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Accumulation of some heavy metals in the muscles of *Diplodus sargus*, inhabiting El-Mex Bay (Alexandria, Mediterranean Sea) with special references to its physiological responses.

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ABSTRACT

The present study aimed to determine the heavy metals concentrations (cadmium, copper, nickel, lead, iron zinc) in the sediment of El-Mex Bay during the period from December, 2012 to November, 2013; to investigate the qualitative analysis of heavy metals and determination of some biochemical parameters (total proteins, total lipids and total carbohydrates) in the muscles of white seabream, *Diplodus sargus*.

Results revealed that, the highest values of cadmium and copper concentrations in the sediment were recorded during winter and the lowest levels were observed during summer, while the maximum values of iron, lead and nickel occurred during autumn and their minimum values were detected during winter. Zinc ion concentration showed a high peak during spring and declined during autumn.

Data revealed that, the highest values of cadmium, lead, nickel and zinc in the muscles of small and large fish, *D. sargus* were recorded during autumn and their lowest values were detected during summer, except zinc; its lowest level was observed during spring in the two sizes. Concerning copper and iron, their maximum concentrations were measured during summer in the two sizes and the minimum occurred during winter. Biochemical analysis indicated devastating effects in metabolic parameters at the high bio-concentrations of metals through the different seasons. ANOVA ($p< 0.05$) showed significant differences between the different seasons and parameters. Also, there was a non-significant difference between the two sized groups.

1. INTRODUCTION

El-Mex Bay (Fig. 1) is a part of Alexandria Coast on the Mediterranean Sea, is adjacent to Alexandria City Centre and extends for about 15 km between El-Agami headland and the Western Harbour and from the coast to a depth line of about 15 meters (Said *et al*., 1994). The Bay is elliptical shape in the west of Alexandria, its surface area is about 19.4 km^2 and has volume about 190.30 x 10^6 m^3 (Mikhail, 2008).

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The natural water bodies may extensively be contaminated with various heavy metals released from industrial and mining effluents, combustion of fossil fuels, discharge of sewage and sewage sludge; fertilizers and residues of domestic & pesticides, dumping of hospital and anthropogenic activities, etc. (Laxmi-Priya *et al*., 2011; Ghanem, 2014).

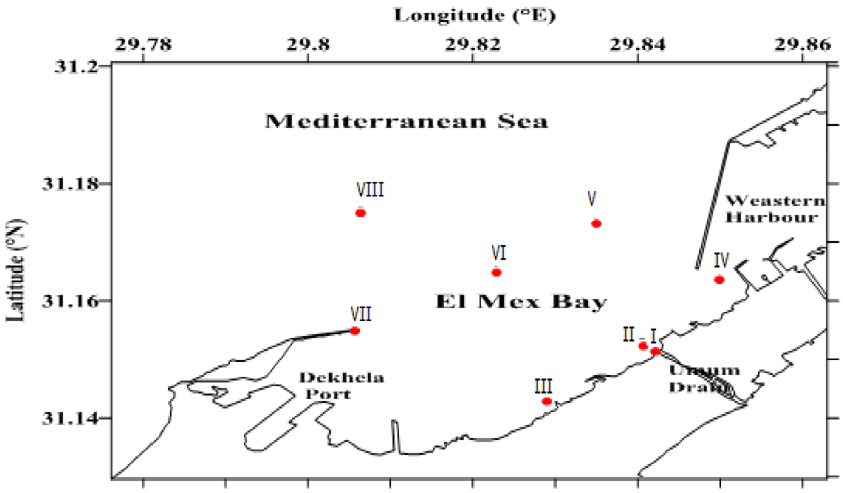

Fig.1: Map of El-Mex Bay showing the study area.

The area of El-Umum drain reflects the effects of discharged materials which thus shift the sediment towards a finer grades character (Younis, 2005). Heavy metal contamination may cause devastating effects on the ecological balance of the recipient environment and its diversity of aquatic organisms (Vinodhini & Narayanan, 2008; Ghanem, 2014).

Traces of heavy metals play a biochemical role in the life processes of some aquatic plants and animals, while it becomes toxic when it is present at high concentrations (Kotickhoff, 1983). Fish are often the top of aquatic food chain and is an important source of protein for human who may absorb large amounts of some metals such as cadmium, copper, iron, nickel, lead and zinc. Low concentrations of heavy metals may not kill individuals of the fish but affect their size, reproduction and body weights, thus reducing their ability to compete for food and habitat which affect directly the metabolic and enzymes activities correlated with changes in the rate of protein synthesis (Vosyliene & Jankaite, 2006; Jovanovic *et al*., 2011; Ghanem, 2014). Feeding behavior of the fish in El-Mex Bay may influence metabolic process i.e. the fishes are mainly bottom feeder; the direct contact with sediment may cause organ dysfunction depending on the degree of sediment contamination (Khalaf-Allah & Shehata, 2011).

Heavy metals have badly influence on blood parameters of living organisms and lead to haematological disorders and cause oxidative stress and may on cell components due to devastating effects (Yapici *et al*., 2006). The trace metals are uptake more rapidly in high temperature by marine organisms (Raymont and Shields, 1994).

Muscles is one of the most organs which are varied in composition according to the species, sex and maturity as well as seasons (Rubbi *et al*., 1985). Biochemical and physiological biomarkers are frequently used for detecting or diagnosing the harmful effects in fish exposed to different toxic substances. Therefore, in the present study attempts have been made to assess the heavy metals concentrations in the sediment and fish muscles collecting from El-Mex Bay and their effects on biochemical parameters in an important commercial species feeding on the substrate in fauna.

2. MATERIALS AND METHODS

2.1 Determination of heavy metals in the sediment:

Sediment samples were collected seasonally from the bay using Van Veen, grab. After collection, the sediment samples were transferred into the laboratory in plastic bags. Concentrations of heavy metals in the sediment samples were determined using the method described by Oregioni and Aston (1984). The results were expressed by µg/g dry weight.

2.2 Determination of heavy metals in the muscles of the fish:

2.2.1 Specimens collection:

Samples of *Diplodus sargus*, (Fig. 2) were collected during the period from December, 2012 to November, 2013 by using gill net and encircling net. After collection, fish were freshly examined or immediately preserved in an ice box and transferred to the laboratory for latter examination. In the laboratory, standard and total length of each fish were measured to the nearest centimeter, while the body weight was determined to the nearest gram. The fish specimens were selected carefully to cover two sized groups. One of these was small sized groups (12.00-17.00 cm) and the second was large sized groups (17.10-27.00 cm). After dissection of the collected fish samples, a known weight of the muscles was kept under freezing condition at 4°C until latter examinations.

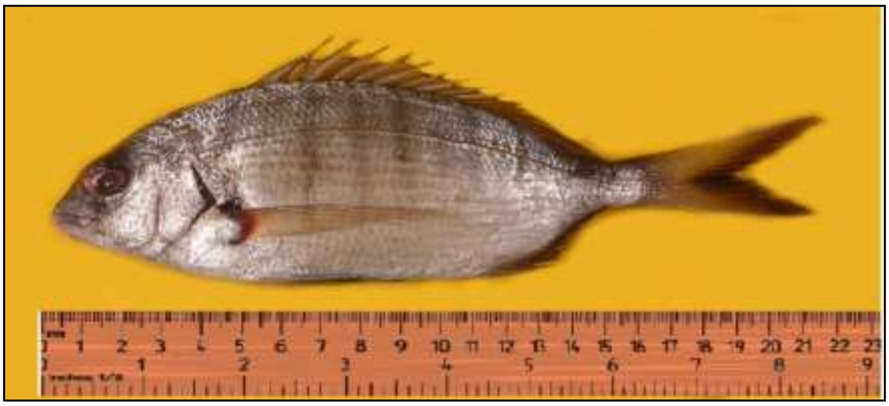

Fig. 2: Lateral view of *Diplodus sargus*, collected from El-Mex Bay, during 2012-2013.

2.2.2 Concentration of heavy metals in the fish muscles were measured according to APHA (1992). Results were expressed by µg/g wet weight.

2.2.2.1 Physiological studies:

After the dissection of the collected fish, a known weight of the muscles was homogenized by using the electric homogenizer for 2 min. The homogenate specimens were centrifuged at 4000 r.p.m. for 15 min. at 2 ^{o}C in a refrigerator centrifuge. The supernatant solution was used directly or stored at 4 C^{o} until the use for the biochemical analysis.

Total protein content was determined according to Doumas (1975), while total lipids were detected according to the method of Knight *et al*. (1972) by using a kit of Bioadwic Company. Total carbohydrates content were determined according to the method of Singh and Sinha (1977). Results were expressed by g/100 g wet weight.

2.2.2.2 Statistical analysis:

Results were expressed in tables as mean ±S.D. Data were analyzed by using analysis of variance (ANOVA) for biochemical parameters and correlation coefficient for concentration of heavy metals in the sediment and muscles according to Bailey (1981).

3. RESULTS AND DISCUSSION

3.1 Heavy metals determinations:

Sediment is a very important sector to ecologists because it gives a good indication in the environmental changes of water and aquatic organisms. It is important host for different pollutants and tends to concentrate the heavy metals and other organic pollutants (Saloman *et al.,* 1987). It plays a very important role in physicochemical and ecological dynamics; any changes in toxic concentrations of heavy metal residues in the sediments will affect the natural aquatic life (Jumbe and Nandini, 2009). Aquatic animals can accumulate the metals by ingestion of particulate materials suspended in the water, ingestion of food and adsorption of tissue (Hassouna, 1989).

The present study attempts to create awareness concerning the potential severe public health issues resulting from the toxic effects of heavy metals as pollutants on invertebrates which considered the important food source for *Diplodus sargus*.

Bioaccumulation of heavy metals and consequent alterations in the muscles of most economic fish were examined.

Results (Table 1) revealed that, the maximum value of cadmium ion concentration in sediment of the bay was recorded during winter (5.82 µg/g dry wt.) and the minimum one (3.60 µg/g dry wt.) was observed during autumn, with an annual average of 4.52±1.11 µg/g dry wt. However, the concentrations of copper ion in the bay sediment were fluctuated between 39.88 µg/g dry wt. during summer and 55.32 µg/g dry wt. during winter, with an annual average of 48.22±6.49 µg/g dry wt. (Table 1). Detection of the lowest values during autumn may be due to the low amounts of discharged wastes and the highest one during winter may be attributed to the increasing of discharged waste into the bay. These observations agree with Mostafa *et al.* (2004); Masoud (2007); Abdallah (2008); Masoud *et al.* (2012) and disagree with Vukovic *et al*. (2011) who found that, the highest value of this ion occurred during autumn.

Table 1: Seasonal variations of heavy metals concentration (µg/g dry wt.) in the sediment of El-Mex Bay, during 2012- 2013.

Seasons / Metals	Winter	Spring	Summer	Autumn	Annual average
Cd	5.82	5.07	3.58	3.60	4.52±1.11
Cu	55.32	50.55	39.88	47.13	48.22±6.49
Fe	1573.33	1617.83	1616.83	1619.00	1606.75±22.30
Pb	98.55	106.00	114.22	137.40	114.04±16.84
Ni	78.87	86.38	95.03	105.95	91.56±11.65
Zn	161.00	190.15	161.42	122.98	158.89±27.55

Regarding iron ion concentration (Table 1), the highest value of iron ion concentration was recorded during autumn (1619.00 µg/g dry wt.). While, the lowest value was detected during winter (1573.33 µg/g dry wt.), with an annual average of 1606.75±22.30 µg/g dry wt. The large amounts of wastes and organic matter that reached to the bay from El-Umum Drain may be associated with iron and deposited on the substrate. These processes could explain the high iron contents in the sediments of El-Mex region.

These findings were agree with Masoud *et al.* (2012) and differ with El-Enany (2004) who reported that, the highest value of iron ion may be due to the increasing of wastes discharged into this area which considered the main source of metals adsorption of iron on the sediment as iron oxide.

In the present study, it is clear that, lead ion concentrations (Table 1) in the sediment of the bay are varied considerably from season to season. Its concentration in the sediment was peaked during autumn (137.44

µg/g dry wt.) and declined to 98.55 µg/g dry wt. during winter, with an annual average of 114.04±16.84 µg/g dry wt. The maximum value of lead ion concentration in the sediment of El-Mex Bay may be attributed to the presence of different effluents from El-Umum drain. This observation was agreed with Beukemo *et al.* (1986) who reported that, agricultural wastes enriched the sediment with suspended organic matter; humic acids acquire a large fraction of Pb burden from the overlying water through suspended particles. Additional amounts deposited after the humic acids are buried in the sediments.

Moreover, the concentration of nickel ion in the sediment was varied from 78.87 µg/g dry wt. during winter to 105.95 µg/g dry wt. during autumn, with an annual average of 91.56±11.65 µg/g dry wt. (Table 1). The higher concentration of nickel ion in the sediment was associated with its concentration in the water follow and different sewage effluents from El-Umum drain. These results are agreed with Ali (2013); Ibrahim & Omar (2013), while differ with Gueddah and Djebar (2014) who found that, the concentrations of nickel ion are lower than that recorded in the obvious data.

Concerning zinc ion in the sediment, its maximum value (190.15 µg/g dry wt.) was observed during spring and the minimum (122.98 µg/g dry wt.) occurred during autumn, with an annual average of 158.89±27.55 µg/g dry wt. (Table 1). The highest value of zinc ion concentration may be due to the continuous discharges through El-Mex pumping station or attributed to the highest amounts of organic matter in the silt and clay sediments at this area. The low dissolved oxygen may also contribute to the increasing rate of zinc sedimentation as ZnS, which is favored only under anaerobic conditions. Similar observations were mentioned by Goher (1998).

The present data (Table 2) showed that, correlation coefficient between different metals in the sediment indicated to a positive correlation in the case of interaction between cadmium with copper& zinc; copper with zinc; iron with lead & nickel and the case of lead with nickel.

Metals such as iron, copper, zinc, lead, cadmium and others when discharged into water can enter the food chain and bioaccumulated in the fish tissue and hence become a threat to human (Ajmal *et al.*, 1985).

Table 2: Correlation coefficient between different heavy metals in the sediment of El-Mex Bay.

Metals	Cd	Cu	Fe	Pb	Ni	Zn
Cd	1					
Cu	0.891	1				
Fe	-0.780	-0.711	1			
Pb	-0.822	-0.474	0.636	1		
Ni	-0.922	-0.647	0.742	0.977	1	
Zn	0.550	0.197	-0.075	-0.807	-0.723	1

On the other hand, some heavy metals are essential for the different metabolic processes. But they are highly toxic for aquatic organisms and their consumers when exceeding the recommended safety levels (Zaghloul, 2000). Bioaccumulation of metals in the different tissues was varied from metal to another, differs in various organisms and from organ to another in the same organism (Al-Ghamdi and Al-Mohanna, 1993). Feeding behavior and the direct contact with contaminated sediment may influence the metabolic process causing organ dysfunction (Khalaf-Allah & Shehata, 2011).

The present study (Table 3) revealed that, the maximum average value of cadmium ion concentrations in the muscles of small size *D. sargus* (0.80 µg/g wet wt.) was detected during autumn and the lowest

(0.45 µg/g wet wt.) was observed during summer. During winter and spring, however, it was ranged between 0.60 µg/g wet wt. in the former and 0.75 µg/g wet wt. in the latter. In the large fish, however, cadmium ion concentration in the muscles was increased during autumn (0.95 µg/g wet wt). It decreased gradually during spring (0.70 µg/g wet wt.) through winter (0.65 µg/g wet wt.) and reached its minimum value (0.55 µg/g wet wt) during summer.

Results showed that, cadmium ion concentrations in the muscles of this fish increase with increasing length groups and decreasing in temperature. This finding agrees with Ghanem (2011) who concluded that, it increased with the increasing in length groups, at very high concentrations of metals in the different tissues with size-dependent. On the other hand, it differ with Al-Ghamdi & Al-Mohanna (1993) whom reported that, its level were decreased with the increasing length groups according to that, the uptake of metals is influenced by many factors including fish species, age, type of fish organs and various environmental factors.

Regarding copper ion concentrations in the small size, its lowest value was recorded during winter and the highest value was determined during summer; being 2.05 µg/g wet wt. and 3.90 µg/g wet wt, respectively. However, it was ranged between 2.75 µg/g wet wt. during winter and 4.95 µg/g wet wt. during summer in the large group. At the remaining seasons, however, small fish showed a nearly similar concentration; being 2.40 µg/g wet wt. during spring and 2.70 µg/g wet wt. during autumn. In the large one, however, it was varied from 2.85 µg/g wet wt. during spring to 4.75 µg/g wet wt. during autumn. Results indicated that, the copper ion concentrations in the muscles of large fish are relatively higher than the small ones along the year. These findings were in agreement with Ghanem (2011 and 2014) who concluded that, the concentrations of heavy metals in the fish tissues increases with the increasing age of the fish and with increasing temperature and differ with Al-Ghamdi & Al-Mohanna (1993); Shakweer & Abbass (1997) whom concluded that, the concentration of heavy metals were decreased with the increasing the fish size.

On the other hand, the minimum average value of iron ion concentration in the muscles of small fish was recorded during winter (70.85 µg/g wet wt.) and the maximum (85.20 µg/g wet wt.) was observed during summer. In the large size, however, it was fluctuated between 83.00 µg/g wet wt. during winter and 98.00, during summer. At the remaining seasons, it was varied from 75.40 µg/g wet wt. during spring in the small size to 95.55 during the same season in the large ones. Results declared that, iron ion concentrations in the muscles of small fish are relatively lower than the large one during the year and increased with increasing temperature (Table 3). From the above findings, it can be concluded that, iron ions in the muscles of *D. sargus* increases with the increasing age groups and water temperature. These observations agree with Raymont & Shields (1994); Ghanem (2014) who reported that, the trace metals are uptake more rapidly at high temperature by marine organisms specially iron ion concentration in the muscles of *D. sargus* inhabiting Mediterranean Coast and disagree with Said & El-Agroudy (2003); Ghanem (2011) whom attributed that, metal concentration in the soft tissues increases with the decreasing of water temperature

Furthermore, the minimum average value of lead ion concentration in the small fish muscles of *D. sargus* was observed during spring and summer and the maximum value was determined during winter and autumn; being 1.70µg/g wet wt. and 2.70 µg/g wet wt, respectively. In large fish, however, the highest value (4.90 µg/g wet wt.) was recorded during autumn and decreased gradually during spring (4.10 µg/g wet wt.) followed by 4.00 µg/g wet wt. during winter and reached its lowest one (2.95 µg/g wet wt) during summer. Results revealed that the lead ion concentrations in the muscles of large fish are relatively higher than the small one along the year and increased with temperature (Table 3). These observations agree with Said & El-Agroudy (2003); Ghanem (2011) whom attributed that metal concentration in the soft tissues increases with the decreasing of water temperature and disagree with Raymont & Shields (1994); Ghanem (2014) who reported that, the trace metals are uptake more rapidly at high temperature by marine organisms.

The present study (Table 3) declared that, nickel ion concentration showed a lower peak in the muscles of small and large fish, *D. sargus* during summer; being 28.70 µg/g wet wt. and 31.15 µg/g wet wt, respectively and the highest peak were observed during autumn in the small fish (30.85 µg/g wet wt.) and during autumn & spring in the large one (39.20 µg/g wet wt. & 41.05 µg/g wet wt., respectively). At the remaining seasons, it was fluctuated between 29.50 µg/g wet wt. during winter in the small fish and 32.25 µg/g wet wt. during the same season in the large one.

Table 3: Seasonal variations of heavy metals concentration (µg/g wet wt.) in the muscles of *Diplodus sargus,* collected from El-Mex Bay area, during 2012 - 2013.

Metals \ Seasons		Winter	Spring	Summer	Autumn
Cd	Small fish	0.60	0.75	0.45	0.80
	Large fish	0.65	0.70	0.55	0.95
Cu	Small fish	2.05	2.40	3.90	2.70
	Large fish	2.75	2.85	4.95	4.75
Fe	Small fish	70.85	75.40	85.20	75.70
	Large fish	83.00	95.55	98.00	90.80
Pb	Small fish	2.70	1.70	1.70	2.70
	Large fish	4.00	4.10	2.95	4.90
Ni	Small fish	29.50	30.45	28.70	30.85
	Large fish	32.25	41.05	31.15	39.20
Zn	Small fish	17.30	13.55	14.35	25.85
	Large fish	19.70	13.90	30.05	41.75

Nickel ion concentrations in the muscles of large fish were relatively higher than the small one along the year specially the cold weather. Similar observations obtained by Said & El-Agroudy (2003) and was different with Bahnasawy (2001); Radwan &Lotfy (2002); Ghanem (2014) whom stated that, the concentrations of nickel increased during the hot weather.

Concerning to zinc ion concentration in the muscles of *D. sargus*, the depletion was shown during spring and increased to reach its highest value during autumn; being 13.55 µg/g wet wt. & 25.85 µg/g wet wt., respectively in the small size and being;. 13.90 µg/g wet wt. & 41.75µg/g wet wt., respectively in the large one. At the remaining seasons, it was ranged between 14.35 µg/g wet wt. during summer in the small sized group and 30.05 µg/g wet wt. during the same season in the large one. Data indicated that, zinc ion concentrations in the muscles of large fish are relatively higher than the small one. In the present study, the concentrations of zinc ion in the muscles of *D. sargus* were increased with increasing length of the fish. A higher level of zinc ion may be due to the increase of heavy metals in the drainage waters during the same season, decomposition of organic matter and discharge remnants of fertilizer factories and other chemicals lead to these facts; the presence of one metal deletes the accumulation of another metal; the uptake of metals is influenced by many factors including fish species, age, type of fish organs, season and various environmental factors. Similar observation was detected by Ptashynski & Klaverkamp (2002); Ghanem (2014) and differ with that obtained by Falandysz (1992) who stated that, the concentrations of zinc in the muscles of fish decreases with the increasing length of the fish. However, Yacoub and Gad (2012) reported that, cold season exhibited the high level of this metal than the hot one. Also, Said & El-Agroudy (2003); El-Serafy *et al*. (2003) mentioned that, no marked seasonal variations in the concentration of metals in *Patella Caerulea* lived in polluted area of Alexandria Coast were detected.

Bio-sedimentation of heavy metals in the muscles of *Diplodus sargus* collected from El-Mex Bay, during the study period indicated a high sedimentation rate was recorded in the case of nickel ion at the small and large fishes, while, its low rate was observed in the case of lead at the different sized groups (Fig. 3). Similar observation was obtained by Ali *et al*. (2011); Ibrahim & Omar (2013) whom found that, the highest bio-sediment accumulation factor was obtained for Ni, which could be explained by higher levels of these metals in the sediment.

Correlation coefficient between different metals in the sediment and muscles of the different sized groups exhibited that, it was a negative correlation in the case of copper and zinc between the sediment and small fish muscles. Furthermore, it was positive correlation in the case of iron, lead and nickel between the sediment and two sized groups (Table 4).

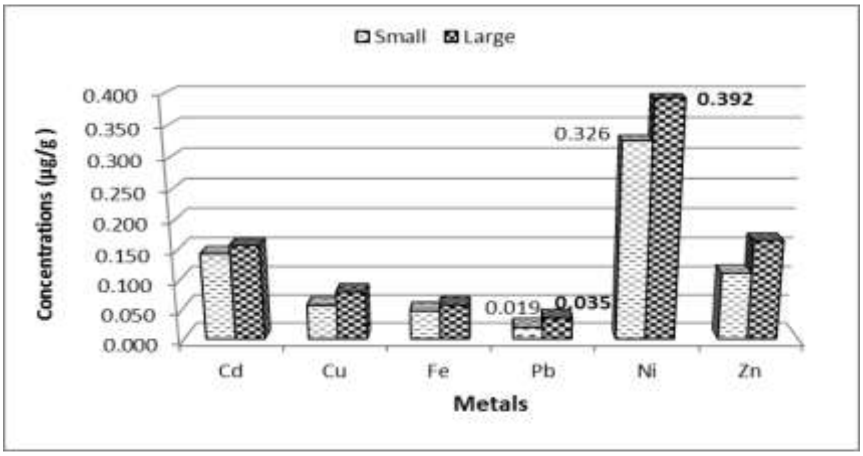

Fig. 3: Bio-sedimentation of heavy metals in the muscles of ***Diplodus sargus***, collected from El-Mex Bay, during 2012- 2013.

Table 4: Correlation coefficient between different heavy metals in the sediment and muscles of *Diplodus sargus,* collected from El-Mex Bay.

Sized groups / *Sediment*	*Small fish*	*Large fish*
Cd	0.074	-0.272
Cu	**-0.979**	**-0.877**
Fe	**0.631**	**0.873**
Pb	0.270	0.496
Ni	0.347	0.293
Zn	**-0.928**	**-0.935**

3.2 Biochemical studies:

Muscle compositions are varied according to the species, sex and maturity as well as seasons (Rubbi *et al*., 1985). The present results (Table 5) indicated that, the highest values of total proteins in the muscles of small and large fish, *D. sargus* were determined during spring and the lowest values were detected during winter; being 39.79±1.21 g/100 g wet wt. & 38.99±6.10 g/100 g wet wt., respectively in the first season and 28.86±2.57 g/100 g wet wt. & 29.55±0.60 g/100 g wet wt., respectively in the second one. At the remaining seasons, it was varied from 32.84±1.30 g/100 g wet wt. during summer in the small size and 37.61±1.82 g/100 g wet wt. during autumn in the same group.

The depletion in proteins content during winter may be due to the lower in fish activity that depends on water temperature and their distribution was limited by their thermal tolerance. In addition to the changes in water quality by the action of heavy metals dissolved in agriculture drainage waters and humic matter that may critically influenced the growth rate and quality of the fishes; which feed on biota induced by bioaccumulation of metals. This may be explained as the exposure to metals may lead to high accumulation in muscles sharp reduction in the metabolic rate of fish and consequently decrease protein contents in tissues. These results agree with Khalil & Hussein (1997) Fayed *et al.* (2001); Ghanem (2014) and differ with Kaur & Saxena (2001); Abdel-Tawwab *et al*. (2010) whom concluded that the depletions in proteins content were observed in the muscles of large size, due to the protein utilization decrease with the increasing age and dietary of protein levels.

Table 5: Seasonal variations of biochemical parameters (g / 100 g wet weight) in the muscles of *D. sargus,* collected from El-Mex Bay, during 2012- 2013.

Parameters \ **Seasons**		**Winter**	**Spring**	**Summer**	**Autumn**
Total protein	Small fish	28.86±2.57	39.79±1.21	32.84±1.30	37.61±1.82
	Large fish	29.55±0.60	38.99±6.10	37.21±1.64	35.72±3.22
Total lipid	Small fish	2.04±0.83	2.74±1.91	3.19±0.49	2.79±1.92
	Large fish	2.55±1.54	3.51±0.61	2.65±0.82	3.13±0.78
T. carbohydrates	Small fish	0.37±0.01	0.41±0.02	0.37±0.02	0.32±0.01
	Large fish	0.39±0.01	0.43±0.02	0.42±0.03	0.35±0.02

However, the elevation in total proteins was recorded during spring as results to suitable temperature and food availability, in addition to type of food, fish size and maturity stages. Total protein contents in the study fish were increased with the increasing in length groups. This results agree with Shakweer & Abbas (2005); Ghanem (2011) and differ with Sancho *et al.* (1998) who mentioned that, total proteins in the different organs of immature and mature fish were nearly similar.

Concerning total lipids, the maximum value of total lipids in the muscles of small fish, *D. sargus* was observed during summer and the minimum was detected during winter; being 3.19±0.49 g/100 g wet wt. and 2.04±0.83 g/100 g wet wt., respectively. Moreover, total lipids percentage was increased from 2.55±1.54 g/100 g wet wt. during winter to 3.51±0.61 g/100 g wet wt. during spring in the large group. At the remaining seasons, it was varied from 2.74±1.91 g/100 g wet wt. during spring to 2.79±1.92 g/100 g wet wt. during autumn in the small size. While, it was fluctuated between 2.65±0.82 g/100 g wet wt. during summer and 3.13±0.78 g/100 g wet wt. during autumn in the large ones (Table 5).

In the present data, total lipids in the muscles of *D. sargus* were slightly increased during spring in the large fish and summer in the small ones. The depletion of total lipids in the muscles during winter may be due to the use of energy-rich lipids for energy production during toxic stress from one or more of metals. Similar observations were recorded by Sancho *et al*. (1998); Blaner *et al*. (2005); Ghanem (2011) whom reported that, the smaller sized group had lower level of fat than the larger one and differ with Virk & Sharma (1999). Total lipids in the fish tissues reached the maximum level during spring and summer. This may be due to the favorable climatic condition, the availability of more amount of food consumed by the fish and increase the feeding intensity of the fish during this period. These results agree with Jain & Singh (1987).

The high peak of total carbohydrates in the muscles of small sized group of *D. sargus* was detected during spring and the minimum was measured during autumn; being 0.41±0.02 g/100 g wet wt. and 0.32±0.01 g/100 g wet wt. respectively. In the large fish, however, it was varied from 0.35±0.02 g/100 g wet wt. during autumn to 0.43±0.02 g/100 g wet wt. during spring. At the remaining seasons, it was nearly similar; being 0.37±0.01 g/100 g wet wt. during winter and 0.37±0.02 g/100 g wet wt. during summer in the small sized group. However, it was fluctuated between 0.39±0.01 g/100 g wet wt. during winter and 0.42±0.03 g/100 g wet wt. during summer in the large one.

The high level of carbohydrate content was found during spring; may be due to the favourable temperature and high feeding activity. This result agrees with Bumb (1992) who recorded the variation of carbohydrate content with feed intake and intensive of feeding in *Ambassis commersoni* coincides with the occurrence of high carbohydrate content in the muscle of fish. However, the low levels of total carbohydrates may be due to the fact that they does not contribute much to the reserves in the body. This depletion in glycogen level may be due to its utilization for supplying the energy to the fish under stress condition leading to retardation of growth and alter the physiological mechanism (Ghanem, 2011; Mathana *et al.*, 2012).

From the above mentioned results, total proteins, total lipids and total carbohydrates in the muscles of the large fish are relatively higher than the small group during the year except autumn. Statistically (Table 6), ANOVA showed that, there are highly significant differences ($p<0.01$) between the parameters and seasons. However, it was non-significance difference between the different sizes. Moreover, two ways of ANOVA appeared that, there were non-significant differences between the interaction of size groups and parameters at the same season and between the different sizes groups and seasons at the same parameter. On the other hand, the interaction between seasons and parameters at the same size indicated a significant difference ($p<0.05$).

Table 6: Analysis of variance (ANOVA), on physiological parameters in the different sizes of *Diplodus sargus,* during 2012-2013.

Source of variance	D.F.	S.S.	M.S.	F.V.
Seasons	3	38.24	12.75	9.45**
Sizes	1	0.97	0.97	0.72 n.s.
Parameters	2	5913.63	2956.82	2191.14**
Seasons* Sizes	3	2.45	0.82	0.60 n.s.
Seasons* Parameters	6	58.83	9.80	7.27
Sizes* Parameters	2	0.83	0.42	0.31 n.s.
Errors	23	6023.04	261.87	--
Total	6	8.10	1.35	--

Note: * = Significant at $p<0.05$.
** = Significant at $p<0.01$.
n.s. = non-significant.

4. REFERENCES

Abdallah, A. M. (2008). Trace element levels in some commercially valuable fish species from coastal waters of Mediterranean Sea, Egypt., Journal of Marine Systems,. 73(1-2):114-122.

Abdel-Tawwab, M.; Ahmed, M. H.; Khattab, Y. A. E. and Shalaby, A. M. E. (2010). Effect of dietary protein level, initial body weight and their interaction on the growth, fed utilization and physiological alterations of Nile tilapia, *Oreochromis niloticus*. Aquacult., 298: 267-274.

Ajmal, M.; Khan, M. A. and Nomani, A. A. (1985). Distribution of heavy metals in plants and fish of the Yamuna River, India. Environ. Monit. Assess., (5): 361 – 367.

Al-Ghamdi, H. S. and Al-Mohanna, M. M. (1993). Determination of some heavy metals in five fish species imported to Saudi Arabia for human consumption. Proc. Zool. Soc. A. R. E., 24: 249-257.

Ali, D.; Alireza, N.; Seyed, M.; Alireza, S. and Mehrdad, K. (2011). Environmental monitoring of heavy metals in seaweed and associated sediment from the strait of Hormuz, I.R. Iran., World Journal of Fish and Marine Sciences, 3 (6): 576-589.

Ali, D. and Masoud, N. (2013). Heavy metals contamination in sediments from the north of the Strait of Hormuz**.,** Journal of the Persian Gulf (Marine Science)., 4(11): 39-46.

A.P.H.A. (American Public Health and Associated) (1992). Standard methods of the examination of water and wastewater. 18th ed Washington D.C. 1015 pp.

Bahnasawy, M. H. (2001). Levels of heavy metals in catfish, *Clarias gariepinus*, from different habitats and their effects on some biochemical parameters. Egypt. J. Aquat. Boil. & Fish., 5(1): 99-125.

Bailey, N. T. J. (1981). Statistical methods in biology. 2nd Ed. (Biological Science Texts): 216pp.

Beukemo, A. A. Hekestra, G. P. and Venema, C. (1986). Environmental Monitoring and Assessments., 7: 117.

Blaner, C.; Curitis, M. and Chan, H. (2005). Growth, nutritional composition and hematology of Artic chair (*Salvelinus alpinus*) exposed to toxaphene and tapeworm (*Diphyllobothrium dendritiam*), Archive of Environmental Contamination and Toxicology., 48: 397-404.

Bumb, S. (1992). Studies on the biology of Commersoni's Glassy perchlet, *Ambassis commersonii*. (Cuvier). Ph.D Thesis, Goa University, 214 pp.

Doumas, B. T. (1975). Colorimetric determination of total protein in serum or plasma. Clin. Chem., 21(8): 1159-1166.

El-Enany, H. R. (2004). Ecological and biological studies on Lake Manzalah with special reference to their water quality and sediment productivity. M. Sc.Thesis, Fac. Sci., Al-Azhar Univ.

El-Serafy, S. S.; El-Gamal, M. M. and El-Sayed, D. S. (2003). Seasonal variations of trace metals levels in water and the limpet, *Patella caerulea*, of Alexandria coast, Egypt. Egypt. J. Aquat. Boil. & Fish., 7(4): 283-312.

Falandysz, J. (1992). Metal content of muscles of herring taken from the Gulf of Gdansk. Stud. Mater. Oceano., 62: 19-26.

Fayed, H. M.; Zaghloul, K. H.; Abdel-Monem, S. and Mohamed, H. A. (2001). Biological responses of the Nile Tilapia, *Oreochromis niloticus*, and the Nile catfish, *Clarias gariepinus*, induced by agricultural and industrial pollutants. J. Union Arab Biol., 16(A): 543-568.

Ghanem, M. H. M. (2011). Seasonal variations of total proteins, lipids and carbohydrates in cultivated, brackish and salt water fishes with special references to their nutrient values. .Ph. D. Thesis, faculty of Science, Al-Azhar Univ., Cairo., 395pp.

Ghanem, M. H. M. (2014). Effect of heavy metals on some physiological responses in two fish species inhabiting Mediterranean Sea Coast; Damietta Governorate, Egypt., Al-Azhar Bull. Sci., 25(1):1-14.

Goher, M.A. (1998). Factors affecting the precipitation and dissolution of some chemical elements in River Nile at Damitta Branch. M.Sc. Thesis, Faculty of Science, Menofiya University, Egypt.

Gueddah, D. and Djebar, A. B. (2014). Pollution by the heavy metals of the superficial marine sediment of Skikda city (Northeastern Algeria)Annals of Biological Research, 5 (1):40-45.

Hassouna, A. F. A. (1989). Some analytical study on sediments on Lake Nasser, River Nile and Aswan Dam Zone Reservoir. M. Sc. Thesis, Fac. of Sci., Al-Azhar Univ.

Ibrahim, A. Th. and Omar, H. M. (2013). Seasonal variation of heavy metals accumulation in muscles of the African

Catfish, *Clarias gariepinus* and in River Nile water and sediments at Assiut Governorate, Egypt., J Biol Earth Sci., 3(2): 236-248.

Jain, K. F. and Singh, C. P. (1987). Monthly variation in the fat content of a Carp, *Labeo gonius* (Ham). Comp. Phvsiol. Ecol., 12(2): 100-1 02.

Jovanovic B.; Mihaljev, E.; Maletin, S. and Palic, D. (2011). Assessment of heavy metal load in chub liver (Cyprinida: *Leuciscus cephalus*) from the Nisava River (Serbia), Biologica Nyssana., 2(1):1-7.

Jumbe, A. S. and Nandini, N. (2009).Heavy metals analysis and sediment quality values in Urban Lakes. American J. of Environ. Sci., 5 (6): 678-687.

Kaur, T. and Saxena, P. K. (2001). Impact of pollution on the flesh of some fishes inhabiting River Satluj waters' a biochemical study, Indian J. Environ. Health., 44 (1): 58-64.

Khalaf-Allah, H. M. and Shehata, S. M. A. (2011). Report on state of atresia in the ovaries of some Mediterranean Sea fishes from the Egyptian coast., Al-Azhar Bull. Sci., 22(1): 1-12.

Khalil, M. T. and Hussein, A. H. (199٧). Recycling and reuse of wastewater for fish farming: An experimental field study at Sewage Treatment Plant, Egypt. J. Egypt.Ger. Soc. Zool., 19(B): 59-79.

Knight, J. A.; Anderson, S. and RAwie, J. M. (1972). Chemical basic of the sulfophosphovanillin reaction for estimating total serum lipid. Clin. Chem., 18: 199-202.

Kotickhoff, S. W. (1983). Pollutant sorption in environmental systems. EPA- 6001D, 80 -83, NTTS, Spring Field, VA.

Laxmi-Priya, S.; Senthilkumar, B.; Hariharan, G.; Paneer-Selvam, A.; Purvaja, R. and Ramesh, R. (2011). Bioaccumulation of heavy metals in mullet (*Mugil cephalus*) and Oyster (*Crassostrea madrasensis*) from Pulicat Lake, south east cost of India, Toxicol. Ind. Health., 27(2):117-126.

Mathana, P.; Raj, S. T.; Nair, C. R. K. and Selvamohan, T. (2012). Seasonal changes in the biochemical composition of four different tissues of red spotted emperor *Lethrinus lentjan* (Family:Lethrinidae) Scholars Research Library Annals of Biological Research, 3 (11):5078-5082.

Masoud, M. S., EL-Samra, M. and EL-Sadawy, M. M. (2007). Heavy-metal distribution and risk assessment of sediment and fish from El-Max Bay, Alexandria, Egypt. Chemistry and Ecology, 23(3): 201-216.

Masoud, M. S.; Wagdi M.; El-Saraf, A. M.; Abdel–Halim, A. E. and Hamad M.I. (2012). Heavy metals accumulation in sediments of Alexandria Coastal Areas. Bull. Facu. of Sci., 47(1,2):12-28.

Mikhail, S. K. (2008)**.** Dynamics of estuarine phytoplankton assemblages in El-Mex Bay, Alexandria (Egypt): Influence of salinity gradients, Egypt J. Aquat. Biol. & Fish., 12(4):231– 251.

Mostafa, A.; Barakat, A.; Qian, Y. and Wade, T. (2004). An overview of metal pollution in the Western Harbor of Alexandria, Egypt. Soil and Sediment Contamination, (13):299-311.

Oregioni, B. and Aston, S. (1984). The determination of selected trace metals in marine sediments by flame atomic absorption spectrophotometry. IAEA, Monaco laboratory, Internal Report.

Ptashynski, M. D. and Klaverkamp, J. F. (2002). Accumulation and distribution of dietary nickel ion in lake whitefish, Coregonus clupeaformis. Aqua. Toxicol., 58: 249-264.

Radwan, A. R. and Lotfy, I. M. (2002). On the pollution of Burullus Lake water and sediments by heavy metals. Egypt. J. Aquat. Boil.&Fish., 6(4): 147-164.

Raymont, J. E. G. and Shields, J. (1994). Advances in water pollution research. Pergamon press. London, Oxford. 290 pp.

Rubbi, S. F.; Muslemuddin, M.; Majeda, B.; Shamin, J. S. and Ahmed, A. T. (1985). Handling of six species of fresh fish of

Bangladesh. FAO Fisheries., 317: 108-122.

Said, M. A.; El-Deek, M. S.; Mahmoud, Th. H. and Shriadah, M. A. (1994). Effect of pollution on the hydrochemical characteristics of different water types in El-Max Bay area, west of Alexandria, Egypt. ActaAdriatica, 34 (1/2): 9-19.

Said, T. O. and El-Agroudy, N. A. (2003). Concentrations of some heavy metals in the Red Sea water and their effects on the coral reef community. Egypt. J. Aquat. Boil. & Fish., 7(2):1-17.

Saloman, W. N.; Rooij, H. and Bril, J. (1987). Sediments as a source for contaminants. Hydrobiol., J. 149: 13-30p.

Sancho, E.; Ferrando, M.D.; Fernandez, C. and Andreu, E. (1998). Liver energy metabolism of *Anguilla anguilla*, after exposure to fenitrothion. Ecotoxicol. Environ. Saf., 41(2): 168-175.

Shakweer, L. M. and Abbas, M. M. (1997). Heavy metals concentration levels in some fish species of Lake Mariut and the Nozha Hydrodome, Egypt. Bull. Nat. Inst. Oceanogr. & Fish. A.R.E., 23: 167-186.

Shakweer, L. M. and Abbas, M. M. (2005). Effect of ecological and biological factors on the uptake and concentration of trace elements by aquatic organisms at Edku Lake. Egyptian Journal of aquatic research, 1110-0354.

Singh, N. B. and Sinha, R. N. (1977). Carbohydrates, lipids and protein in the developmental stages of *Sitophilus oryzae* and *Sitophilus granaries*. Soc. Am., (70):107-111.

Vinodhini, R. and Narayanan, M. (2008). Bioaccumulation of heavy metals in organs of fresh water fish *Cyprinus carpio* (Common carp). Int. J. Environ. Sci. Technol., 5(2):179-182.

Virk, S. and Sharma, R. C. (1999). Biochemical changes induced by nickel and chromium in the liver of *cyprinus carpio* (L). *Pollution Research.*, 18(3): 217-222.

Vosyliene, M. Z. and Jankaite, A. (2006). Effect of heavy metal model mixture on rainbow trout biological parameters, Ekologija. 4: 12-17.

Vuković, Z.; Radenković, M.; Stanković, S. and Vuković, D. (2011). Distribution and accumulation of heavy metals in the water and sediments of the River Sava., J. Serb. Chem. Soc. 76 (5) 795–803.

Yacoub, A. M. and Gad, N. S (2012). Accumulation of some heavy metals and biochemical alterations in muscles of Oreochromis niloticus from the River Nile in Upper Egypt. INTERNATIO. J. OF ENVIRON.SCI. and Engineer. (IJESE)., 3: 1- 10.

Yapici, G.; Timur, I. H. G. and Kaypmaz, A. (2006). Lead and cadmium exposure in children living around a coal-mining area in Yatagan, Turkey", Toxicol. Ind. Health., 22: 357-62.

Younis, A. E .M. (2005). Speciation of Organotin Compounds in Semi-closed Areas along the Mediterranean Coast of Alexandria. M. Sc. in Chemistry, Faculty of science; Al-Azhar University, Egypt.

Zaghloul, H. K. (2000). Effect of different water sources on some biological and biochemical aspect of the Nile tilapia, *Oreochromis niloticus* and Nile cat fish, *Clarias gariepinus*. Egyptian J. Zool., 34:353-377.