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Seasonal Variations Of Heavy Metals In Common Carp (CyprinusCarpio L., 1758) Collected From Sikkak Dam Of Tlemcen (Algeria)

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ABSTRACT
Seasonal variations in the concentrations of six heavy metals (Cd, Fe, Cu, Pb, Zn, and Ni) were determined from December 2010 to November 2011 in four different tissues (muscles, gills, gonads, and livers) of common carp (*Cyprinuscarpio* L., 1758) from Sikkak dam at Ainyoucef (Wilaya of Tlemcen) which is an important water source for irrigation and drinking in northwest part of Algeria. Heavy metals in fish samples were analyzed by atomic absorption spectrophotometry (AAS) after dry digestion. One-way ANOVA and principal component analysis (PCA) were used to compare the data among seasons (level of 0,05). Mean concentrations were found to decrease in sequence of *Cyprinuscarpio* samples, in muscle and liver as Fe > Zn >Pb> Ni > Cu > Cd, in gills as Zn > Fe >Pb> Ni > Cu > Cd, and in gonads Fe > Zn > Ni >Pb> Cu > Cd. In samples Cd, Pb and Zn concentrations exceeded the tolerable values provided by FAO/WHO.
The highest metals concentrations were found in liver followed by gills, by gonads, and by muscle. Heavy metal levels in tissues of carp were decreased in winter. The obtained results showed that the average values of Cd, Cu, Zn and Ni were at the highest levels in summer. The highest Pb levels were measured in the spring and Fe in autumn. It found that all tested metals in organs did not reveal any significant difference between different seasons (P > 0,05), (inter-season comparison).
Keywords - Heavy metal, pollution, *Cyprinuscarpio*, Sikkak dam, Tlemcen.

I. INTRODUCTION

Aquatic pollution started long back but intensified during the last few decades, and now the situation has been alarming. Contamination of aquatic ecosystems with metals has been receiving increased worldwide attention [1-12].

Fish species are widely used to biologically monitor variations in environmental levels of anthropogenic pollutants [7-12]. Fish are often at the top of the aquatic food chain and may concentrate large amounts of some metals from the water [9]. In fish, the toxic effects of heavy metals may influence physiological functions, individual growth rates, reproduction and mortality [9-13]. The concentrations of heavy metals in tissues are the result of uptake and release processes with characteristic kinetics for the elements and their biological halftime, influenced by the age and size of individuals, the feeding habits of the species, their life cycle and life history, and also the seasons [9, 14-17]. Heavy metals may enter fish bodies in three possible ways; through the body surface, the gills or the digestive tract [18-19]. The gills are regarded as the important site for direct uptake from the water [18, 20-21], whereas the body surface is generally assumed to play a minor role in heavy metal uptake of fish [18-19, 22]. Food may also be an important source for heavy metal accumulation [18, 22], potentially leading to bio-magnifications, the increase of pollutants up to the food chain [6]. Muscle, gills, gonads and liver were chosen as target organs for assessing metal accumulation. The concentrations of metals in gills reflect those of metals in the water. The metal content in the dorsal muscle was analyzed because of its importance for human consumption and that in the gonads.

The Sikkak dam is located in Ainyoucef on the east of Tlemcen (North West of Algeria). The dam was constructed to help alleviate the water problems of the Sikkak basin and many inhabitants of the area also carry out fisheries on the dam. The quality of this ecosystem has been degrading due to agriculture and humane activities. To the best of our knowledge, from literature survey, no work has been carried out on the environmental quality biota of the dam.

The present study has been conducted to determine cadmium, iron, copper, lead, zinc, and nickel concentrations in the four different tissues (muscle, gills, gonads, and liver) of *cyprinuscarpio*L., 1758 from Sikkak dam since this fish is an important component of the humane diet in this zone. This will be useful as an alarm signal to minimize the rate of pollution of heavy metals in the dam and for management programs of the dams.

II. MATERIALS AND METHODS

2.1 Area descriptions

DamSikkak, located in wadiSikkak in the northwest of Algeria (Fig. 1), is used for irrigation, drinking water supply and fisheries in the region. It confluence with WadiIsser takes place at 81 m of Remchi.It volume is about 27 hm³.Recently, agricultural developments as well as increase in pollution substantially increased the contamination of fish with heavy metals.

2.2 Sampling and sample preparation

40 specimens of fish species (*Cyprinuscarpio*) were collected from Sikkak dam during the fourseasons from december 2010 to november 2011 using a motorized boat put at our disposal by the administration of the dam and also using a tri mesh net. Fish species were kept in a cooler and transported to the laboratory, where their age total body length and total wet weight were recorded (Table 1).

The age of carp was determined from scales, which were removed from the left side between the posterior end the pectoral fin and the anterior one of the dorsal fin. Four parts, muscle, gonads, gills, and liver were removed by plastic knife, weighted, and kept in polyethylene bags, closed and labeled at low temperature until digestion. The determination of the sex (males, females) is made after the dissection.

Digestion was conducted according to dry method. All the samples of fish went through dry mineralization. One to two grams of organic matter were placed in an oven at 110°C for 3 hours. For ash reduction, they were placed in a muffle furnace for 15 min at 450°C, next moistened with HNO_3 and then put back in the furnace at 350°C for 1h30min. The ashes were then filtered using a filter paper of 0,45 µm of porosity and a swinex, and adjusted to 25 ml with nitric acid solution (1%), then packed in polyethylene bags and kept in the refrigerator until analysis. AAURORA AI. 1200 Flame Atomic Absorption Spectrophotometer was used to determine the concentrations of metals (Cd, Fe, Cu, Pb,Zn and Ni). The concentrations of heavy metals in mg/kg represent the amount of metal per kilogram of dry weight of fish.A blank was added to each digestion series to determine the exogenous contamination. The validity and reproducibility of the method were verified by analyzing the Certified Standard DORM-2 from the National Research Council of Canada.The later was analysed under the same experimental conditions. Replicate analysis of these reference materials showed good accuracy with recovery rates for metals between 104 %. The absorption wavelengths and detections limits were 228,6 nm and 0,001 ppm for Cd, 248,3 nm and 0,0046 ppm for Fe, 324,8 nm and 0,007 ppm for Cu, 283,3 nm and 0,0042 ppm for Pb, 213,9 nm and 0,0059 ppm for Zn, and 232 nm and 0,015 ppm for Ni respectively.

2.3 Statistical procedures

Statistical analysis of data was carried out using XLSTAT statistical package programs. One way ANOVA test was used to compare the data among seasons at the level of 0,05 and also we performed principal component analysis (ACP) on the mean of the metal concentrations in carp's organs of each season.

III. RESULTS AND DISCUSSION

Table 1 shows length, weight ranges and their relationships. The levels of Cd, Fe, Cu, Pb, Zn, Ni measured in *Cyprinuscarpio*tissues (muscles, gills, gonads, and livers) during four seasons are presented in Table 2.

Figure 2 shows the temporal variations of the mean content of the various metallic elements in the different organs obtained from samples analyzed by atomic absorption spectrophotometry (AAS). The horizontal line in each graph indicates the recommended values for each metallic element in fish according to FAO/WHO [23].

Different tissues showed different capacities for accumulation of heavy metals. Concentrations of heavy metals were highest in livers, and lowest in the muscles. Cadmium concentrations in muscle, gills, gonads, and liver of *Cyprinuscarpio*ranged respectively from 0,13 to 0,22 mg/kg, from 0,14 to 0,34 mg/kg, from 0,05 to 0,10 mg/kg, and from 0,06 to 0,10 mg/kg, (Table 2). The highest Cd concentrations were recorded in gills in summer. These average concentrations in muscles, gills and gonads exceed tolerable values(0,1mg/kg)throughout the study period [23].

Iron levels in muscle, gills, gonads, and liver of carp ranged respectively between55,29 and 73,50 mg/kg, between 79,75 and 74,20 mg/kg, between 179,21 and 252,42 mg/kg and between 123,75 and 268,33 mg/kg (Table 2).In liver, iron concentrations were foundto be highest during autumn season.

Copper levels in muscle, gills, gonads, and liver of *Cyprinuscarpio* ranged respectively between 0,08 and 0,13 mg/kg, between 0,29 and 0,46 mg/kg, between 0,19 and 0,30 mg/kg and between 0,20 and 0,27 mg/kg (Table 2).In gill copper concentrations were foundto be highest during summer season.

These concentrations recorded do not exceed the tolerable values, 10 mg/kg [23].

Concentrations of lead, in muscle, gills, gonads, and liver of the carpranged respectively from1,61 to 2,28 mg/kg, from 1,36 to 2,36 mg/kg, from 0,20 to 0,33 mg/kg and from 2,69 to 2,88 mg/kg (Table 2). The highest lead concentrations were found in liver in winter. Lead concentrations exceed the recommended values 0,5mg/kg [23].

Zinc levels in muscle, gills, gonads, and liver of *Cyprinuscarpio* ranged respectively between 14,58 and 20,33 mg/kg, between 67,42 and 117,50 mg/kg between 42,75 and 80,94 mg/kg, and between 62,71 and 72,45 mg/kg (Table 2). The highest zinc concentrations were recorded in gills in summer. Zinc concentrations exceed the recommended values 50 mg/kg [23].

Nickel levels in muscle, gills, gonads, and liver ranged respectively between 0,39 and 0,85 mg/kg, from 1,18 to 1,78 mg/kg, from 0,89 to 1,60 mg/kg and from 1,20 to 1,64 mg/kg (Table 2), and reached the highest level in gills in summer. These concentrations recorded do not exceed the tolerable values, 10 mg/kg [13].

All tested metal levels in organs of all carp samples were found to be insignificant (P > 0,005) from season to another. Mean concentrations in muscle, and liver of common carp followed the sequences Fe > Zn >Pb> Ni > Cu > Cd, in gills, Zn > Fe >Pb> Ni > Cu > Cd and in gonads Fe > Zn >Pb> Ni > Cu > Cd.

Knowledge of heavy metal concentrations in fish is important for both human consumption and nature management. In this study, we examined metals in tissues of carp, to evaluate heavy metal concentrations in Sikkak dam. It was also aimed to investigate whether metal concentrations varied seasonally in the study. The target organs, such as livers and gills have tendency to accumulate heavy metals in high values, as shown in many species of fishes in different areas [22, 24]. In this study, highest concentrations of Fe were observed in liver of *Cyprinuscarpio*, followed by gonads, by muscle and by gills. The concentrations of metal in gills reflect the concentrations of metal in waters where the fish species live where as the concentrations in liver represent storage of metals [14]. It is well known that induction levels of metallothionein occur in liver tissue of fishes [25]. Metal concentrations in gills could be due to element complexion with the mucus, which is impossible to be completely removed from the lamellae, before tissues are prepared for analysis. The adsorption of metals onto the gills surface, the first target for pollutants in water, may also influence the total metal levels of the gill [26].

Heavy metal concentrations were lower in the muscle compared to gonads gills and liver. Similar results were reported from a number of fish species that the muscle is not an active tissue in accumulating heavy metals [27-28]. The maximum concentrations of cadmium, copper, zinc, and nickel were reached in summer, while their lowest concentrations were in winter and spring for nickel**.** The maximum concentrations of iron were reached in autumn, while their lowest concentrations were in spring. For lead the maximum concentrations were reached in summer, while their lowest concentrations were in spring. The relatively high concentrations of heavy metals in winter were also found in *cyprinuscarpio* in Karakaya dam lake [25], in *stizostedionluciperca* and *Tincatinca in* Kovadalake[1]. The increase of heavy metal levels in summer and winter could be related to increasing physiological activity of fish during summer, primarily caused by the increasing water temperature and decrease in waste water from agricultural activities during winter. For all seasons we found cadmium, lead and zinc concentrations higher than the FAO/WHO values [13], Cd: 0,1 mg/kg Cu: 10 mg/kg, Pb: 0,5 mg/kg, Zn: 50 mg/kg, Ni: 10 mg/kg.

PCA was applied using as variables the mean of the metal concentrations in organs of carp caught seasonally, in order to verify possible bioaccumulation patterns in organs and to detect possible different contamination levels among seasons in the area of study. PCA indicated that both organs and seasons explained significantly 73,43 % of the total variance (44,42% for factor 1 and 29,01 % for factor 2) of the metal concentration (Fig. 3).

IV. CONCLUSION

The dam's water has been observed to deteriorate in quality very rapidly because of the anthropogenic activities. Agricultural wastes are additionally discharged to dam. These may be the possible causes of the high metal amounts mainly dam sediment and biotic components. Levels of heavy metal varied depending on different tissues.

In this study, the effects of the seasons on heavy metal accumulation in organs of *Cyprinuscarpio*were determined. In general, the concentration of iron was observed to be higher in all seasons. For all seasons we found cadmium, lead, and zinc, concentrations higher than the FAO/WHO values [23]. The maximum concentrations of Cd, Cu, Zn and Ni were reached in summer. The present data showed that metal concentrations in livers were generally higher than gills, gonads, and muscles. Liver and gills are metabolically actives tissues and accumulate heavy metals in higher levels. Muscle is not an active tissue in accumulating heavy metals.

From above mentioned result it is clear that the concentration of heavy metals (Cd, Pb and Zn) of Sikkak dam is high. We can conclude there should be effort to protect dam from pollution to reduce environmental risks.

V. Acknowledgments

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FIGURE AND TABLE CAPTIONS

Figure captions

Figure 1: Map of the studied area showing Sikkak dam where samples were collected

Figure 2: Seasonal variations of heavy metals concentrations (Cd, Fe, Cu, Pb, Zn, and Ni) in muscles, gills, gonads, and in the livers

Figure 3: Biplots for first and second axis of the PCA bases on mean values of metal concentrations (Cd, Fe, Cu, Pb, Zn, and Ni) in organs of *Cyprinuscarpio* from Sikkak dam, (M: muscle, Gi: gills, Go: gonads, and L: Liver)

Table Captions:

Table 1: The relationships between weight (W) (g) and total length (L) (mm) of *Cyprinuscarpio* from theSikkak dam at Ainyoucef(Algeria)

Table 2: Mean heavy metal concentrations (mg/kg dry weight) in muscles, gills, gonads and liver of *Cyprinuscarpio* fromSikkak dam, Algeria

Figure 1:

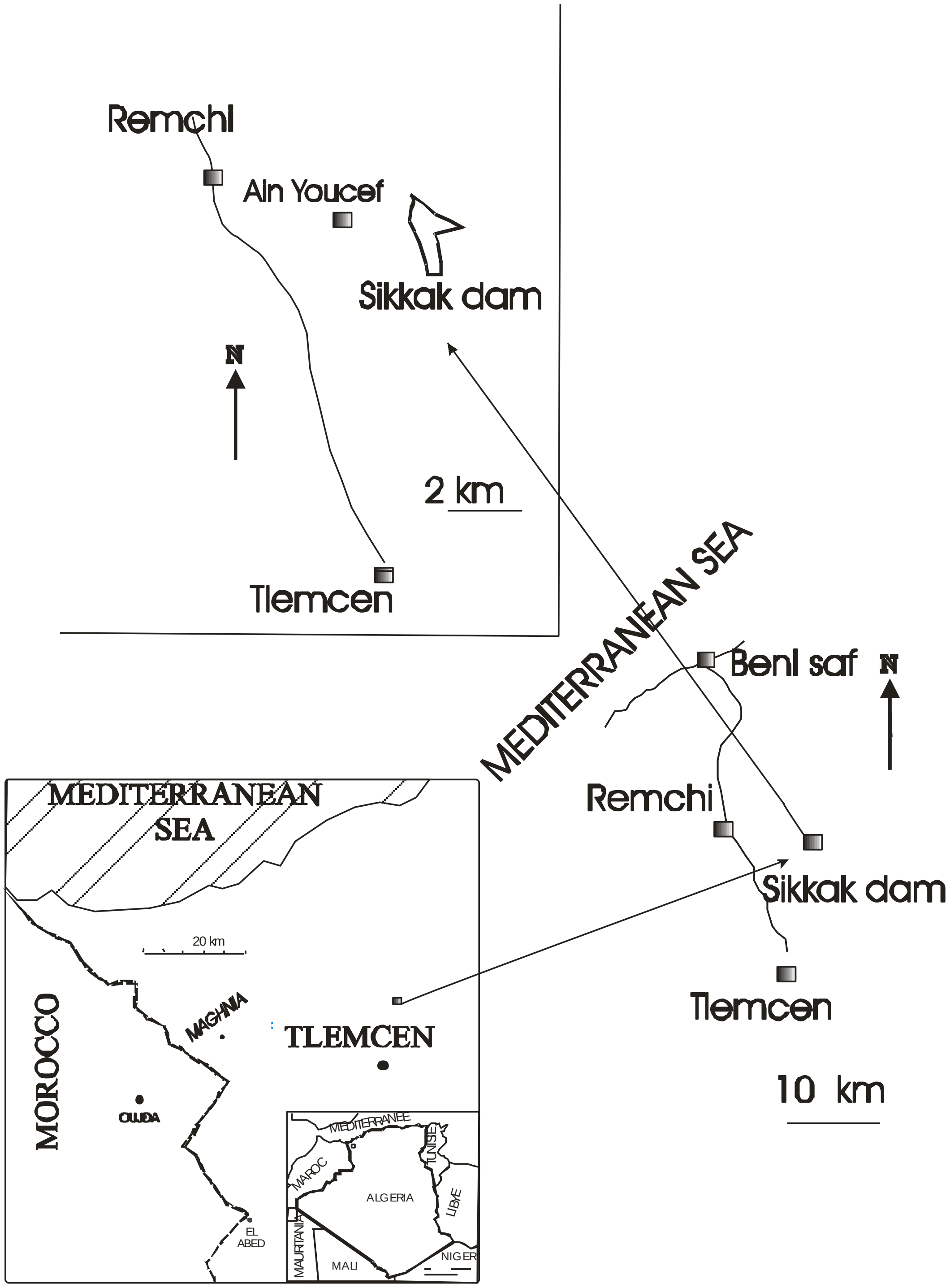
Remchi
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Figure 2:

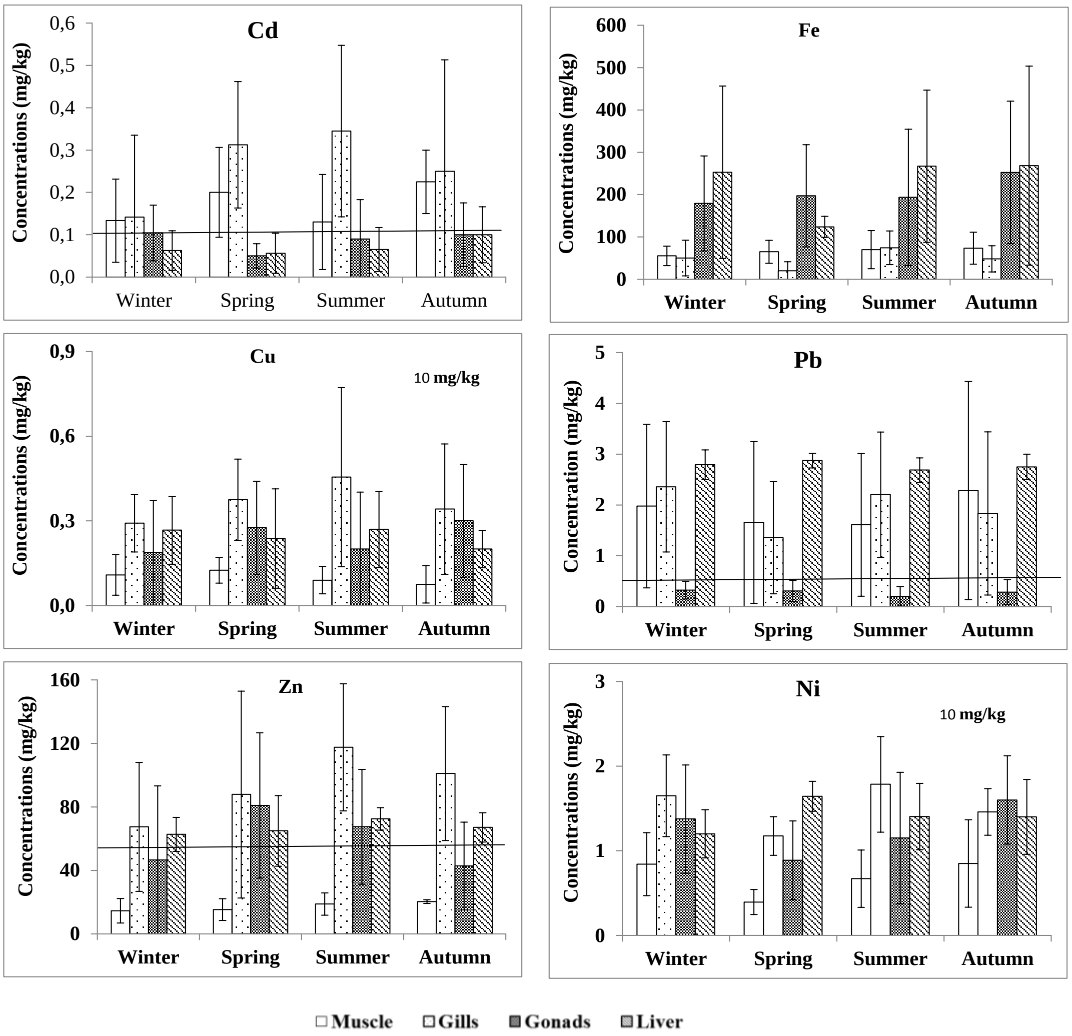

Figure 3 :

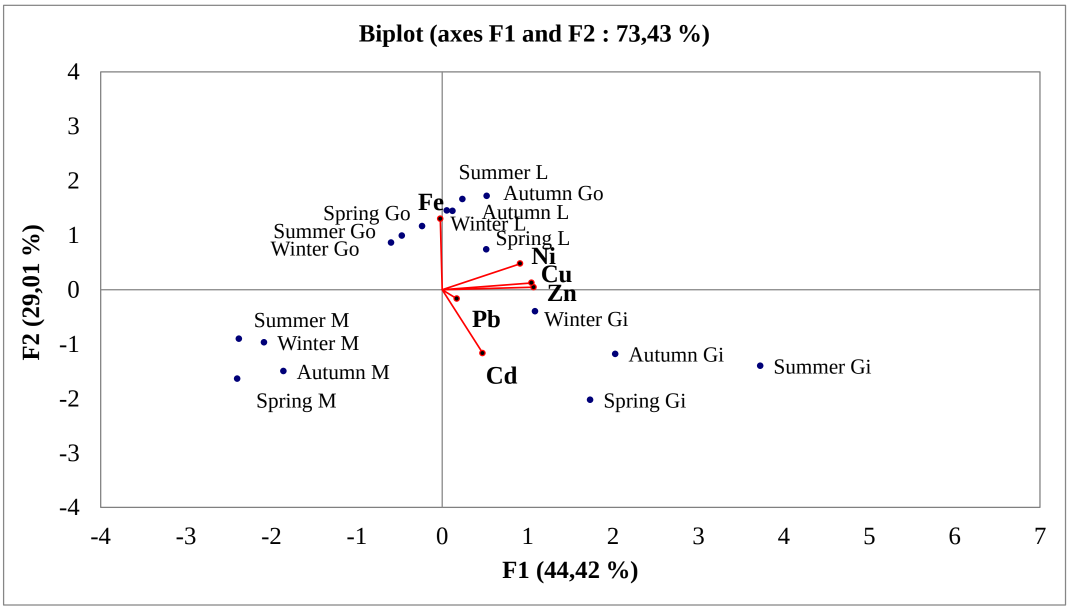

Table 1:

Seasons	L ranges	W ranges	Equation[a]	R value
Winter	18,4-29,1 24,30 ± 4,32	70-230 160,00 ± 66,33	Y = 0,062 X + 14,373	0,952
Spring	15,7-26,7 20,52 ± 4,73	50-220 121,25 ± 73,53	Y = 0,0642 X+ 12,736	0,998
Summer	15,9-25,6 20,32 ± 4,08	50-200 120,00 ± 64,42	Y = 0,0631 X + 12,751	0,995
Autumn	17,1-25,2 20,13 ± 4,42	60-200 110,00 ± 78,10	Y = 0,0557 X +14,011	0,984

[a]Y is total fish length (cm) and X is total weight (g).

Table 2

Organs	Seasons	Cd	Fe	Cu	Pb	Zn	Ni
Muscle	**Winter**	0,13± 0,10	55,29 ± 23,09	0,11 ± 0,07	1,98 ±1,61	14,58 ±7,74	0,84 ± 0,37
	Spring	0,20 ±0,11	64,94 ± 27,17	0,13 ± 0,05	1,66 ±1,59	15,31 ± 6,91	0,39 ± 0,15
	Summer	0,13±0,11	69,70 ± 45,04	0,09 ± 0,05	1,61 ±1,41	18,80 ±7,04	0,67 ± 0,34
	Autumn	0,22±0,08	73,50 ±37,93	0,08 ± 0,07	2,28 ± 2,15	20,33 ±1,18	0,85 ± 0,52
Gills	**Winter**	0,14 ± 0,19	50,08 ±42,15	0,29 ± 0,10	2,36 ± 1,28	67,42 ± 40,59	1,65 ± 0,48
	Spring	0,31± 0,15	19,75 ±21,54	0,38 ± 0,14	1,36 ± 1,11	87,81 ± 65,17	1,18 ± 0,23
	Summer	0,34 ± 0,20	74,20 ± 39,83	0,46 ± 0,32	2,21 ±1,23	117,50 ± 40,06	1,78 ± 0,57
	Autumn	0,25 ± 0,26	48,08 ±30,99	0,34 ± 0,23	1,83 ±1,61	101,00 ± 42,15	1,46 ± 0,28
Gonads	**Winter**	0,10 ±0,07	179,21 ± 112,02	0,19 ± 0,19	0,33 ± 0,17	46,46 ± 46,76	1,38 ± 0,64
	Spring	0,05± 0,03	197,13 ±120,82	0,28 ± 0,17	0,31 ±0,21	80,94 ± 45,76	0,89 ± 0,47
	Summer	0,09 ± 0,09	193,55 ± 161,07	0,20 ± 0,20	0,20 ±0,19	67,45 ± 36,22	1,15 ± 0,78
	Autumn	0,10 ± 0,08	252,42± 168,46	0,30± 0,20	0,28 ±0,25	42,75 ± 27,67	1,60 ± 0,52
Liver	**Winter**	0,06 ± 0,05	252,83±203,90	0,27 ± 0,12	2,79 ± 0,29	62,71 ± 10,73	1,20 ± 0,28
	Spring	0,06 ± 0,05	123,75 ± 25,07	0,24± 0,18	2,88 ± 0,14	64,94 ± 22,25	1,64 ± 0,18
	Summer	0,07 ± 0,05	266,95±179,85	0,27±0,14	2,69±0,24	72,45 ± 7,07	1,41 ± 0,39
	Autumn	0,10 ± 0,07	268,33± 35,23	0,20± 0,07	2,75 ±0,25	67,08 ± 9,21	1,40 ± 0,44