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Trace Metals Distribution in *Anguilla anguilla* (Linnaeus, 1758) Tissues, Sediments and Water from Tersakan Stream (Mugla), Turkey

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Abstract

In this study bio-accumulation of trace metals (Co, Cr, Cu, Fe, Mn, Ni and Zn) were determined in water, sediment and European eel (Anguilla anguilla) tissues from Tersakan Stream (Mugla) between September 2011- August 2012. Totally 160 eels have been caught and investigated seasonally. To determine the heavy metal levels in eel tissue samples from gill, liver and muscle of eel were obtained and analyzed in ICP-AES after dried in microwave unit. The mean metal amounts of all investigated tissues were also statistically analysed and discussed results. According to the findings; pH value varied between 7, 55-8, 06. In water; Co were not determined in any seasons but Cr, Cu, Fe, Mn, Ni and Zn were present almost in the whole seasons while Fe level was highest. In sediment; Co, Cr, Cu, Fe, Mn, Ni and Zn were present in all seasons while Fe was highest. In the gill, liver and muscle tissues mean highest accumulation was observed at Fe with 66,08 μg/g, 206,63 and 31,21 μg/g levels respectively. The accumulation of investigated heavy metals in muscle tissue was in order as; Zn> Fe> Mn> Cr> Cu> Ni> Co. In gill tissue the order of accumulation of the investigated heavy metals was as; Fe> Zn> Mn> Cu> Cr> Ni> Co. In liver tissue the order of the accumulation of the investigated heavy metals was; Fe> Zn> Cu> Mn> Cr> Ni> Co. In generally, it was found that the metal accumulation levels of edible portions (muscle) were lower than other tissues. The trace metal concentrations in the edible tissue (muscle) of Anguilla anguilla was compared with the tolerable national and international values in fish. The result obtained showed that the trace metal concentrations in edible tissue did not exceed the WHO/FAO, TFC, Range of International Standards, EPA and IAEA-407guidelines. The results of this study revealed that consuming the Anguilla anguilla from the Tersakan Stream may not have harmful effects.

Keywords: Anguilla anguilla; Trace Metal; Food Safety; Mugla

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Abbreviations: WHO**:** World Health Organization; FAO: Food and Agriculture Organization; CIW: Criterions of the Irrigation Water; PEC: probable effect concentration; SEL: levels and severe effect levels; TWPCR: Turkish Water Pollution Control Regulations; SE: Standart Error.

Introduction

The pollution ofthe aquatic environment with heavy metals has become a world wide problem during recent years, because they are indestructible and most of them have toxic effects on organisms [1]. Among pollutants, trace metals are great concern because of their toxicity, persistence and prevalence. The subsequent problems have received considerable scientific attention for many years. Various abnormalities have been observed in wildlife [2,3] and even in humans [4-6], have been proved to be linked with the high degree of exposure to toxic metal contaminanats. Therefore, enviromental monitoring activities play an important role in assessing and predicting the impact of contaminants to environment. As a consequence, pollutants discharged in aquatic environment are likely to accumulate in fish and represent a potential risk not only to the fish, but also to other fish consumers, particularly humans. Catadromous eels, widely distributes throughout the world, are one of the top predator in freshwater ecosystem [7]. Due to the long-life cycle and the spesific biological and ecological features of anguillid eels, they are vulnerable to adverse impacts from nature and human activities [8-10]. Therefore, the eel populations have declined dramatically in recent years and causes are attributed to over-fishing, construction, climate change, other enviromental factors, especially enviromental pollution [11-13]. Hence, anguillid eels has been widely used as bioindicator for environmental monitoring to assess the aquatic system quality in many countries [14-16].

The results of monitoring work in field by using eels as bioindicator are, therefore, rather useful to assess the health of aquatic system, fish consumers and risk for human consumption [17-19]. However, the use of European eel *A. anguilla* as bio-indicator or bio-monitor. Has mostly been done in EU countries for decades [14-16]. Moreover, less information is available for the use of European eel *A. anguilla* as bio-indicator though European eel are abundant in aquatic system of Turkey countries including Mugla. The aim of this study was to determine thetrace metals concentrations (Co, Cr, Cu, Fe, Mn, Ni and Zn) in water, sediments and in muscle, liver and gill of fish species (*A. anguilla*) from the Tersakan Stream (Mugla), since this fish is an important component of the human diet in this zone. The results obtained from this study would provide information for background levels of metals in the water, sediment and fish species of the river, contributing to the effective monitoring of both environmental quality and the health of the organisms inhabiting the lake ecosystem.

Materials and Methods

Study Area

Dalaman- Tersakan Stream, (36°45’51”N, 28°49’20”E) is located in province Mugla in the southwest of Turkey (Figure 1). The sampling site (Tersakan Stream) is a temperate stream which is impacted by unpredictable environmental conditions associated with a Mediterranean climate. Its length is 30 km and this stream has temporal and spatial water flow variations throughout the water course (48-780 m^3/s). The lower section of the stream was channelized by local authorities to prevent seasonal floods. The stream flows into Mediterranean Sea [20] (Figure 1). Vegetation is usually abundant throughout the stream banks and depth varies between 0.5-2m. The sampling site was characterized by muddy substrate, limited vegetation and slow flow velocity. It had recently been affected by heavy floods, which occur seasonally because of high annual precipitation. There are eight known species inhabiting the stream of which the most abundant are *Mugil cephalus* (Linnaeus, 1758), *Leuciscus cephalus* (Linnaeus, 1758), *Gambusia affinis* (Baird and Girard, 1853) and *Anguilla anguilla* (Linnaeus, 1758) [20].

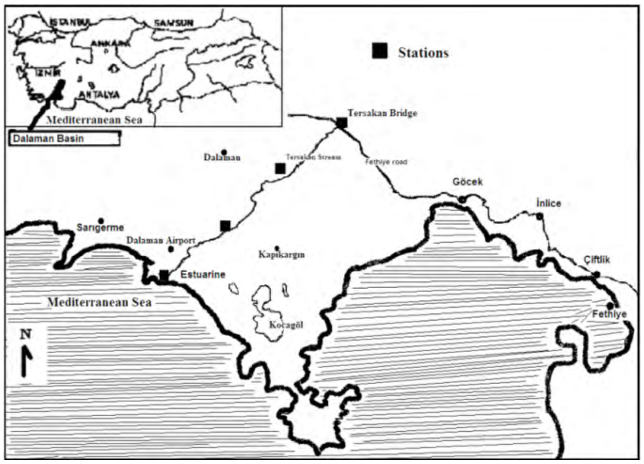

Figure 1: Sampling area of Anguilla anguilla specimens in Mugla province (Turkey).

Sampling and analysis

Water and fish samples were collected 4 times (from September 2011 to August 2012) for every 3 months at four stations in the Tersakan Stream. The sampling bottles were pre-conditioned with 5% nitric acid and later rinsed thoroughly with distilled de-ionized water. At each sampling site, the polyethylene sampling bottles were rinsed at least three times before sampling was done. Pre-cleaned polyethylene sampling bottles were immersed about 10 cm below the water surface. About 0.5L of the water samples were taken at each sampling site. Samples were acidified with 10% HNO_3, placed inan ice bath and brought to the laboratory. The samples were filtered through a 0.45 µm micropore membrane filter and kept at 4°C until analysis. The samples were analyzed directly analyses were performed according to APHA, [21] sediment samples were collected using grab sampler from four sites. Samples were transported to the laboratory and air-dried in the laboratory at room temperature. Once air-dried, sediment samples were powdered and passed through 160 µm sieve. The samples packed in polyethylene bags and stored below -20°C prior to analysis. Sediments samples were weighed placed into the digestion bombsn with 10mL of HNO_3/HCl (1:3 v/v) and digested in a microwave digestion system. Sediments analysis was carried out according to the procedure described earlier [22]. A total of 160 samples of eel, (*A. anguilla*) were captured by backpack electrofishing with a battery-powered unit (550 V, 5–100 Hz) at four different stations along the Tersakan Stream in the period of September 2011to August 2012. Fish samples were immidiately transported to the laboratory in a thermos flask with ice. The fish species selected were based on their abundance along the river water course and frequency in use as food by the inhabitants of these areas. The main characteristics of the fish species are given in Table 1.

Age	Number of species (N)	Total Length (TL) (mm) X±SE	Weight (W) (g) X±SE
3	61	216,70±39,234	42,02±8,653
4	53	279,45±56,378	49,12±4,468
5	26	352,65±51,573	54,53±7,451
6	12	443,58±77,774	58,20±9,524
7	8	571,37±64,272	63,93±9,776
SE: Standart Error			

Table 1: Main characteristics of the *Anguilla anguilla* from Terskan Stream.

The samples were carefully cut opened using a plastic knife in order to remove the muscle, and freze dried and pulverized into a uniform particle size prior to analysis. The small sized particles were subjected to acid digestion using nitric acid. Approximately 5 g of muscle from each sample were dissected, washed with deionized water, weighed, packed in polyethylene bags, and stored at -20°C prior to analysis. The tissue samples were digested with concentrated nitric acid. Dissected samples were transferred to a 100 mL teflon beaker. Thereafter, 10 mL ultrapure concentrated HNO_3 (Merck) was added, and the sample heated at (100, 150,210 and 280°C on a hot plate for 0.5, 0.5, 0.5 and 2 hours) with a DK-20 Heating Digester respectively. Two mL of 1 N HNO_3 was added to the residue, and the solution continuously evaporated on the hot plate, until it was digested in every sample. After cooling, a further 10 mL of 1 N HNO_3 was added. The solution was transferred, diluted and filtered through 0.45 µm nitrocellulose membrane fitler [23].

All samples were analysed in duplicates for Co, Cr, Cu, Fe, Mn, Ni and Zn by ICP/AES (Optima 2000-Perkin Elmer), which is a fast multi-element technique with a dynamic linear range and moderate-low detection limits [24]. Detection limits are given in Table 2. The detection limit is defined as the lowest analytical signal to be distinguished qualitatively at a specified confidence level from the background signal [25]. The accuracy of analytical procedure was checked by analyzing the standard reference materials (Water: SRM-143d, National Institute of Standards and Technology; Fish: DORM–2, National Research Council). Recovery rates ranged from 79 to 96% for all the elements analysed.

Element	Wavelength (nm)	Detection limit (mg/L)
Co	228.616	0.0070
Cr	267.716	0.0071
Cu	327.393	0.0097
Fe	238.204	0.0046
Mn	257.610	0.0014
Ni	231.604	0.0150
Zn	202.548	0.0040

Table 2: Spectral lines used in emission measurements and detection limit for the elements measured by using ICP-AES.

Statistically analysis

Statistical analysis of data was carried out using SPSS 14.0 statistical package program. One-Way Analysis of Variance (ANOVA)and Duncan's Multiple Comparison Test were used to compare the data among seasons at the level of 0,05. Relationship of metal levels in muscle tissue with metal levels in water samples and fish biometric parmeters was performed using the Pearson's Correlation Analysis.

Results and Discussions

The mean concentrations of trace metals inwater samples for all seasons were presented in Table 3. According to the findings; In water; Co were not determined in any seasons but Cr, Cu, Fe, Mn, Ni and Zn were present almost in the whole seasons. It was determined that Fe was the highest metal in water. Iron is one of the most abundant elements in the earth's crust [26,27]. The levels of trace metal exhibited seasonal fluctuations. The highest levels in the water were found during summer and winter, while the lowest values occurred during autumn and spring. These seasonal variations may be due to the fluctuation of the amount of agricultural drainage water, sewage effluents, and industrial wastes discharged into the Tersakan Stream. Industrial discharge and agricultural runoffs forms of human activities that are released into river caused heavy metal pollution in water seasonally, and that these persistent pollutants accumulated more readily in fish during the dry season [28,29].

Trace metals	Autumn Mean ±SE (range)	Winter Mean ±SE (range)	Spring Mean ±SE (range)	Summer Mean ±SE (range)	CIW (Criterions of the Irrigation Water [30])	The Classes of the Water Quality according to TWPCR [31]			
						I	II	III	IV
Co	ND	ND	ND	ND	0.05	10	20	200	> 200
Cr	0,02±0,01^{c} (0,01–0,04)	0,39±0,21^{b} (0,14–0,66)	0,01±0,01^{c} (0,01–0,03)	0,72±0,21^{a} (0,48–0,94)	0.1	20	50	200	> 200
Cu	0,68±0,09^{b} (0,56–0,79)	0,9±0,28^{b} (0,55–1,24)	0,18±0,09^{b} (0,12–0,25)	2,37±0,07^{a} (1,18–3,74)	0.2	20	50	200	> 200
Fe	10,79±2,12^{c} (6,58–16,5)	30,75±4,43 ^{b} (23,56–38,7)	19,74±5,46^{c} (12,45–31,13)	49,2±5,21^{a} (42,4–59,3)	5	300	1000	5000	> 5000
Mn	0,4±0,11^{c} (0,26–0,53)	0,8±0,14^{b} (0,66–0,98)	0,18±0,06^{c} (0,11–0,25)	1,16±0,12^{a} (0,98–1,45)	0.2	100	500	3000	> 3000
Ni	0,19±0,11^{b} (0,09–0,34)	0,45±0,08^{b} (0,39–0,58)	0,17±0,07^{b} (0,1–0,26)	1,12±0,24^{a} (0,73–1,79)	0.2	20	50	200	> 200
Zn	0,99±0,03^{c} (0,96–1,03)	2,02±0,07^{b} (1,96–2,13)	0,42±0,11^{c} (0,23–0,69)	3,72±0,47^{a} (2,99–4,5)	2	200	500	2000	> 2000

I: High quality water, II: weakly polluted water, III: Polluted water, IV: High polluted water
[a,b,c]: The different word in each row is statically different ($p<0,05$). TWPRC: Turkish Water Pollution Control

Table 3: The mean trace metals concentrations in the Tersakan stream's water and comparison with guidelines (µg/L). Regulation [31] ND: Not Detectable.

Because of evaporation resulting from increased temperatures during the dry season, heavy metal concentrations in the water were generally higher compared to those in the wet season. Lower heavy metal concentrations in the wet season are attributable to water dilution caused by moderate to heavy rainfall during the rainy season [32]. Ali and Abdel-Satar [33] attributed the increase of metal concentrations in the water during hot seasons (summer) to the release of heavy metals from the sediment to the over lying water under the effect of both high temperature and a fermentation process resulting from the decomposition of organic matter. Also, the seasonal variations of metals in water have been reported by Asaolu, et al. [34], Calıskan [35], Tekin-Ozan and Kir, [36] and Abdel-Baky, et al. [37]. Our results were similar with their studies. The Cr, Cu, Fe, Mn, Ni and Zn concentrations in Tersakan Stream's water samples in the four seasons were compared with international standards. The values of Criterions of the Irrigation Water (CIW) [30], these values and our results were compared and it was found as high trace metal concentrations almost in whole seasons. The obtained results showed that the concentrations of Cu, Fe, Mn and Ni in water exceeded the values of CIW [30] in whole seasons. However the concentrations of Cr and Zn in water only exceeded the values of CIW [30] in winter and summer seasons. In this case, the water taken from Tersakan Stream is not proper for irrigation. Trace metal concentrations in water samples were determined to be class I according to Turkish Water Pollution Control Regulations (TWPCR) [38].

According to the our results it was identified that Tersakan Stream has good quality water but, becomes under pressure because of the pollution periodically. Especially theuse of fertilizers and pesticides in agriculture, domestic and urban sewage and industrial waste is believed to would increase the metals concentrations. There is no doubt that most waste generated due to human activities are discharged on landor in streams around rural areas which would be transported by runoff water into the Tersakan Stream during rains. Our statistical analysis showed that there was a significant different between trace metal concentrations in four seasons ($p<0.05$). The mean concentrations of trace metals in sediment samples for four seasons were presented in Table 4. The concentration of tracemetals were higher in the winter and summer seasons than those of the autumn and spring seasons. The result is similar to the observation of Asaolu, et al. [34] who studied the determination and seasonal variation of heavy metals inalgae and sediments in sewers from industrial area of Lagos state Nigeria and Adefemi, et al. [39] when studying seasonal variation in heavy metal distribution in the sediment of major dams in Ekiti state. Also, our results were similar with studies of Caliskan [35], tekin-Ozan and Kir [36] and Kir and Tumantozlu [40]. Of the heavy metal examined Fewas found to be the most abundant, the high content of Fe compared to other heavy metals in the sediment is expected because it has been reported by several workers, that it occurred at high levels in earth's crust [26,27].

A similar high level of Fecontent was reported by Bourhane-Eddine, et al. [41] in the evaluation of metal contamination in the surface sediment of the Quberra Lagoon, Algeria. The knowledge of the heavy metal concentrations in sediment could give vital information regarding their sources, distribution and degree of pollution, since sedimentation is one of the most important fluxes in aquatic systems [49]. The trace metal concentrations obtained from the sediment samples were compared with Sediment Quality Guideline which showed that Co, Cr, Fe, Mn and Ni concentrations exceeded the probable effect concentration (PEC) levels and severe effect levels (SEL), for except Cu and Zn. From the results obtained, it is clear that, the metals Co, Cr, Cu, Fe, Mn, Ni and Zn were accumulated in sediment at high concentration levels amounting to thousands times those accumulating in water. By comparing the accumulation of heavy metals in *A. anguilla* tissues, water and sediments, it can be concluded that the heavy metals are highly accumulated in sediments than water and *A. anguilla* tissues, since the sediments act as reservoir for all contaminants and dead organic matter descending from the ecosystem above. Similar findings were reported by other authors [42,43]. Inaddition to, this has been attributed to the fact that bottom sediment in an aquatic environment used to act as traps for most of the metals due to high level of organic substance. Many studies have reported that the maximum heavy metal accumulation in sediment [44-46].

Trace metals	Autumn	Winter	Spring	Summer	PEC	SEL
(μg/g)	(range)	(range)	(range)	(range)		
Co	1,12±0,26b	2,79±0,453 ab	2,07±0,373 b	3,95±0,437 a	-	-
	(0,56–2,05)	(1,04–4,02)	(1,36–3,03)	(2,94–5,96)		
Cr	239,77±17,45	242,65±19,46	201,3±28,12	284,4±38,56	111	110
	(187–273,9)	(202,7–274)	(149,8–262)	(183,1–390,3)		
Cu	16,07±0,12 ab	19,75±0,32 ab	14,39±1,45 b	21,54±4,1 a	149	110
	(15,8–16,24)	(18,79–20,98)	(11,04–16,31)	(11,56–26,43)		
Fe	1831,25±77,56	1933±96,55	1833±87,67	2043,5±69,89	-	75
	(1469–2136)	(1435–2154)	(1376–2226)	(1944–2164)		
Mn	359,55±19,78	403,47±51,71	324,8±15,32	428,67±17,45	-	-
	(321–414,9)	(233–520)	(258,3–362,9)	(388,8–469,5)		
Ni	416,77±26,67	427,5±34,72	375,1±45,15	453,5±21,56	48,6	75
	(338,2–500)	(303,3–492,8)	(279,1–425)	(409,5–508)		
Zn	110,77±39,1	167,1±47,68	54,27±15,54	171,1±59,56	459	820
	(28,29–246,5)	(98,53–295,9)	(31,2–78,08)	(77,17–327,4)		

Table 4: The mean Trace Metal Concentrations in The Tersakan Stream's Sediment and Comparision with Sediment Quality Guideline. (Mean±SE.,μg g^{-1} metal/ wet w).
[a,b,c]: The different word in each row is statically different ($p<0,05$)
SEL = Severe effect level, dry weight [47], PEC= Probable effect concentrations [48].

There was a significant difference ($p<0.05$) between concentrations of Co and Cu in spring and summer periods while there was no a significant difference ($p>0.05$) between concentrations of Cr, Fe, Mn, Ni and Zn according to seasons (Table 4). Mean concentrations of trace metals (Co, Cr, Cu, Fe, Mn, Ni and Zn) in muscle, gills and liver of *A. anguilla* from the Tersakan Stream is shown in Table 5. The highest concentrations were found in liver, except for Co, Mn and Ni which is found in gills to be highest. However, majorly muscle has the least concentration of the trace metals compared with gills and liver in the *A. anguilla* sampled. This is in agreement with previous study by Ishaq, et al. [49] which showed that muscle is not an active organ in the accumulation of heavy metals. The high level of metals in the liver may be due to the role of metals in physiological activities of organs. The liver tissue is highly active in the uptake and storage of heavy metals [50]. It is well known that large amount of metallothionein induction occurs in the liver tissue of fish and liver has also an important role in redistribution transformation of contaminants and also act as an active site of pathological effects induced by contaminants. The results are in accordance with the findings of Evans, et al. [51] and Shanker, et al. [52] on the trace element concentration in some fishes of fresh water ecosystem. Gill tissues play an important role in interface with the environment in performing its functions in gas exchange, ion regulation, balance and waste excretion while muscle is not an active tissue in bioaccumulation [53-55]. Gills, on the other hand, has been reported as metabolically active site and can accumulate heavy metals in higher level. Target organs such as liver and gills, have a tendency to accumulate heavy metals in high values, as shown in many species of fish in different areas [56-58]. Lower concentration of metals in muscles were observed during this study may be due to the fact that the absence of metals binding protein and also muscles was less active as compared with other tissues. It is generally accepted that muscle is not an organ in which metals accumulate [59]. Similar results were observed by Al Yousuf, et al. [60] in the Lethrinus lentjan. The above observation was also supported by Romeo, et al. [61]. Also similar results were reported from a number of fish species showing that muscle is not an active tissue in accumulating heavy metals [62]. This is agreed with the present study.

The accumulation of investigated trace metals in muscle tissue were in order as; Fe > Zn > Mn > Cr > Cu > Ni > Co. In gill tissue the order of accumulation of the investigated trace metals were as; Fe > Zn > Mn > Cu > Cr > Ni > Co. In liver tissue the order of the accumulation of the investigated trace metals were; Fe > Zn > Cu > Mn > Cr > Ni > Co. In the gill, liver and muscle tissues mean highest accumulation was observed at Fe with 66, 08 µg/g, 206, 63µg/g and 31,21 µg/g levels respectively (Table 5). In our study, it is obvious that Fe has the highest concentration, while Co has the lowest concentration of all measured metals in fish tissues, also Cu and Zn were found to have high accumulation rate in fish tissues. This could be explained by the fact that, Fe, Zn and Cu are essential elements in the bodies of living organisms and has an important role in different physiological processes. Furthermore, this order might be attributed to the different uptake, metabolism and detoxification of metals in fish. Similar observation was recorded by Ibrahim and ElNaggar [63].

Trace metals (µg/g)	**Co**	**Cr**	**Cu**	**Fe**	**Mn**	**Ni**	**Zn**
	Mean±S.E.	Mean±S.E.	Mean±S.E.	Mean±S.E.	Mean±S.E.	Mean±S.E.	Mean±S.E.
	(Range)	(Range)	(Range)	(Range)	(Range)	(Range)	(Range)
Gills	0,44±0,14 a	1,65±0,28 b	1,85±0,02 b	66,08±16,36 b	3,52±0,07 a	1,54±0,08a	23,47±4,84 b
	0,003–2,01	0,24–4,02	0,13–7,56	12,98–322	0,98–15,51	0,35–6,53	9,49–78,2
Liver	0,32±0,08 b	2,17±0,09 a	15,92±3,48 a	206,63±37,6 a	3,08±0,56 b	1,38±0,2 b	35,73±7,36 a
	0,01–1,65	0,69–6,79	2,48–55,31	10,59–685,2	0,39–8,74	0,14–3,58	8,32–86,59
Muscle	0,07±0,02 c	1,73±0,27 b	1,66±0,28 b	31,21±6,75 c	2,04±0,65 c	1,29±0,2 b	22,11±4,67 b
	0,001–0,34	0,34–4,57	0,16–4,82	10,03–79,19	0,05–9,47	0,15–3,7	8,36–65,24
FAO/WHO limits [64]	-	-	30	-	-	-	40
EPA [65]	27	4,1	54	410	190	27	410
TFC [31]	-	-	20	-	20	-	50

Range of International Standards	-	-	10-100	-	-	-	40-100
[66]							
IAEA-407 [67]	-	0,73	3,28	146	3,52	0,60	67,10

Table 5: The mean Trace metals Concentration (µg g^{-1} metal/ wet w) in *A. anguilla* tissues and comparison with the tolerance levels of metal concentrations in fish according to guidelines.
[a,b,c]: The different word in each column is statically different (p<0,05).

The highest level of Fein all the tissues of *A. anguilla* may be due to they forms hydrated ionic condition and may form a variety of complexes with inorganic and organic ligands. In water they may occur as complex and diverse mixture of soluble and insoluble forms such as inorganic and organic complex and or associated with colloids and suspended particulate matter. From this it enters into the organ of fishes through some vulnerable sites such as gills and skins. The fishes also get this Fe from their prey item and biomagnified in the different parts of the body. Feis not considered hazardous to health. In fact, it is essential for good health to transport oxygen in blood. Similar finding was observed by Bamishaiye, et al. [68], on the determination of some trace elements in water samples within Kano meteropolis. The above findings were supported by Farkas, et al. [69] and Olsson [70]. The results confirm the differences of heavy metal accumulation in the different tissues. It is well known that all metal ions taken up by a fish through any route are not totally accumulated because fish can regulate metal concentrations to a certain extent, after which accumulation will occur. Therefore, the ability of each tissue to either regulate or accumulate metal ions can be directly related to the total amount of metal accumulated in that specific tissue. This metal regulation is due to induction of low molecular weight metal-binding proteins, such as metallothionein are closely related to heavy metal exposure and metals taken up from the environment can be detoxified by binding on these proteins [71-73]. Furthermore, the physiological differences and the position of each tissue in the fish can also influencethe accumulation of a particular metal [50,73]. In other words, the amount of a metal accumulated is influenced by various environmental, biological andgenetic factors, leading to the differences in metal accumulation between different individuals, species, tissues, seasons and sites [74]. On the other hand, high levels of metals in tissues of fishes could be originated from different sources around the study area. These sources are; i) for Zn, compossed fertilizer enriched with Zn, ii) for Mn, micro-elements fertilizer and iii) for Cu, conservative fungicides that contains Cu used for citrus fruits plantations and green-houses around Tersakan Stream. Concentrations of trace metals in fish tissues were always higher than that of water [75].

Entry of metals occurs either through gill membrane or through ingestion. A difference in concentration of trace metals in fish organs as indicated by the study was reported by Kalay and Canli [71], Bury, et al. and Chatterjee, et al. [76-78]. Statistical analysis showed that there was a significant difference (p< 0.05) between concentrations of Co, Fe and Mn in all tissues. Also, there was a significant difference (p<0.05) between concentrations in muscle and gill tissues with the concentrations of Cr, Cu and Zn in liver tissue while there was a significant difference (p<0.05) between concentrations in gill tissue with the concentrations of Ni in liver and muscle tissues. Unfortunately, there is no uniform source of guidance or standards for most metal residues in aquatic ecosystems, especially in fish tissue. We could not find single references for acceptable levels of most metals in marine or fresh water fish, self-caught or commercial ones. Information was compiled from documents of the Codex Alimentarius Commission assembled under the aegis of the United Nations Food and Agriculture Organization and the World Health Organization [64], Environmental Protection Agency [65] and national sources as Turkish Food Codes [31].

The trace metal concentrations in the edible tissue (muscle) of *A. anguilla* was compared with the tolerable national and international values in fish. The result obtained showed that the trace metal concentrations in edible tissue did not exceed the WHO/FAO [64], TFC [31], Range of International Standards, EPA [65] and IAEA-407 [67] guidelines. The results of this study revealed that consuming the *A. anguilla* from the Tersakan Stream may not have harmful effects, because the levels of heavy metals are below the permissible limits (Table 5). However, a potential danger may exist in the future, depending on the agricultural development in this region. As the Tersakan Stream is also used for agricultural irrigation purposes, performance of pollution researches at certain periods is of significance for both environment and public health.

Conclusions

Fish samples can be considered as one of the most significant indicators in freshwater systems for the estimation of metal pollution level [79]. The commercial and edible species have been widely investigated in order to check for those hazardous to human health [80]. The present results show that the metal concentrations in muscle, gills and liver tissues of *A. anguilla* are closely associated with metal content of water and sediments in Tersakan Stream.The changes in the metal concentration in the different tissues are associated with metal concentration in eco system, time of exposure, ecological needs, metabolic processes, feeding habits, salinity, temperature and interacting agents. Form the present study, it can be concluded that the trace metal concentration in the different tissues of *A. anguilla* are below the prescribed level. Therefore the muscle of *A. anguilla* caught was suitable for human consumption and did not posed a risk for human health. Considering the results of this study, *A. anguilla* is adequate and most suitable species for use as bio-monitors of trace metals pollution in the Tersakan Stream. Consequently, we recommend the use of these species as biological indicators as a tool for future monitoring programs, to evaluate the evolution of trace metal pollution in this area. Finally, this work may provide valuable database for future research on Tersakan Stream. Regular evaluation of pollutants in the stream is also very important.

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