreement with the research of Tiimub *et. al.* (2013) who found out that the increased concentrations of heavy metals Cd and Pb accumulated in the catfish and tilapia samples from Densu River at Weija might be due to the increased anthropogenic activities, domestic wastes and agricultural influx flows coming into the river.

This can be associated with the absence of Industrial and domestic activities near that particular Coast. Önder *et. al.* (2013) studied Cd, Cr and Pb metal levels in the male and female cuttlefish from Mediterranean and Cd/Pb were found over limited. Hossam *et. al.* (2017) analyzed manganese (Mn), copper (Cu), zinc (Zn), nickel (Ni), cobalt (Co), lead (Pb) and cadmium (Cd) concentrations in the muscles of grey mullet (Mugil cephalus) collected from Gaza fishing harbor and concluded that there was no human health risk elevated from the consumption of fish from this site. However, Quntulan *et. al.* (2015) studies showed that the fish coast of Baluchistan, Pakistan was safe site for consuming fish since the heavy metals did not exceed the safe limits which was also confirmed by Rauf *et. al.* (2009). Önder *et. al.* (2013) studied Cd, Cr and Pb metal levels in the male and female cuttlefish from Mediterranean and Cd, Pb were found over limited.

Conclusion

The results of the findings of this research revealed valuable knowledge on the heavy metals accumulation in the muscle tissue of the freshwater fish obtained especially from River Ravi, a natural resource as compared to the fish collected from Private fish farms and Government fish ponds.

The concentrations of the heavy metals in river fish were mostly above the maximum permissible limits as recommended by regulatory agencies and these portrait a human health risk through their intake/frequent consumption and thereby their accumulation in the human body. However, the Government fish ponds and Private fish farms depicted a bright picture of healthy fish production mainly attributed to the availability of cleaner water supply sources.

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