

Research Article

Assessment of heavy metal bioaccumulation in the muscle tissue of Caspian kutum (*Rutilus frisii kutum*) from the Siah Darvishan River, Guilan Province, Iran

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Keywords

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Abstract

In this study, the bioaccumulation of eleven metals (As, Fe, Hg, Zn, Pb, Se, Cd, Cr, Cu, Mn, and Ni) in the muscle tissue of 25 Caspian kutum (*Rutilus frisii kutum*) collected from the Siah Darvishan River, Guilan Province, Iran, during the summer of 2016 was investigated using varian atomic absorption spectroscopy. The mean concentrations of the metals ($\mu\text{g g}^{-1}$ d.w.) ranged as follows: Zn (34.87–36.12), Fe (29.15–33.34), Cu (7.57–8.84), Mn (1.89–2.23), As (0.80–0.87), Pb (0.73–0.88), Se (0.68–0.80), Cd (0.25–0.28), Ni (0.27–0.32), Cr (0.15–0.16), and Hg (0.08–0.09). According to the results, the mean concentrations of all elements in the muscle tissue of Caspian kutum were below the threshold limits recommended by FAO/WHO international standards, except for Mn (2.045 ± 0.169), As (0.840 ± 0.038), Pb (0.805 ± 0.077), and Cd (0.265 ± 0.011). Overall, the results indicate that the consumption of Caspian kutum from the Siah Darvishan River does not pose a significant health risk to consumers. Nevertheless, continuous environmental monitoring and regular assessment of metal contamination are essential to ensure long-term ecosystem health and food safety.

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Introduction

The Caspian kutum (*Rutilus frisii kutum*) is a semi-anadromous species belonging to the family *Cyprinidae*, which includes around 3,000 species (Ghasemi *et al.*, 2014). This unique species inhabits the Caspian Sea, forming part of the Arctic ichthyofauna that entered the sea after the last glacial period approximately 10–12 thousand years ago and gradually acquired the native characteristics of the Caspian ecosystem (Gussev *et al.*, 1993; Coad, 1998). The species has a lifespan of about 9–10 years, with maximum recorded length and weight of approximately 72 cm and 6 kg, respectively (Kotlik *et al.*, 2008). It is distributed along the Iranian coast of the Caspian Sea and migrates to southern rivers during the spawning season, particularly for feeding and overwintering (Yousefian and Mosavi, 2008; Bandpei *et al.*, 2009; Esmaili and Gholami, 2011). Although it also occurs in the Volga and Ural rivers, approximately 90% of the total population is found in Iranian waters (Naseka and Bogutskaya, 2009; Ustarbekov, 2011).

The Caspian kutum is one of the most economically and nutritionally valuable fish species in Iran due to its high-quality flesh and favorable taste (Ahmadian *et al.*, 2015). Consequently, fish consumption is common in the coastal regions and southern river basins of the Caspian Sea, particularly in Guilan Province (Coad, 1998). Although the International Union for Conservation of Nature (IUCN) currently classifies the species as “Least Concern” (Freyhof and Kottelat, 2008; Abdolhay *et al.*, 2012), its populations have declined in recent years as a result of overfishing, pollution of aquatic environments, and changes in water

chemistry, especially from heavy metal contamination (Esmaili *et al.*, 2015).

The Siah Darvishan River is one of the key habitats of the Caspian kutum in the southern Caspian basin and serves as an important tributary to the Anzali Wetland. Originating from the highlands of Masal, Fouman, Gasht-e Rudkhan, Nazar Alat, Heydar Alat, and Ghale Rudkhan, the river covers a watershed area of approximately 290.5 km², with a main channel length of 45 km, an average slope of 3.2%, and an annual discharge of 6.37 m³/s (Ghaderi *et al.*, 2001). However, over recent decades, extensive human activities—including agriculture, livestock farming, and the discharge of domestic and industrial wastewater—have caused substantial fluctuations in the river’s chemical composition (Ghafouri *et al.*, 2010).

Trace elements such as Zn, Fe, and Cu are essential micronutrients required for physiological and reproductive functions in aquatic organisms. However, elevated concentrations of these metals in aquatic environments can induce toxicity due to their bioaccumulation in vital tissues such as muscle, liver, kidney, and gills (Järup, 2003; Castro-González and Méndez-Armenta, 2008). Therefore, assessing the bioaccumulation of heavy metals in aquatic organisms is crucial for understanding environmental contamination levels. Fish are particularly valuable bioindicators due to their ecological relevance and capacity to reflect pollutant concentrations in aquatic ecosystems (Authman *et al.*, 2015; Khanipour *et al.*, 2018; Łuczyńska *et al.*, 2018; Roomiani *et al.*, 2018).

Industrialization in riparian zones has intensified the discharge of contaminants

into rivers (Islam *et al.*, 2015). Given the high consumption of fish and the role of muscle tissue as a major entry point for trace elements into the food chain—leading to biomagnification and potential transfer to humans (Bosch *et al.*, 2016)—monitoring metal bioaccumulation in fish muscle is essential. Previous studies have examined heavy metal accumulation in *R. frisii kutum* (Varedi *et al.*, 2012) and other cyprinid species such as *Cyprinus carpio* (Siraj *et al.*, 2018; Sobhan Ardakani and Jafari, 2018; Solgi *et al.*, 2018).

The present study aimed to evaluate the composition and concentration of heavy metals in the edible muscle tissue of *R. frisii kutum* from the Siah Darvishan River, an

economically important species with high nutritional value. Furthermore, this research assessed the potential health risks associated with the consumption of this fish to provide a scientific basis for environmental and food safety management in the region.

Materials and methods

Preparation of samples

A total of 25 Caspian kutum (*Rutilus frisii kutum*) specimens of various sizes were collected randomly during the summer of 2016 from different stations along the Siah Darvishan River (latitude 37°25'N, longitude 49°30'E; elevation approximately -15 m, Sowmeh Sara, Guilan, Iran; Fig. 1).

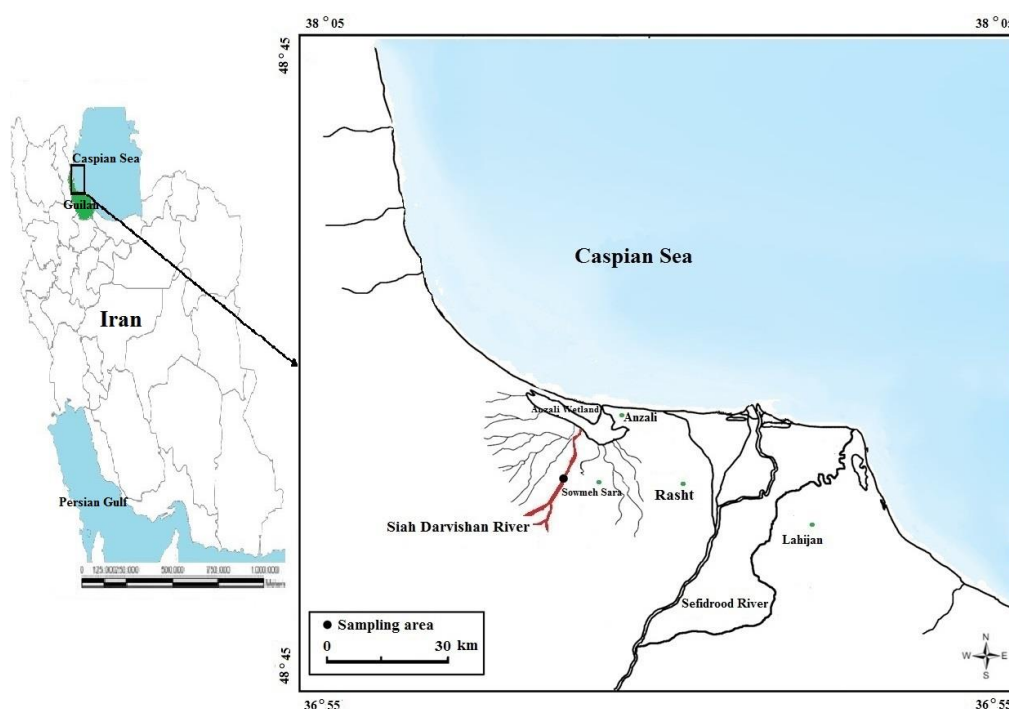


Figure 1: Map of Caspian kutum collection area in Siah Darvishan River (latitude 37°25'N, longitude 49°30'E; elevation approximately -15 m; Sowmeh Sara, Guilan province, Iran).

The fish were caught using a cast net (mesh size: 10 mm; diameter: 3 m) and transported in EPS foam containers with

crushed ice to the Fisheries Research Laboratory, Faculty of Natural Resources, University of Guilan. Upon arrival, the fish

were rinsed with double-distilled water to remove surface debris and potential external metal contaminants. Biometric measurements were then conducted: total length was recorded using a Mitutoyo caliper (503-501 series, Takatsu-ku, Japan; precision 0.1 mm), and body weight was measured with a Sartorius digital scale (CPA series, Göttingen, Germany; precision 0.01 g). Recorded data were tabulated for analysis. Subsequently, non-target organs—including skin, viscera, bones, and scales—were removed using sterile surgical blades, and the muscle tissue was carefully separated. Each muscle sample was labeled and freeze-dried using a Zirbus freeze-dryer (VaCo 5, Bad Grund, Germany) at -50°C for 9–10 hours. The dried tissues were ground into fine powder using a porcelain mortar. All laboratory glassware and mortars were prewashed with 10% HNO_3 and rinsed thoroughly with deionized water (Kimia Tehran Acid, Tehran, Iran) to prevent contamination (Authman *et al.*, 2015; Łuczyńska *et al.*, 2018).

Chemical digestion of tissues

Sample digestion was performed using the closed-vessel microwave-assisted acid digestion method. One gram of the dried, powdered muscle sample was accurately weighed using a Sartorius analytical balance (ED series, Göttingen, Germany; precision 0.001 g) and transferred into digestion tubes of the CEM microwave system (MARS 5, Matthews, USA). Each sample was treated with 9 mL of high-purity HNO_3 (65%, Merck Millipore, Billerica, USA) and digested at 180°C . After cooling to room temperature (23–

25°C), the digested solutions were filtered through Whatman® No. 40 filter papers (Sigma-Aldrich, St. Louis, USA) and diluted to 50 mL with ultrapure water in volumetric flasks. Samples were stored in pre-cleaned polypropylene containers (Nalgene®, series 2126, Rochester, USA) until metal analysis (Authman *et al.*, 2015; Łuczyńska *et al.*, 2018).

Evaluation of the heavy metal concentration

Metal concentrations were determined using atomic absorption spectroscopy (AAS, Varian, Palo Alto, USA). Zn, Mn, Cu, and Fe were measured using flame AAS (acetylene–air method, Varian 280 FS), whereas Pb, Cd, Cr, As, Ni, and Se were analyzed by graphite furnace AAS (Varian 280Z/120GTA). Hg was quantified using cold vapor AAS (Varian VGA-77). Accuracy and precision were verified by the standard addition method using certified reference material (CRM) of mussel tissue (SRM® 2976, NIST®, Gaithersburg, USA). All analyses were performed in triplicate (mean $\pm$ standard deviation), and recovery rates were calculated to assess analytical performance (Authman *et al.*, 2015; Łuczyńska *et al.*, 2018).

Statistical analysis

Data normality was tested using the Kolmogorov–Smirnov test. Differences in mean metal concentrations were evaluated by one-way ANOVA, followed by Tukey's post-hoc test to identify statistically distinct groups at the 95% confidence level ($p < 0.05$). Comparisons with international permissible limits (FAO/WHO, 1993) were

conducted using a one-sample t-test. Statistical analyses were performed using SPSS v22 (IBM, North Castle, USA), and results were expressed as mean $\pm$ standard deviation ($\mu\text{g g}^{-1}$ d.w.). Graphs and tables were generated using Microsoft Excel 2013 (Redmond, USA).

Results

Biometrics of the samples

A total of 25 Caspian kutum (*Rutilus frisii kutum*) specimens collected from the Siah Darvishan River were subjected to biometric analysis. The mean total length and weight were 42.80 ± 3.61 cm and 851.37 ± 120.21 g, respectively (Table 1).

Table 1: Biometric measurements of Caspian kutum collected from Siah Darvishan River.

Variables	Number	Average	Std. deviation	Minimum	Maximum
Total length (cm)	25	42.80	3.61	35.71	50.62
Total weight (g)	25	851.37	120.21	647.93	1132.79

Evaluation of the accuracy of the metal extraction method

To verify analytical accuracy, the certified reference material SRM[®] 2976 (Mussel Tissue, NIST[®]) was analyzed alongside the samples using the standard addition method. The measured concentrations of metals were within 85–103% of the

certified values (Table 2), with Zn and Hg showing the highest and lowest recovery rates, respectively. These results confirm that the applied digestion and analytical methods were reliable and suitable for determining metal concentrations in fish muscle tissue.

Table 2: Comparison of certified and measured heavy metal Concentrations ($\mu\text{g g}^{-1}$ d.w.) in standard reference material SRM[®] 2976 (Mussel Tissue).

Element	Operating conditions		SRM [®] 2976 (Mussel Tissue)		
	Wavelength (nm)	Slit width (nm)	Certified values $\mu\text{g g}^{-1}$	Measured values $\mu\text{g g}^{-1}$	Recovery (%)
As	188.98	0.5	13.3 ± 1.8	11.61 ± 1.52	90
Cd	214.44	0.7	0.82 ± 0.16	0.70 ± 0.21	86
Cr	267.70	0.2	0.50 ± 0.17	0.44 ± 0.21	89
Cu	327.40	0.7	4.02 ± 0.33	4.21 ± 0.11	97
Fe	259.94	0.2	171.52 ± 4.9	158.52 ± 6.74	92
Hg	253.71	0.5	0.061 ± 0.0036	0.052 ± 0.007	85
Mn	257.61	0.2	33 ± 2	31.57 ± 0.81	95
Ni	231.60	0.2	0.93 ± 0.12	0.97 ± 0.06	102
Pb	220.35	0.7	1.19 ± 0.18	1.07 ± 0.24	91
Se	196.03	1.0	1.80 ± 0.15	1.61 ± 0.09	88
Zn	213.90	0.7	137 ± 13	141.54 ± 8.62	103

Accumulation of the heavy metals in the muscle tissue

The concentrations of heavy metals in the muscle tissue of Caspian kutum are summarized in Table 3. The mean ($\pm$ SD) concentrations ($\mu\text{g g}^{-1}$ d.w.) were as follows: Zn (35.54 ± 0.63), Fe (31.20 ± 1.96),

Cu (7.85 ± 0.28), Mn (2.05 ± 0.17), As (0.84 ± 0.04), Pb (0.81 ± 0.08), Se (0.75 ± 0.06), Ni (0.29 ± 0.02), Cd (0.27 ± 0.01), Cr (0.15 ± 0.00), and Hg (0.08 ± 0.00). Among the analyzed elements, Zn exhibited the highest mean concentration ($36.12 \mu\text{g g}^{-1}$ d.w.), while Hg

showed the lowest ($0.08 \mu\text{g g}^{-1} \text{ d.w.}$). The average accumulation order in the muscle tissue followed the sequence: $\text{Zn} > \text{Fe} > \text{Cu} > \text{Mn} > \text{As} > \text{Pb} > \text{Se} > \text{Ni} > \text{Cd} > \text{Cr} > \text{Hg}$.

Table 3: Comparison of heavy metals concentrations ($\mu\text{g g}^{-1} \text{ d.w.}$) in muscle tissue of Caspian kutum with the threshold limit value of the international standard.

Element	Concentration ($\mu\text{g g}^{-1} \text{ d.w.}$)		R.S.D. (%)	Range ($\mu\text{g g}^{-1} \text{ d.w.}$)		International standard (FAO/WHO)
	Mean	Std. deviation		Minimum	Maximum	
As	0.840	0.038	4.59	0.80	0.87	0.05
Cd	0.265	0.011	4.00	0.25	0.28	0.2
Cr	0.153	0.004	2.85	0.15	0.16	0.2
Cu	7.849	0.284	3.63	7.57	8.84	30
Fe	31.195	1.957	6.28	29.15	33.34	100
Hg	0.084	0.004	5.19	0.08	0.09	0.5
Mn	2.045	0.169	8.27	1.89	2.23	0.05
Ni	0.293	0.021	7.20	0.27	0.32	0.4
Pb	0.805	0.077	9.63	0.73	0.88	0.5
Se	0.745	0.061	8.19	0.68	0.80	1
Zn	35.536	0.625	1.76	34.87	36.12	1000

When compared with the permissible limits established by the Food and Agriculture Organization and World Health Organization (FAO/WHO, 1993), the concentrations of As, Cd, Pb, and Mn in the edible muscle tissue exceeded the recommended thresholds. In contrast, all other metals were within the acceptable limits for human consumption.

Discussions

Heavy metals are persistent and resistant to degradation, attracting significant attention due to their potential for bioaccumulation in aquatic organisms, including fish tissues such as muscle, bone, and skin. The increasing input of these elements into aquatic ecosystems as a result of industrialization has become one of the major environmental challenges in recent decades, since the rate of metal entry into aquatic systems often exceeds the natural purification capacity of these environments

(Ahalya *et al.*, 2003). Because most fish species cannot escape contaminated habitats, they are particularly vulnerable to heavy metal exposure (Yi *et al.*, 2011; Mance, 2012). Over time, these elements can be transferred through the food web, undergoing biomagnification as they move to higher trophic levels (Mirzajani *et al.*, 2016). Consequently, the consumption of contaminated fish poses potential risks to human health, highlighting the importance of evaluating metal accumulation in edible fish tissues (Rai, 2009).

Muscle tissue, the most frequently consumed part of fish, has been the focus of numerous studies investigating heavy metal accumulation (Carvalho *et al.*, 2005; Jia *et al.*, 2017). Determining the concentration of these elements in muscle is critical because it reflects both the contamination level of the habitat and the degree of exposure for consumers (Azaman *et al.*, 2015). It also serves as a reliable biomarker,

indicating the extent of metal pollution in aquatic environments (Birungi *et al.*, 2007). As one of the main edible portions of fish (Venugopal and Shahidi, 1996), muscle tissue was selected as the target for assessment in this study.

The observed variations in metal accumulation among fish species and habitats may result from environmental conditions and species-specific physiological factors such as differences in metallothionein and lysozyme production. These proteins play key roles in detoxifying harmful metals at low pollution levels, thereby influencing interspecific variation in metal bioaccumulation (Moore and Ramamoorthy, 2012).

Table 4 compares the sequence of metal accumulation in the present study with results from previous investigations on various *Cyprinidae* species. In this study, Zn and Fe exhibited the highest concentrations in muscle tissue, consistent with findings from Canbek *et al.* (2007), who reported Zn and Fe levels of 8.92 and 1.8 $\mu\text{g g}^{-1}$ in *Capoeta capoeta* from the Porsuk River, Turkey. Similarly, Uysal *et al.* (2009) found Zn and Fe concentrations of 16.31 and 11.32 $\mu\text{g g}^{-1}$, respectively, in *Squalius cephalus* from Enne Dam Lake (Turkey), and Xie *et al.* (2010) recorded corresponding values of 65.3 and 29.5 $\mu\text{g g}^{-1}$ in *Cyprinus carpio* from the Pearl River Delta, China. Comparable results were also reported by Kaçar *et al.* (2017) for *Barbus grypus* from Batman Lake (Turkey), with Zn and Fe concentrations of 5.35 and 3.32 $\mu\text{g g}^{-1}$, respectively. These results support the present findings, where Zn and Fe concentrations were 35.54 and 31.19 $\mu\text{g g}^{-1}$, respectively. However, other studies reported different accumulation sequences. For instance, Oymak *et al.* (2009) observed Zn

and Fe values of 10.94 and 3.98 $\mu\text{g g}^{-1}$, respectively, in *Barbus grypus* from Atatürk Dam Lake (Turkey), while Uysal *et al.* (2009) found Zn and Fe concentrations of 24.88 and 21.10 $\mu\text{g g}^{-1}$ in *Alburnus alburnus* from Enne Dam Lake (Turkey), showing Fe > Zn.

The higher Zn concentration in the present study may be related to its crucial role in metabolic processes at trace levels, including enzymatic catalysis, mitigation of Cd toxicity, and its gradual release from tissues (Leung *et al.*, 2014). Fe, while essential for hemoglobin formation and cellular respiration, is also one of the most abundant industrial metals and can enter aquatic systems through anthropogenic sources. Although Fe constitutes approximately 0.005% of total fish tissue mass, its deficiency may lead to severe microcytic and hypochromic anemia, reducing the size and volume of red blood cells (Uysal *et al.*, 2008; Dhanakumar *et al.*, 2015; Rakocevic *et al.*, 2018).

The bioaccumulation sequence of the essential metals Cu and Mn observed in this study (Cu: 7.85 $\mu\text{g g}^{-1}$; Mn: 2.05 $\mu\text{g g}^{-1}$) followed Zn and Fe, consistent with Uysal *et al.* (2009), who reported Cu and Mn concentrations of 1.51 and 0.48 $\mu\text{g g}^{-1}$ in *Carassius carassius*, and Köse *et al.* (2015), who recorded 0.945 and 0.91 $\mu\text{g g}^{-1}$ in *Capoeta sieboldii*. However, the results differ from Farhadi and Yavari (2013), who found distinct accumulation patterns in *Capoeta damascina* from the Sezar River (Iran). Cu plays a critical role in metabolic activities, pigment formation, and enzyme function, while Mn and Se contribute to muscle structure and coenzyme activity, explaining their relatively high accumulation in muscle tissue (Tepe *et al.*, 2008; Tuzen, 2009; Spanopoulos-Zarco *et al.*, 2017).

Table 4: Comparison between the orders of heavy metals accumulation in the muscle tissue of Caspian kutum with other Cyprinidae species from different parts of the world.

Species	Rank order	Sampling area	Reference
<i>Rutilus frisii kutum</i>	Zn>Se>Cu>Mn>Hg>Pb>Cd>Ni	Total southern coasts, Caspian Sea, Iran	(Anan <i>et al.</i> , 2005)
<i>Barbus plebejus</i>	Zn>Fe>Ni>Mn>Pb>Cu>Cd	Porsuk River, Eskişehir, Turkey	(Canbek <i>et al.</i> , 2007)
<i>Capoeta capoeta</i>	Zn>Fe>Ni>Mn>Cu>Pb>Cd	Porsuk River, Eskişehir, Turkey	(Canbek <i>et al.</i> , 2007)
<i>Barbus grypus</i>	Fe>Zn>Pb>Cu>Mn>Ni	Atatürk Dam, Adıyaman, Turkey	(Oymak <i>et al.</i> , 2009)
<i>Carassius carassius</i>	Zn>Fe>Cu>Mn>Cd	Enne Dam, Kütahya, Turkey	(Uysal <i>et al.</i> , 2009)
<i>Chondrostoma nasus</i>	Fe>Zn>Ni>Cu>Cd	Enne Dam, Kütahya, Turkey	(Uysal <i>et al.</i> , 2009)
<i>Squalius cephalus</i>	Zn>Fe>Ni>Cu>Mn>Cd	Enne Dam, Kütahya, Turkey	(Uysal <i>et al.</i> , 2009)
<i>Alburnus alburnus</i>	Fe>Zn>Cd	Enne Dam, Kütahya, Turkey	(Uysal <i>et al.</i> , 2009)
<i>Cyprinus carpio</i>	Zn>Fe>Cu>Pb>As>Mn>Cd>Hg	Pearl River Delta, Guangdong, China	(Xie <i>et al.</i> , 2010)
<i>Barbus grypus</i>	Zn>Pb>Ni>Cu>Mn>Cd	Shadegan Wetland, Khuzestan, Iran	(Alhashemi <i>et al.</i> , 2012)
<i>Barbus sharpeyi</i>	Zn>Pb>Cu>Mn>Ni>Cd	Shadegan Wetland, Khuzestan, Iran	(Alhashemi <i>et al.</i> , 2012)
<i>Cyprinus carpio</i>	Zn>Pb>Cu>Ni>Mn>Cd	Shadegan Wetland, Khuzestan, Iran	(Alhashemi <i>et al.</i> , 2012)
<i>Rutilus frisii kutum</i>	Zn>Ni>Cu>Pb>Hg>Cd	Farah Abad Sari coasts, Caspian Sea, Iran	(Varedi <i>et al.</i> , 2012)
<i>Capoeta damascina</i>	Fe>Zn>Pb>Ni>Cu>Cd	Sezar River, Lorestan, Iran	(Farhadi and Yavari, 2013)
<i>Cyprinus carpio</i>	Zn>Cu>Hg>Cd>Ni>Pb	Total southern coasts, Caspian Sea, Iran	(Nasrollahzadeh <i>et al.</i> , 2013)
<i>Ctenopharyngodon idella</i>	Mn>Cu>Pb>As>Cd	Lhasa, Tibetan Autonomous Region, China	(Jiang <i>et al.</i> , 2014)
<i>Cyprinus carpio</i>	Mn>Cu>Pb>As>Cd	Lhasa, Tibetan Autonomous Region, China	(Jiang <i>et al.</i> , 2014)
<i>Barbus tauricus</i>	Zn>Ni>Cu>Mn	Porsuk River, Eskişehir, Turkey	(Köse <i>et al.</i> , 2015)
<i>Capoeta baliki</i>	Zn>Ni>Cu>Mn	Porsuk River, Eskişehir, Turkey	(Köse <i>et al.</i> , 2015)
<i>Carassius gibelio</i>	Zn>Ni>Cu>Mn>As>Pb	Porsuk River, Eskişehir, Turkey	(Köse <i>et al.</i> , 2015)
<i>Capoeta sieboldii</i>	Zn>Cu>Mn>Ni	Porsuk River, Eskişehir, Turkey	(Köse <i>et al.</i> , 2015)
<i>Barbus grypus</i>	Zn>Fe>Mn>Cu	Batman Dam, Diyarbakır, Turkey	(Kaçar <i>et al.</i> , 2017)
<i>Squalius fellowesii</i>	Zn>Cu>Mn>Pb>Cd	Tersakan River, Turkey	(Şaşı <i>et al.</i> , 2017)
<i>Cyprinus carpio</i>	Zn>Fe>Ni>Cu>Pb>Mn>Cd>Hg	Kabul River, Khyber Pakhtunkhwa, Pakistan	(Siraj <i>et al.</i> , 2018)
<i>Cyprinus carpio</i>	Cu>Mn>Pb>Cd	Nowshahr coasts, Caspian Sea, Iran	(Solgi <i>et al.</i> , 2018)
<i>Rutilus frisii kutum</i>	Zn>Fe>Cu>Mn>As>Pb>Se>Ni>Cd>Cr>Hg	Siah Darvishan River, Sowmeh Sara, Guilan, Iran	Present study

In the present study, Zn, Fe, and Cu concentrations were below the international standard limits, whereas Mn exceeded the recommended values.

The accumulation sequence of the non-essential and toxic metals (As, Pb, Ni, Cd, Cr, and Hg) was also in agreement with several previous studies. Anan *et al.* (2005)

reported similar results for *R. frisii kutum* collected from the southern Caspian Sea. Comparable trends were observed by Xie *et al.* (2010) in *C. carpio*, Varedi *et al.* (2012) in Caspian kutum from Farah Abad (Sari, Iran), and Siraj *et al.* (2018) in *C. carpio* from the Kabul River (Pakistan). In these studies, toxic metals appeared in lower concentrations than essential elements, consistent with the present results. The relatively low levels of toxic metals in muscle may be attributed to species-specific differences within *Cyprinidae* and the limited biological requirement of fish for these elements (Makedonski *et al.*, 2017).

Anthropogenic sources—including industrial effluents, agricultural runoff, animal husbandry, medical waste, petroleum products, pesticides, and domestic sewage—are the primary contributors to these contaminants (Duruibe *et al.*, 2007; Alloway, 2013; Mance, 2012). High concentrations of such metals can severely affect aquatic organisms by disrupting osmoregulatory and reproductive processes, damaging vital tissues, and even causing mortality (Mance, 2012).

Conclusion

According to the results of this study, the concentrations of all analyzed metals in the edible muscle tissue of Caspian kutum (*Rutilus frisii kutum*) from the Siah Darvishan River, Guilan Province, were below the permissible limits recommended by FAO/WHO standards, except for Mn, Pb, As, and Cd. Although the detected levels of these elements were slightly above the international thresholds, they indicate a

potential risk associated with the input of various pollutants—such as residues of agricultural chemicals, herbicides, pesticides, and petroleum derivatives—into the habitat of this species. These findings highlight the necessity of continuous environmental monitoring and comprehensive assessment of contamination sources to prevent the further accumulation of toxic metals in aquatic ecosystems. Regular surveillance and the identification of pollutant entry pathways are essential for maintaining ecological health and ensuring the safety of fish species consumed by humans. Implementing effective management strategies to control industrial, agricultural, and domestic discharges can play a critical role in preserving the environmental integrity of the Siah Darvishan River and the broader Caspian basin.

Conflicts of interest

The authors have declared no conflict of interest.

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