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RESEARCH ARTICLE

The effects of season on the metal levels of tissues of some lessepsian species caught from the Northeastern Mediterranean Sea.

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ABSTRACT

The concentrations of iron, copper, zinc, chromium, arsenic, cadmium and lead were determined by inductively coupled plasma mass spectrometer (ICP-MS) in the muscle, gill, brain and liver tissues of lessepsian fish species sampled from Taşucu region located on the south of Turkish coastal waters in the Mediterranean Sea for all seasons. While iron showed the highest levels, cadmium showed the lowest levels in the examined tissues of all fish species (*Pelates quadrilineatus*, *Upeneus moluccensis*, *Nemipterus randalli*, *Saurida lessepsianus*). Metal levels showed tissue-dependent changes in the species studied. Cadmium was detected only in the liver tissue in all fish, in addition to a few other tissues (*U. moluccensis*'s brain tissues in summer season and *P. quadrilineatus*'s gill tissues in winter season). The maximum accumulation of Fe except for *N. randalli* and *S. lessepsianus* was detected in liver tissue. The maximum accumulation of Cu in all species was detected in liver tissue. The maximum accumulation of Zn except for *S. lessepsianus* was also detected in liver tissue. The highest accumulations of As and Cr in other fish species except for *N. randalli* were also detected in liver tissue. Except for As accumulation in *N. randalli*, the least accumulation for metals in all species was determined in muscle tissue. According to the seasons, there was no statistically significant relation between metal accumulations. Metal concentrations in edible parts of fish species were 17.26-108.22 μg g^{-1} dw for iron, 0.54-3.65 μg g^{-1} dw for copper, 11.50-31.17 μg g^{-1} dw for zinc, 0.32-1.09 μg g^{-1} dw for chromium, 4.32-69.44 μg g^{-1} dw for arsenic, below limit (not detectable) for cadmium, N.D.-1.12 μg g^{-1} dw for lead. In this study, for all metals except arsenic there is no health risk through an exposure of consumption of certain fish. Additionally, the results obtained for the elements in analyzed fish species were within acceptable limits for human consumption.

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Introduction

Lakes, rivers, seas and oceans have been ignored by people for a long time and used as waste zones which are thought to have unlimited capacity. The widespread discharge of industrial, agricultural and domestic wastewater to coastal waters is becoming widespread in many parts of the world. As a result, pollution has increased rapidly in the water environments of coastal and inland areas and affects the living things in this environment negatively.

In recent years, fish consumption in the world has increased in parallel to its nutritive and therapeutic properties (Storelli, 2008). Besides being an important protein source, fish has rich nutritional content such as basic minerals,

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vitamins and unsaturated fatty acids (Medeiros et al., 2012). The American Heart Association recommended eating fish at least twice per week in order to reach the daily intake of omega-3 fatty acids (Kris-Etherton et al., 2002). However, despite their known benefits, fish are susceptible to metal contamination and when the metal accumulation in the food chain is taken into consideration, fish species can become a serious risk for human health, especially the most consumable fish species (Castro-Gonzalez et al., 2008). Therefore, it is important to investigate the amount of metal accumulation of different fish species in order to provide data on how heavy metals will reach human beings by consuming fish (Visnjic-Jeftic et al., 2010; Kalyoncu and Arslan, 2012).

There are many studies on the target organs of heavy metals in fish used as bioindicators in the aquatic ecosystem (Kalay et al., 1999; Atli and Canli, 2003; Karayakar et al., 2010, 2017; Kulcu et al., 2014). Fish can accumulate high amounts of metal by absorbing metals from the gill surface, kidney, liver and intestinal system (Annabi et al., 2013). Fish can also accumulate heavy metals that are absorbed directly from the water environment or food chain at higher levels than the environmental concentration (Bervoets and Blust, 2003). The accumulation of heavy metals by organisms can be passive or selective. Differences in the accumulation of heavy metals among the living beings are due to assimilation, egestion, or differences in both (Egila and Daniel, 2011).

As the metal accumulation in fish changes according to the metal it is exposed to, it varies according to many factors such as species, tissue (Petrovic et al., 2013), gender, age, size, breeding cycle, swimming style, feeding behavior and living environment of fish (Canli and Atli, 2003; Zao et al., 2010). Mersin Bay, which is located in the Eastern Mediterranean coast of Turkey, has high potential for pollution due to heavily used in agricultural activities fertilizers and pesticides, domestic waste in the region, wastes from the chrome, plastics, fertilizers, glass and port industries (Kalay et al., 2004) and due to the increased population (Karayakar et al., 2010).

With the opening of the Suez Canal and the establishment of the Aswan Dam, no geographical obstacles remain between the Red Sea and the Mediterranean Sea. Indo-Pacific species have begun to migrate to the Eastern Mediterranean in terms of ecological parameters such as salinity and temperature, similar to those of the Red Sea. These species are able to find suitable areas in the Mediterranean Sea due to their feeding habits, habitat and their depth of distribution. That's why, their transition from the Red Sea to the Mediterranean has been accelerated (Golani, 2002).

The most important lessepsian species for the fishing industry of the Northeastern Mediterranean Sea are the Randall's threadfin bream (*N. randalli* Russell, 1986), the brushtooth lizardfish (*S. lessepsianus* Russell, Golani and Tikochinski, 2015), the goldband goatfish (*U. moluccensis* Bleeker, 1855) and the fourlined terapon (*P. quadrilineatus* Bloch, 1790). The aim of this study is to reveal the seasonal changes in the levels of metal accumulation in the tissues (muscle, gill, brain, liver) of some important Lessepsian fish species; *N. randalli* (Russell, 1986), *S. lessepsianus* (Russell, 2015), *U. moluccensis* (Bleeker, 1885), *P. quadrilineatus* (Blotch, 1790) in the Northeast Mediterranean Sea.

Material and Methods

Materials

Samples were caught by trawl net from Taşucu region located on the south Turkish coastal waters of the Mediterranean Sea for all seasons. In every season, 40 specimens in each species were taken (except for *N. randalli* and *S. lessepsianus* in the summer season, for *P. quadrilineatus* except for spring season) and placed in polystyrene boxes with ice and brought to the laboratory within 1 hour and kept at −18°C until analysis. The size and weight of all samples were measured and given in Table 1. The tissues from 40 individuals of each species were taken out. All samples of the homogenates were analyzed on triplicate.

Table 1. The mean total lengths and weights of the samples

Season	Species	Lengths (cm)		Weights (g)	
		$\bar{X}\pm S_{\bar{x}}$	Min-Max.	$\bar{X}\pm S_{\bar{x}}$	Min-Max.
Spring	NR	16.03 ± 0.28^{x}	15-17.3	61.38 ± 2.31^{a}	53-71
	SL	-	-	-	-
	UM	10.56 ± 0.33^{x}	8-12	9.78 ± 0.41^{a}	8-27
	PQ	13.00 ± 0.41^{x}	11-15	42.44 ± 4.00^{a}	25-63
Summer	NR	15.66 ± 0.56^{x}	13-17.3	55.75 ± 5.29^{a}	37-65
	SL	-	-	-	-
	UM	12.22 ± 0.14^{y}	11-13.2	28.78 ± 1.14^{c}	22-35
	PQ	14.01 ± 0.33^{x}	11.6-15.8	48.69 ± 3.01^{b}	36-59
Autumn	NR	19.00 ± 0.23^{y}	17.5-19.5	87.00 ± 2.28^{b}	75-91
	SL	19.74 ± 1.30^{x}	14-24	67.44 ± 12.49^{a}	36-99
	UM	$10,82\pm0,11^{x}$	10-12	15.93 ± 0.56^{b}	11-21
	PQ	14.00 ± 0.20^{x}	33-47	40.31 ± 1.56^{a}	12.7-14.5
Winter	NR	-	-	-	-
	SL	19.48 ± 1.01^{x}	13-22	64.72 ± 10.07^{a}	28-78
	UM	11.50 ± 0.29^{y}	10-15	25.07 ± 1.23^{c}	17-42
	PQ	13.48 ± 0.12^{x}	13-14.,5	41.13 ± 1.20^{a}	36-52

Note: *Superscript letters (a, b, c, d) in the same rows for each season indicate significant differences ($p<0.05$). Superscript letters (x, y, z, q) in the same columns for each tissue in the same species indicate significant differences ($p<0.05$). $\bar{X}\pm S_{\bar{x}}$: mean±standard error, NR: N. randalli, SL: S. lessepsianus, UM: U. moluccensis, PL: P. quadrilineatus.*

Metal Analysis

The samples (0.1 g dry weight) used for metal analysis were dried at 105°C to reach constant weights, and then concentrated nitric acid (4 mL, Merck, Darmstadt, Germany) and percholoric acid (2 mL, Merck, Darmstadt, Germany) were added to the samples, and they were put on a hot plate set to 150°C until all tissues were dissolved (Canli and Atli, 2003).

Inductively coupled plasma mass spectrometer (ICP-MS, Agilent, 7500ce Model, Japan) was used to determine metals. ICP-MS operating conditions were as following: radio frequency (RF) (W), 1500; plasma gas flow rate (L min^{-1}), 15; auxiliary gas

flow rate (L min^{-1}), 1; carrier gas flow rate (L min^{-1}), 1.1; spray chamber T (°C), 2; sample depth (mm), 8, 6; sample introduction flow rate (mL min^{-1}), 1; nebuliser pump (rps), 0.1; extract lens (V), 1.5. The levels of trace element (Fe, Cu, Zn) and toxic metal (Cd, Pb, As, Cr) in samples were detected as µg metal g^{-1} dry weight. High Purity Multi Standard (Charleston, SC 29423) was used for determination of the metal analyses. Standard solutions for calibration curves were prepared by dilutions of the macro, trace elements and toxic metals. Solutions that have been prepared for the toxic metals had a content of lead, cadmium, arsenic and chromium in the range of 1-50 ppb (0.001 to 0.050 mg L^{-1}), for the trace elements had a content of copper, iron, and zinc in the range of 1-50 ppm (1 to 50 mg L^{-1}).

Statistical Analysis

One-way analysis of variance (ANOVA) was run using the SPSS version 22 software. Duncan's multiple range test comparisons at 0.05 significance level were used to evaluate the effects of season on the metal levels of the tissues (muscle, gill, brain and liver) of species.

Results

The highest accumulation of Fe in *N. randalli* is in the gills (1355.6 µg g^{-1} dw) in the autumn season and the lowest accumulation is in the muscle (16.6 µg g^{-1} dw) in the spring season. Similarly, the highest Fe accumulation in *S. lessepsianus* is in the gills (1332.55 µg g^{-1} dw) in autumn season and the lowest Fe accumulation is in muscle tissue in spring season (17.26 µg g^{-1} dw). The lowest accumulation of Fe in *U. moluccensis* is in muscle tissue in spring season (41.64 µg g^{-1} dw), the highest Fe accumulation is in liver tissue in autumn season (1912.31 µg g^{-1} dw). The lowest accumulation of Fe in *P. quadrilineatus* is in muscle tissue in winter season (50.27 µg g^{-1} dw), the highest Fe accumulation is in liver tissue in winter season (927.95 µg g^{-1} dw) (Table 2).

According to the seasons, copper accumulates in the tissues: The highest accumulation in *N. randalli* is in the liver (122.56 µg g^{-1} dw) in spring season and the lowest accumulation is in the muscle (0.88 µg g^{-1} dw) in winter season. The highest accumulation in *S. lessepsianus* is in the liver (32.37 µg g^{-1} dw) in winter season and the lowest accumulation is in the muscle (0.54 µg g^{-1} dw) in winter season. The highest accumulation in *U. moluccensis* is in the liver (132.06 µg g^{-1} dw) in winter season and the lowest accumulation is in the muscle (1.33 µg g^{-1} dw) in spring season. The highest accumulation in *P. quadrilineatus* is in the liver (33.72 µg g^{-1} dw) in winter season and the lowest accumulation is in the muscle (0.75 µg g^{-1} dw) in autumn season (Table 3).

The highest accumulation of Zn in *N. randalli* is in the liver (554.4 µg g^{-1} dw) in spring season and the lowest accumulation is in the muscle (11.50 µg g^{-1} dw) in autumn season. The highest Zn accumulation in *S. lessepsianus* is in the brain tissue (135.08 µg g^{-1} dw) in winter season and the lowest Zn accumulation is in muscle tissue in the same season (14.48 µg g^{-1} dw). The lowest accumulation of Zn in *U. moluccensis* is in muscle tissue in spring season (16.13 µg g^{-1} dw), the highest Zn accumulation is in liver tissue in summer season (640.55 µg g^{-1} dw). The lowest accumulation of Zn in *P. quadrilineatus* is in muscle tissue in autumn season (15.98 µg g^{-1} dw), the highest Zn accumulation is in liver tissue in winter season (153.25 µg g^{-1} dw) (Table 4).

Table 2. The effects of season on iron levels of some tissues of the lessepsian species ($\bar{X}\pm S_{\bar{x}}$, µg g^{-1})

Species	Season	Muscle	Gill	Brain	Liver
NR	*Spring*	$16.60\pm1.63^{a,x}$	$387.99\pm68.86^{c,x}$	$142.7\pm23.66^{b,x}$	$616.33\pm73.22^{c,x}$
	Summer	-	-	-	-
	Autumn	$38.89\pm14.41^{a,x}$	$1355.6\pm235.26^{b,y}$	$112.9\pm22.35^{a,x}$	$929.46\pm180.82^{b,x}$
	Winter	$19.82\pm3.38^{a,x}$	$462.67\pm107.99^{c,x}$	$162.71\pm39.61^{b,x}$	$426.43\pm71.47^{c,x}$
SL	*Spring*	$17.26\pm2.33^{a,x}$	$598.58\pm130.63^{b,x}$	$221.29\pm88.37^{b,x}$	$425.69\pm78.97^{b,x}$
	Summer	-	-	-	-
	Autumn	$41.92\pm11.71^{a,x}$	$1332.55\pm79.11^{c,y}$	$384.19\pm102.59^{b,x}$	$301.01\pm20.83^{b,x}$
	Winter	$17.59\pm4.27^{a,x}$	$956.95\pm127.23^{c,xy}$	$244.89\pm50.59^{b,x}$	$863.95\pm50.84^{c,y}$
UM	*Spring*	$41.64\pm4.46^{a,x}$	$274.1\pm38.55^{b,x}$	$256.07\pm87.98^{b,x}$	$1841.91\pm214.88^{c,y}$
	Summer	$50.35\pm8.10^{a,xy}$	$494.44\pm115.39^{b,xy}$	$293.49\pm76.45^{b,x}$	$540.36\pm94.63^{b,x}$
	Autumn	$62.74\pm5.19^{a,xy}$	$486.58\pm36.94^{c,y}$	$193.54\pm17.61^{b,x}$	$1912.31\pm371.25^{d,y}$
	Winter	$67.09\pm4.94^{a,y}$	$868.42\pm207.88^{c,y}$	$216.43\pm30.90^{b,x}$	$1058.7\pm179.81^{c,xy}$
PQ	*Spring*	-	-	-	-
	Summer	$108.22\pm33.51^{a,x}$	$211.93\pm26.42^{ab,x}$	$142.21\pm24.29^{a,x}$	$394.02\pm86.23^{b,x}$
	Autumn	$52.39\pm11.63^{a,x}$	$372.05\pm36.45^{b,y}$	$240.62\pm46.09^{b,x}$	$920.68\pm117.05^{c,y}$
	Winter	$50.27\pm6.26^{a,x}$	$522.19\pm60.33^{c,y}$	$240.51\pm17.21^{b,x}$	$927.95\pm180.66^{c,y}$

Note: *Superscript letters (a, b, c, d) in the same rows for each season indicate significant differences ($p<0.05$). Superscript letters (x, y, z, q) in the same columns for each tissue in the same species indicate significant differences ($p<0.05$). $\bar{X}\pm S_{\bar{x}}$: mean±standard error, NR: N. randalli, SL: S. lessepsianus, UM: U. moluccensis, PQ: P. quadrilineatus.*

Table 3. The effects of season on copper levels of some tissues of the lessepsian species ($\bar{X}\pm S_{\bar{x}}$, µg g^{-1})

Species	Season	Muscle	Gill	Brain	Liver
NR	*Spring*	$0.93\pm0.29^{a,x}$	$4.29\pm0.38^{b,y}$	$9.15\pm2.73^{b,x}$	$122.56\pm28.49^{c,z}$
	Summer	-	-	-	-
	Autumn	$0.92\pm0.18^{a,x}$	$2.07\pm0.42^{a,x}$	$6.91\pm1.56^{b,x}$	$12.08\pm1.57^{b,x}$
	Winter	$0.88\pm0.08^{a,x}$	$4.05\pm0.60^{b,xy}$	$13.62\pm2.78^{c,x}$	$35.78\pm6.24^{d,y}$
SL	*Spring*	$0.78\pm0.12^{a,x}$	$2.52\pm0.25^{b,x}$	$4.83\pm1.28^{b,x}$	$22.42\pm5.10^{c,x}$
	Summer	-	-	-	-
	Autumn	$3.65\pm1.82^{a,x}$	$2.93\pm0.75^{a,x}$	$15.79\pm8.85^{a,x}$	$23.34\pm9.66^{a,x}$
	Winter	$0.54\pm0.08^{a,x}$	$2.43\pm0.20^{b,x}$	$5.55\pm0.53^{c,x}$	$32.37\pm3.60^{d,x}$
UM	*Spring*	$1.33\pm0.17^{a,x}$	$3.60\pm0.40^{b,x}$	$5.56\pm0.93^{c,x}$	$44.30\pm13.22^{d,x}$
	Summer	$1.53\pm0.05^{a,x}$	$3.50\pm0.17^{b,x}$	$14.68\pm2.72^{c,y}$	$42.39\pm8.87^{d,x}$
	Autumn	$2.42\pm0.15^{a,y}$	$5.36\pm0.74^{b,x}$	$15.13\pm3.24^{c,y}$	$36.50\pm9.54^{c,x}$
	Winter	$2.21\pm0.19^{a,y}$	$3.04\pm0.90^{a,x}$	$13.05\pm2.79^{b,xy}$	$132.06\pm20.10^{c,y}$
PQ	*Spring*	-	-	-	-
	Summer	$1.79\pm0.45^{a,xy}$	$1.71\pm0.22^{a,x}$	$2.48\pm0.74^{a,x}$	$9.00\pm1.70^{b,y}$
	Autumn	$0.75\pm0.13^{a,x}$	$1.36\pm0.19^{ab,x}$	$2.65\pm0.62^{b,x}$	$1.05\pm0.04^{a,x}$
	Winter	$1.95\pm0.26^{a,y}$	$7.52\pm2.01^{b,y}$	$8.30\pm1.09^{b,y}$	$33.72\pm7.82^{c,z}$

Note: *Superscript letters (a, b, c, d) in the same rows for each season indicate significant differences (p<0.05). Superscript letters (x, y, z, q) in the same columns for each tissue in the same species indicate significant differences (p<0.05).* $\bar{X}\pm S_{\bar{x}}$*: mean±standard error, NR: N. randalli, SL: S. lessepsianus, UM: U. moluccensis, PQ: P. quadrilineatus.*

Table 4. The effects of season on zinc levels of some tissues of the lessepsian species ($\bar{X}\pm S_{\bar{x}}$, µg g^{-1})

Species	Season	Muscle	Gill	Brain	Liver
NR	*Spring*	$16.19\pm2.12^{a,x}$	$62.08\pm2.43^{b,y}$	$59.77\pm10.86^{b,x}$	$554.4\pm42.32^{c,z}$
	Summer	-	-	-	-
	Autumn	$11.50\pm0.65^{a,x}$	$30.07\pm7.81^{b,x}$	$38.76\pm6.68^{b,x}$	$94.00\pm9.68^{c,x}$
	Winter	$13.02\pm0.58^{a,x}$	$52.56\pm4.57^{b,y}$	$90.41\pm29.78^{b,x}$	$211.93\pm16.99^{c,y}$
SL	*Spring*	$17.60\pm0.94^{a,x}$	$109.07\pm17.95^{b,x}$	$89.29\pm10.46^{b,x}$	$79.84\pm16.55^{b,x}$
	Summer	-	-	-	-
	Autumn	$20.16\pm2,69^{a,x}$	$91.78\pm10.68^{b,x}$	$84.15\pm21.47^{b,x}$	$96.76\pm13.72^{b,x}$
	Winter	$14.48\pm1,45^{a,x}$	$92.40\pm13.34^{b,x}$	$135.08\pm24.8^{b,x}$	$94.25\pm31.45^{b,x}$
UM	*Spring*	$18.13\pm0.57^{a,x}$	$102.38\pm5.00^{b,y}$	$103.66\pm41.31^{b,x}$	$417.52\pm66.35^{c,x}$
	Summer	$25.57\pm3.55^{a,x}$	$118.07\pm10.11^{c,y}$	$70.97\pm4.36^{b,x}$	$640.55\pm57.39^{d,x}$
	Autumn	$19.33\pm1.90^{a,x}$	$105.7\pm3.05^{c,y}$	$63.84\pm12.72^{b,x}$	$239.59\pm42.83^{d,x}$
	Winter	$20.92\pm1.33^{a,x}$	$60.41\pm11.41^{b,x}$	$46.12\pm6.01^{b,x}$	$587.64\pm154.13^{d,x}$
PQ	*Spring*	-	-	-	-
	Summer	$31.17\pm8.78^{a,x}$	$58.48\pm15.73^{a,x}$	$44.12\pm7.79^{a,x}$	$65.2\pm8.47^{a,x}$
	Autumn	$15.98\pm1.14^{a,x}$	$52.87\pm3.25^{b,x}$	$53.02\pm13.27^{b,x}$	$136.69\pm8.30^{c,y}$
	Winter	$17.07\pm1.17^{a,x}$	$93.48\pm19.22^{bc,x}$	$59.25\pm12.26^{b,x}$	$153.25\pm15.16^{c,y}$

Note: *Superscript letters (a, b, c, d) in the same rows for each season indicate significant differences (p<0.05). Superscript letters (x, y, z, q) in the same columns for each tissue in the same species indicate significant differences (p<0.05).* $\bar{X}\pm S_{\bar{x}}$*: mean±standard error, NR: N. randalli, SL: S. lessepsianus, UM: U. moluccensis, PQ: P. quadrilineatus.*

The lowest accumulation of Zn in *U. moluccensis* is in muscle tissue in spring season (16.13 µg g^{-1} dw), the highest Zn accumulation is in liver tissue in summer season (640.55 µg g^{-1} dw). The lowest accumulation of Zn in *P. quadrilineatus* is in muscle tissue in autumn season (15.98 µg g^{-1} dw), the highest Zn accumulation is in liver tissue in winter season (153.25 µg g^{-1} dw) (Table 4).

According to the seasons, chromium accumulates in the tissues: The highest accumulation in *N. randalli* is in the brain (3.37 µg g^{-1} dw) in spring season and the lowest accumulation is in the muscle (0.32 µg g^{-1} dw) in winter season. The highest accumulation in *S. lessepsianus* is in the brain tissue (8.59 µg g^{-1} dw) in autumn season and the lowest accumulation is in the muscle (0.33 µg g^{-1} dw) in winter season. The highest accumulation in *U. moluccensis* is in the gill (9.97 µg g^{-1} dw) in

summer season and the lowest accumulation is in the muscle (0.55 µg g^{-1} dw) in winter season. The highest accumulation in *P. quadrilineatus* is in the liver (3.92 µg g^{-1} dw) in summer season and the lowest accumulation is in the muscle tissue (0.53 µg g^{-1} dw) in summer season (Table 5).

The highest accumulation of As in *N. randalli* is in the liver (74.43 µg g^{-1} dw) in winter season and the lowest accumulation is in the brain tissue (19.16 µg g^{-1} dw) in autumn season. The highest As accumulation in *S. lessepsianus* is in the liver tissue (96.76 µg g^{-1} dw) in autumn season and the lowest accumulation of As is in brain tissue in spring season (13.37 µg g^{-1} dw). The lowest accumulation of As in *U. moluccensis* is in gill tissue in summer season (13.26 µg g^{-1} dw), the highest As accumulation is in the liver tissue in winter season (165.54 µg g^{-1} dw). The lowest accumulation of As in *P. quadrilineatus* is in muscle tissue in summer season (4.32 µg g^{-1} dw), the highest As accumulation is in liver tissue in winter season (24.52 µg g^{-1} dw) (Table 6).

Table 5. The effects of season on chromium levels of some tissues of the lessepsian species ($\bar{X}\pm S_{\bar{x}}$, µg g^{-1})

Species	Season	Muscle	Gill	Brain	Liver
NR	*Spring*	$0.42 \pm 0.03^{a,x}$	$1.67 \pm 0.27^{b,x}$	$3.37 \pm 1.14^{b,x}$	$1.07 \pm 0.24^{ab,x}$
	Summer	-	-	-	-
	Autumn	$0.71\pm0.19^{a,x}$	$3.21\pm0.92^{b,x}$	$2.32\pm1.01^{ab,x}$	$1.49\pm0.34^{a,x}$
	Winter	$0.32\pm0.02^{a,x}$	$1.75\pm0.36^{b,x}$	$1.48\pm0.27^{b,x}$	$0.68\pm0.11^{a,x}$
SL	*Spring*	$0.40\pm0.05^{a,x}$	$2.76\pm0.95^{bc,x}$	$5.35\pm1.25^{c,xy}$	$1.32\pm0.39^{b,x}$
	Summer	-	-	-	-
	Autumn	$0.94\pm0.17^{a,y}$	$1.42\pm0.33^{a,x}$	$8.59\pm0.57^{b,y}$	$1.02\pm0.16^{a,x}$
	Winter	$0.33\pm0.03^{a,x}$	$3.22\pm0.32^{b,x}$	$2.69\pm0.73^{b,x}$	$0.56\pm0.21^{a,x}$
UM	*Spring*	$0.91\pm0.16^{a,x}$	$0.83\pm0.14^{a,x}$	$2.66\pm0.64^{b,x}$	$3.14\pm0.22^{b,xy}$
	Summer	$1.09\pm0.43^{a,x}$	$9.97\pm1.05^{c,z}$	$2.84\pm0.47^{b,x}$	$0.99\pm0.17^{a,x}$
	Autumn	$0.94\pm0.09^{a,x}$	$1.86\pm0.13^{b,y}$	$2.70\pm0.47^{b,x}$	$4.62\pm0.90^{b,y}$
	Winter	$0.55\pm0.02^{a,x}$	$3.15\pm0.78^{b,y}$	$1.80\pm0.40^{b,x}$	$1.88\pm0.41^{b,x}$
PQ	*Spring*	-	-	-	-
	Summer	$0.53\pm0.10^{a,x}$	$0.59\pm0.06^{a,x}$	0.86 ± 0.18^{a}	3.92 ± 0.98^{b}
	Autumn	$0.75\pm0.13^{a,x}$	$1.36\pm0.19^{ab,xy}$	2.65 ± 0.62^{b}	1.05 ± 0.04^{a}
	Winter	$0.53\pm0.05^{a,x}$	$1.48\pm0.37^{a,y}$	2.93 ± 1.29^{a}	0.72 ± 0.06^{a}

Note: *Superscript letters (a, b, c, d) in the same rows for each season indicate significant differences (p<0.05). Superscript letters (x, y, z, q) in the same columns for each tissue in the same species indicate significant differences (p<0.05).* $\bar{X}\pm S_{\bar{x}}$*: mean±standard error, NR: N. randalli, SL: S. lessepsianus, UM: U. moluccensis, PQ: P. quadrilineatus.*

Table 6. The effects of season on arsenic levels of some tissues of the lessepsian species ($\bar{X}\pm S_{\bar{x}}$, µg g^{-1})

Species	Season	Muscle	Gill	Brain	Liver
NR	*Spring*	$55.57\pm4.27^{a,y}$	$50.48\pm9.19^{a,x}$	$34.21\pm11.43^{a,x}$	$51.44\pm11.42^{a,x}$
	Summer	-	-	-	-
	Autumn	$24.66\pm6.22^{ab,x}$	$27.43\pm13.76^{ab,x}$	$19.16\pm4.10^{a,x}$	$64.11\pm14.65^{b,x}$
	Winter	$69.44\pm12.07^{a,y}$	$61.61\pm8.26^{a,x}$	$49.04\pm13.28^{a,x}$	$74.43\pm11.7^{a,x}$
SL	*Spring*	$23.83\pm4.28^{a,x}$	$17.05\pm1.65^{a,x}$	$13.37\pm1.37^{a,x}$	$16.36\pm3.22^{a,x}$
	Summer	-	-	-	-
	Autumn	$20.16\pm2.69^{a,x}$	$91.78\pm10.68^{b,y}$	$84.15\pm21.47^{b,y}$	$96.76\pm33.72^{b,y}$
	Winter	$25.58\pm7.92^{a,x}$	$19.96\pm6.65^{a,x}$	$13.55\pm2.75^{a,x}$	$19.13\pm5.47^{a,x}$
UM	*Spring*	$32.53\pm6,.28^{a,x}$	$51.6\pm8.81^{ab,y}$	$37.13\pm6.64^{a,y}$	$85.21\pm9.79^{b,xy}$
	Summer	$24.3\pm3.89^{a,x}$	$13.26\pm1.49^{a,x}$	$16.15\pm2.82^{a,x}$	$20.31\pm2.60^{a,x}$
	Autumn	$28.99\pm1.95^{a,x}$	$41.4\pm3.26^{ab,y}$	$31.88\pm2.73^{a,y}$	$54.79\pm5.72^{b,xy}$
	Winter	$38.59\pm9.59^{a,x}$	$14.15\pm4.00^{a,x}$	$30.35\pm3.61^{a,y}$	$165.54\pm35.89^{b,y}$
PQ	*Spring*	-	-	-	-
	Summer	$4.32\pm0.67^{a,x}$	$5.28\pm1.57^{a,x}$	$5.20\pm0,.80^{a,x}$	$6.20\pm0.82^{a,x}$
	Autumn	$4.99\pm0.67^{a,x}$	$4.80\pm0.34^{a,x}$	$4.72\pm0.50^{a,x}$	$13.17\pm1.64^{b,y}$
	Winter	$5.88\pm0.53^{a,x}$	$9.43\pm3.25^{a,x}$	$5.17\pm0.74^{a,x}$	$24.52\pm2.49^{b,z}$

Note: *Superscript letters (a, b, c, d) in the same rows for each season indicate significant differences (p<0.05). Superscript letters (x, y, z, q) in the same columns for each tissue in the same species indicate significant differences (p<0.05).* $\bar{X}\pm S_{\bar{x}}$*: mean±standard error, NR: N. randalli, SL: S. lessepsianus, UM: U. moluccensis, PQ: P. quadrilineatus*

Cadmium has been determined in the liver tissues of fish for all seasons between 0.7-5.51 µg g^{-1} dw and has also been determined in the gill tissue (1.21 µg g^{-1} dw) of *P. quadrilineatus* in winter season and brain tissue (0.99 µg g^{-1} dw) of *U. moluccensis* in summer season. The level of cadmium for the other tissues is below the limits that can be determined (Table 7).

According to the seasons, lead accumulates in the tissues: The highest accumulation in *N. randalli* is in the gill tissue (1.88 µg g^{-1} dw) in autumn season and the lowest accumulation is in the muscle (under the detection limit) in autumn season.

Table 7. The effects of season on cadmium levels of some tissues of the lessepsian species ($\bar{X}\pm S_{\bar{x}}$, µg g^{-1})

Species	Season	Muscle	Gill	Brain	Liver
NR	*Spring*	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	1.08±0.23^{b,x}
	Summer	-	-	-	-
	Autumn	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	1.16±0.06^{b,x}
	Winter	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	1.40±0.30^{b,x}
SL	*Spring*	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}
	Summer	-	-	-	-
	Autumn	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	1.11±0.05^{b,z}
	Winter	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	0.45±0.09^{b,y}
UM	*Spring*	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	2.23±0.70^{b,xy}
	Summer	N.D.^{a,x}	N.D.^{a,x}	0.99±0.04^{a,y}	0.82±0.23^{b,x}
	Autumn	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	2.22±0.40^{b,xy}
	Winter	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	5.51±1.19^{b,y}
PQ	*Spring*	-	-	-	-
	Summer	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	0.83±0.02^{b,x}
	Autumn	N.D.^{a,x}	N.D.^{a,x}	N.D.^{a,x}	0.70±0.09^{b,x}
	Winter	N.D.^{a,x}	1.21±0.21^{b,y}	N.D.^{a,x}	1.14±0.25^{b,x}

Note: *Superscript letters (a, b, c, d) in the same rows for each season indicate significant differences (p<0.05). Superscript letters (x, y, z, q) in the same columns for each tissue in the same species indicate significant differences (p<0.05).* $\bar{X}\pm S_{\bar{x}}$*: mean±standard error, NR: N. randalli, SL: S. lessepsianus, UM: U. moluccensis, PQ: P. quadrilineatus*, N.D.: not detected.

Table 8. The effects of season on lead levels of some tissues of the lessepsian species ($\bar{X}\pm S_{\bar{x}}$, µg g^{-1})

Species	Season	Muscle	Gill	Brain	Liver
NR	*Spring*	0.35±0.01^{a,z}	0.86±0.10^{b,x}	1.85±0.01^{c,q}	0.87±0.19^{b,x}
	Summer	-	-	-	-
	Autumn	N.D.^{a,x}	1.88±0.34^{d,x}	0.81±0.04^{c,y}	0.57±0.02^{b,x}
	Winter	0.14±0.02^{a,y}	1.02±0.12^{bc,x}	1.49±0.09^{c,z}	0.87±0.03^{b,x}
SL	*Spring*	N.D.^{a,x}	1.09±0.33^{b,x}	N.D.^{a,x}	0.80±0.26^{b,y}
	Summer	-	-	-	-
	Autumn	N.D.^{a,x}	0.79±0.05^{c,x}	2.36±0.16^{d,z}	0.38±0.03^{b,y}
	Winter	N.D.^{a,x}	1.15±0.15^{c,x}	1.49±0.15^{c,y}	0.19±0.04^{b,x}
UM	*Spring*	0.52±0.02^{a,y}	0.90±0.11^{b,x}	1.98±0.15^{c,q}	1.25±0.13^{b,y}
	Summer	1.12±0.12^{b,z}	1.79±0.09^{c,y}	0.52±0.03^{a,y}	0.61±0.07^{a,x}
	Autumn	N.D.^{a,x}	0.88±0.14^{b,x}	1.17±0.07^{b,z}	1.10±0.17^{b,xy}
	Winter	N.D.^{a,x}	2.38±0.21^{c,y}	N.D.^{a,x}	1.25±0.11^{b,y}
PQ	*Spring*	-	-	-	-
	Summer	0.77±0.20^{a,y}	1.46±0.24^{a,x}	0.84±0.27^{a,y}	1.39±0.40^{a,x}
	Autumn	N.D.^{a,x}	0.87±0.09 ^{b,x}	N.D.^{a,x}	0.73±0.05^{b,x}
	Winter	N.D.^{a,x}	0.69±0.13^{b,x}	1.10±0.10^{b,y}	0.74±0.03^{b,x}

Note: *Superscript letters (a, b, c, d) in the same rows for each season indicate significant differences (p<0.05). Superscript letters (x, y, z, q) in the same columns for each tissue in the same species indicate significant differences (p<0.05).* $\bar{X}\pm S_{\bar{x}}$*: mean±standard error, NR: N. randalli, SL: S. lessepsianus, UM: U. moluccensis, PQ: P. quadrilineatus*, N.D.: not detected.

The highest accumulation in *S. lessepsianus* is in the brain tissue (2.36 µg g^{-1} dw) in autumn season and the lowest accumulation is in the muscle (under the detection limit) in all season. The highest accumulation in *U. moluccensis* is in the gill (2.38 µg g^{-1} dw) in winter season and the lowest accumulations is in the muscle (under the detection limit) in autumn and winter seasons and in the brain tissue (under the detection limit) in winter season. The highest accumulation in *P. quadrilineatus* is in the gill (1.46 µg g^{-1} dw) in summer season and the lowest accumulations is in the muscle and brain tissues (under the detection limit) in autumn and in the muscle tissue (under the detection limit) in winter season (Table 8).

Discussion

Fish are under the influence of many pollutants in the aquatic ecosystem. The different concentration of contaminants in their tissues can be attributed to the expression of metalloproteins at different ratios in their different tissues (Spanopoulos-Zarco et al., 2017). Three of the seven analyzed metals (Cu, Zn, Cd) were found in liver tissues with the highest accumulation for all seasons and in all species. Liver plays an important role in contaminant storage, redistribution, detoxification or transformation and also acts as an active site of pathological effects induced by contaminants (Evans et al., 1993). As far as relative abundances of analyzed elements are concerned, the sequence of average concentrations in all tissues of fish from all seasons were Fe > Zn > As > Cu > Cr > Pb > Cd.

In fish, metal accumulation differs according to the type of metal (Tchounwou et al., 2012). Cadmium was detected only in the liver tissue in all fish, in addition to a few other tissues (*P. quadrilineatus*'s gill tissues in winter season and *U. moluccensis*'s brain tissues in summer season). The maximum accumulation of Fe except for *N. randalli* and *S. lessepsianus* was detected in liver tissue. The maximum accumulation of Cu in all species was detected in liver tissue. The maximum accumulation of Zn except for *S. lessepsianus* was also detected in liver tissue. The highest accumulations of As and Cr in other fish species except for the *N. randalli* were also detected in liver tissue. Except for As accumulation in *N. randalli*, the least accumulation for metals in all species was determined in muscle tissue. This difference between tissue and organs in terms of metal concentration may be due to ecological needs, swimming behaviors and metabolic activities among different fish species as well as their different structures and functions. Differences in metal concentrations in tissues may be an outcome of inducing capacity for metal binding proteins such as metallothioneins (Heath, 1995; Atli and Canli, 2003).

While low levels of metals such as Cu, Fe and Zn are necessary for enzymatic activity and for various biological processes in fish, their high levels may become toxic (Al-Weher, 2008; Ozden et al., 2010). Essential metals cause toxic effects on fish, such as the alteration of physiological activities and biochemical parameters in their blood (Nemesok, 1988). Kulcu et al. (2014) reported that Fe, Cu and Zn levels of muscles of *N. randalli*, *S. lessepsianus* and *U. moluccensis* from the Northeastern Mediterranean Sea were 85.52, 0.24, 16.81; 78.12, 0.34, 17.59; 83.7, 1.37, 24.57 µg g^{-1} dw, respectively. The findings of the researchers are similar to our findings.

Gills are the first target organs followed by the liver and muscle for iron toxicity (Jahan et al., 2015). The basic biological function of iron is to transport oxygen from the respiratory organs to the cells and the carbon dioxide from cells to the respiratory organs. Adsorption of metals to the gill surface can have a significant effect on the total metal level of the gill tissue (Canli and Furness, 1993). According to our results, the most iron accumulation for *N. randalli* and *S. lessepsianus* was in the gills, the liver tissue was in the second rank. While the maximum accumulation for *U. moluccensis* and *P. quadrilineatus* was in the liver, it was followed by the gill tissue.

Fish are under the influence of many biotic and abiotic factors in the aquatic environment. Heavy metal concentrations in fish tissues show seasonal changes depending on seasonal changes in water parameters (water temperature, pH, salinity) and changes in food consumption habits of fish (Regoli and Orlando, 1994; Foster et al., 2000). This study aimed to reveal the relation between heavy metal concentrations depending on seasons and species in particular. The results show that there is no statistically significant relationship between metal accumulation and seasonality.

The accumulation of metal in fish varies depending on the species of fish (Canli and Atli, 2003; Erdem et al., 2004; Dural et al., 2007; Karayakar et al. 2010, 2017). In this study, statistically significant differences were found in the metal concentrations of each tissue from different fish species. It was found that all of the metals showed the highest accumulation in *U. moluccensis* tissues in all seasons. The lowest accumulation was found in *P. quadrilineatus* for Fe, As, Pb, Cd metals, and *S. lessepsianus* for Zn and Cu metals. This difference can stem from many different biological needs, such as feeding habits and habitats (Kalay et al., 1999; Yılmaz, 2003).

Fish muscle tissue is the most important target tissue for metal accumulation, and also the most analyzed tissue of the reasons why fish muscle tissue constitute the main edible part of the fish (Kumar et al., 2011). Significant differences were found among the tissues in studies carried out by various researchers on various fish species; accumulation was found to be lowest in muscle tissue (Kalay et al., 1999; Dural et al., 2007; Karayakar et al., 2010). In this study, the lowest metal accumulation was found in muscle tissue in all samples except for As accumulation in *S. lessepsianus*.

Lead is a non-essential element that increases mucus formation in fish when taken at high doses and causes lack of survival, growth rates, development and metabolism (Burger et al., 2002). Kalay et al. (1999) determined Pb level in *Mullus barbatus, Caranx crysos* and *Mugi1 cephalus* muscle tissues collected in Mersin Bay as 4.43–9.11 µg g^{-1}. Kulcu et al. (2014) determined Pb level in muscle tissues of *N. randalli*, *S. lessepsianus*, *U. moluccensis* collected from the Northeastern Mediterranean Sea as 6.20, 5.44, 5.63 (µg g^{-1} dw), respectively. Our findings for Pb metal were found to be in the range of N.D.-1.12 µg g^{-1} dw and were also found to be lower than findings obtained in other studies in the region.

Chromium, which has widespread use in the industry, is considered a heavy metal and pollutant, as well as a biologically available form, a microelement that plays an important role in glucose metabolism. Chromium was detected in almost all the samples, with the highest concentration (1.09 µg g^{-1} dw). In muscle of *U. moluccensis* the values were within the limits of 12–13 mg kg^{-1} (USFDA, 1993).

Cd is a serious contaminant and highly toxic element, which is transported in the air. Mormede and Davies (2001) suggested that liver was the target organ showing the detoxification and accumulation role of liver. According to our findings, cadmium was detected only in the liver tissue in all fish (0.7-5.51 µg g^{-1} dw), in addition to a few other tissues (*P. quaderilineatus*'s gill tissues in winter season and *U. moluccensis*'s brain tissues in summer season).

Arsenic is biologically available to aquatic organisms living in contaminated habitats (Eisler, 1988). The exposure of fish to arsenic, as other nonessential metals, is conditioned by the concentration of this element in the surrounding water (Pagenkopf, 1983). Kalantzi et al. (2017) found As values for sardine and anchovy in the Greek coastline as 8.6 to 58.8 mg kg^{-1} dw. Similar to these findings, the amount of As in our study was found to be 4.32-69.44 µg g^{-1} dw. The Joint FAO/WHO Expert Committee (1983) imposed a limit of 0.1 mg kg–1, ww for total arsenic in food (Muñoz et al., 2000; De Gieter et al., 2002). In all species except *P. quadrilineatus*, the amount of As was found to be very high.

The accumulation of As and Fe metals is higher than the other studies. The most important minerals containing arsenic are as follow: As_2S_3 (orpiment), AsS (realgar), FeAsS, $FeAs_2$, NiAs, CoAsS, $Cu_{12}As_4S_{13}$ and Cu_3AsS_4 (Matschullat, 2000; Bissen and Frimmel, 2003). The coexistence of these two heavy metals in the aquatic environment supports our findings.

Conclusion

Heavy metals (Fe, Cu, Zn, Cr, As, Cd and Pb) accumulations were investigated according to the tissues (muscle, gill, brain, liver) in the 4 important lessepsian fish species from Mersin Bay. The highest level in these 7 metals was determined for Fe, and the lowest accumulation for Cd. Nonetheless, these values were in the range stated in the literature.

Metal concentrations in edible parts of fish species were 17.26-108.22 µg g^{-1} dw for iron, 0.54-3.65 µg g^{-1} dw for copper, 11.50-31.17 µg g^{-1} dw for zinc, 0.32-1.09 µg g^{-1} dw for chromium, 4.32-69.44 µg g^{-1} dw for arsenic, below limit (not detectable) for cadmium, N.D.-1.12 µg g^{-1} dw for lead. In this study, for all metals except arsenic there is no health risk through an exposure of consumption of certain fish. In all species except *P. quadrilineatus*, the amount of As was found to be very high. It is thought that this is due to the uncontrolled use of pesticides and fertilizers because of the agricultural area of the region. In this study, for all metals except arsenic there is no health risk through an exposure of consumption of certain fish. Additionally, the results obtained for the elements in analyzed fish species were within acceptable limits for human consumption.

Conflict of Interest

The authors declare that there is no conflict of interest.

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