



Effects of Gender and Season on Potentially Toxic Metal Levels in Muscles of Adult Blue Swimmer Crabs (*Portunus pelagicus*) from the Northeastern Mediterranean Sea

Deniz AYAS^{1*}

Abstract

The effects of gender and season on potentially toxic metal levels in two muscle types (lump crabmeat, LCM, and chela crabmeat, CCM) of adult blue swimmer crabs, *Portunus pelagicus*, caught in Mersin Bay, Northeastern Mediterranean Sea, were investigated. In each season, the muscle tissues from 40 individuals of each sex were taken out by hand. Inductively coupled plasma atomic emission spectrophotometer was used to determine metal levels in samples. The annual ranges of metal levels in the LCM of blue swimmer crabs were: 0.50-1.01 mg Cd kg⁻¹, 0.37-0.45 mg Cr kg⁻¹, 0.10-0.40 mg Pb kg⁻¹, 9.64-33.85 mg Cu kg⁻¹, 37.80-78.16 mg Zn kg⁻¹, and 9.35-25.51 mg Fe kg⁻¹. The annual ranges of metal levels in the CCM of blue swimmer crabs were: 0.63-1.47 mg Cd kg⁻¹, 0.31-0.53 mg Cr kg⁻¹, 0.12-0.52 mg Pb kg⁻¹, 25.13-72.50 mg Cu kg⁻¹, 89.85-178.61 mg Zn kg⁻¹, and 7.31-20.08 mg Fe kg⁻¹. Cu and Zn levels in CCM of blue swimmer crabs were higher than in LCM ($p < 0.05$). Cu and Zn specifically accumulated in CCM. In this study, it was found out that there was no relationship between gender and metal accumulation. In addition, metals such as Cu, Zn, and Fe showed seasonal variations.

Keywords

Blue swimmer crab; *Portunus pelagicus*; Season; Gender; Metal contamination

Introduction

Blue swimmer crab, *Portunus pelagicus*, is an exotic species of Indo-Pacific origin. This species arrived in /migrated to the Mediterranean Sea with the opening of the Suez Canal. Today, they are particularly distributed along the Northeastern Mediterranean coast.

Blue swimmer crab is one of the economically important big sea crabs, and they are caught for consumption around the world and in Turkey, where 55 and 95 metric tons of blue swimmer crabs were caught in 2003 and 2007, respectively [1].

Mersin Bay is densely populated with over one million people

*Corresponding author: Deniz AYAS, Department of Seafood Processing Technology, Faculty of Fisheries, University of Mersin, 33160, Mersin, Turkey, Tel: 90-324-3412813; Fax: 90-324-3413025; E-mail: ayasdeniz@gmail.com

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living around the bay area and considerable agricultural and industrial activities are carried out there. Mersin Port is one of the largest ports in Turkey. Many petrochemical facilities are located in the bay as well. Fertilizers and pesticides are heavily used in the agricultural fields in the bay area. Domestic, industrial, and agricultural pollutants are carried into the bay through the rivers in the region.

Mersin Bay is an area where the fishing is intensive and it is important to determine the pollution in all species caught in Mersin Bay, particularly, given that invertebrate species have weak detoxification mechanisms. A small number of studies have been carried out on the heavy metal accumulation of invertebrate species in Mersin Bay [2-4]. Metal accumulation in the crabs may vary with season, gender, size, sexual maturation, and muscle type. Thus, the objective of this study was to compare and determine the seasonal and gender effects on the metal levels of different muscle tissues of mature blue swimmer crabs harvested from Mersin Bay.

Materials and Methods

Materials

Blue swimmer crabs, *Portunus pelagicus*, were caught by dip net from Mersin Bay on the coast of the Northeastern Mediterranean Sea. Sampling was carried out in April (spring), July (summer), October (autumn), and December (winter) in 2011. In the fishing procedure, a dip net with mesh size of 32 mm was used. In each season, 40 male and 40 female crabs were caught and kept in polystyrene boxes with ice. When they were brought to the laboratory, they were no longer alive. Molting of crabs was observed in October-November, and the crabs caught in these months were semisoft shell crabs.

Sample preparation

Some morphometric measurements of the carapace length (CL), carapace width (CW), and weight of all samples were carried out done (Table 1). Using calipers and weighing balance. After that, the muscle tissues from 40 individuals of each sex were taken out. All assays were conducted on triplicate samples of the homogenates.

Metal analysis

The metal analyses were carried out according to Canli and Atli [5] with some modifications. The LCM and CCM samples (0.5 g dry weight each) were dried at 105°C to constant weight and then analar grade concentrated nitric acid and perchloric acid were added to the samples. They were put on a hot plate set to 150°C until all tissues were dissolved. Inductively coupled plasma atomic emission spectrophotometer (ICP-AES, Varian Model, Liberty series II, Sydney, Australia) was used to determine metal concentrations. The metal concentrations (Cr, Cd, Pb, Cu, Fe, and Zn) in the samples are reported as mg metal kg⁻¹ dry weight. The standard addition method was used to correct for matrix effects. A high purity multi-element standard purchased from Charleston, USA was used for the determination of the metals [2-4,6].

Statistical analysis

Statistical analysis of data was carried out with SPSS 16.0. Duncan's test was used to evaluate the seasonal and gender effects on metal levels in LCM and CCM of blue swimmer crabs.

Table 1: Morphological measurements of blue swimmer crabs in Mersin Bay.

| Season | NS | CL $\bar{X} \pm S_{\bar{X}}$ | Min-max (mm) | CW $\bar{X} \pm S_{\bar{X}}$ | Min-max (mm) | Weight $\bar{X} \pm S_{\bar{X}}$ | Min-max (g) | Sex |
|--------|----|---------------------------------|-----------------|---------------------------------|-----------------|-------------------------------------|----------------|-----|
| Spring | 40 | 71.72 ± 5.60 ^a | 65.0-83.0 | 148.93 ± 12.36 ^a | 130.5-175.0 | 255.80 ± 57.80 ^b | 160.33-390.83 | ♀ |
| | 40 | 65.88 ± 6.10 ^a | 54.0-76.5 | 131.68 ± 10.35 ^a | 112.0-145.0 | 138.48 ± 22.04 ^a | 107.37-177.59 | ♂ |
| Summer | 40 | 70.37 ± 5.35 ^a | 62.0-80.5 | 140.70 ± 9.40 ^a | 124.0-155.5 | 197.66 ± 49.46 ^a | 130.28-308.75 | ♀ |
| | 40 | 69.23 ± 4.16 ^a | 62.5-76.0 | 137.00 ± 6.25 ^a | 125.0-147.0 | 206.38 ± 30.42 ^a | 171.20-290.10 | ♂ |
| Autumn | 40 | 69.70 ± 7.06 ^a | 61.5-82.5 | 133.75 ± 12.78 ^a | 122.0-160.0 | 170.48 ± 47.61 ^a | 132.06-268.64 | ♀ |
| | 40 | 70.92 ± 3.06 ^a | 66.0-75.0 | 138.33 ± 6.01 ^a | 131.0-147.0 | 215.63 ± 18.37 ^a | 201.90-253.40 | ♂ |
| Winter | 40 | 73.75 ± 4.04 ^a | 68.5-81.5 | 143.65 ± 6.52 ^a | 137.0-156.0 | 203.78 ± 30.73 ^a | 166.56-262.73 | ♀ |
| | 40 | 69.10 ± 3.76 ^a | 64.0-74.5 | 137.07 ± 6.02 ^a | 130.5-151.0 | 224.05 ± 32.45 ^a | 177.11-277.47 | ♂ |

NS = Number of specimens; CL= carapace length; CW= carapace width
Different letters (a, b) in the same columns for each season indicate significant differences (p<0.05).

Results and Discussion

The carapace width (CW) of the blue swimmer crabs had a range of 112.0-175.0 mm (Table 1). In a study carried out on the coast of Egypt by Razek, male and female blue swimmer crabs (*P. pelagicus*) with CW between 90-100 mm were considered to be adults [7]. In a similar study carried out in the Kakinada region of India, Devi accepted male and female blue swimmer crabs (*P. pelagicus*) with CW over 95 mm as adults [8]. In the study conducted by Shields and Wood at Moreton Bay in Australia, it was found out that a minimum CW of 96-109 mm was needed for sexual maturation [9]. In a similar study, Chang and Hsu reported that the minimum CW for females should be 82.36 mm and 92.86 mm for males for the sexual maturation of *P. sanguinolentus* living near the coast of Taiwan [10]. Thus, according to the results of these studies, the male and female blue swimmer crabs used in our study were adults.

The annual differences in the Cd, Cr, Pb, Cu, Zn, and Fe levels of muscle tissue in blue swimmer crabs were determined as 0.50-1.47 mg Cd kg⁻¹, 0.31-0.53 mg Cr kg⁻¹, 0.10-0.52 mg Pb kg⁻¹, 9.64-72.50 mg Cu kg⁻¹, 37.80-178.61 mg Zn kg⁻¹, and 7.31-25.51 mg Fe kg⁻¹, respectively. The quantitative relationships of the metal levels of in the blue swimmer crabs generally followed this sequence in all the seasons: Zn > Cu > Fe > Cd > Cr ≥ Pb (Table 2). Ayas and Ozogul had reported Cd, Cr, Pb, Cu, Zn, and Fe levels in muscle tissues of Atlantic blue crab (*C. sapidus*) as 0.4-1.1 mg Cd kg⁻¹, 0.2-0.6 mg Cr kg⁻¹, 0.2-0.6 mg Pb kg⁻¹, 9.7-68.1 mg Cu kg⁻¹, 39.5-175.2 mg Zn kg⁻¹, and 8.8-32.5 mg Fe kg⁻¹, respectively [3]. All our metal levels were similar to those found in that study. This may be due to the fact that the two different species were caught in the same region in both studies. In a similar study, Al-Mohanna and Subrahmanyam reported that Cr, Pb, Cu, and Zn levels in blue swimmer crab (*P. pelagicus*) were 0.2-0.7 mg Cr kg⁻¹, 1.7-2.1 mg Pb kg⁻¹, 110.2-142.8 mg Cu kg⁻¹, and 188.7-221.7 mg Zn kg⁻¹, respectively, in Kuwait Bay [11]. Cr, Pb, Cu, and Zn levels presented by them were higher than the data obtained in our study, possibly because of more fuel transfers into Kuwait Bay than in Mersin Bay. In a similar study, Gökoğlu and Yerlikaya reported Zn, Fe, and Cu levels in muscle tissue of blue swimmer crab (*P. pelagicus*) as 37.2-46.8 mg Zn kg⁻¹, 4.5-6.8 mg Fe kg⁻¹, and 14.9-20.8 mg Cu kg⁻¹, respectively, in Antalya Bay, Northeastern Mediterranean Sea [12]. It can be seen that the results presented by them were lower than those in our study, which may be due to the fact that the crabs had been caught in different areas with different contamination levels.

The findings indicated that the annual ranges of metal levels in the muscle tissues of female blue swimmer crabs were: 0.71-1.23 mg Cd kg⁻¹, 0.32-0.53 mg Cr kg⁻¹, 0.10-0.52 mg Pb kg⁻¹, 11.74-70.62 mg Cu kg⁻¹, 37.80-178.61 mg Zn kg⁻¹, and 7.76-25.32 mg Fe kg⁻¹. The annual ranges of metal levels in the muscle tissues of male blue swimmer crabs were: 0.50-1.47 mg Cd kg⁻¹, 0.31-0.44 mg Cr kg⁻¹, 0.29-0.46 mg Pb kg⁻¹, 9.64-72.50 mg Cu kg⁻¹, 37.98-150.13 mg Zn kg⁻¹, and 7.31-25.51 mg Fe kg⁻¹ (Table 2). Metal levels in muscle tissues of female and male blue swimmer crabs were similar irrespective of the season and muscle types. In the present study, there was no relationship between gender and metal accumulation. Ayas and Ozogul had earlier reported in a study carried out in Mersin Bay that metal levels in muscle tissue of female and male Atlantic blue crabs (*C. sapidus*) were similar irrespective of season and muscle types [3]. Olusegun et al. had stated that metal accumulation differences between the sexes of *C. amnicola* species of portunid crabs were not important [13]. In a similar study, Beltrame et al. also indicated that there was no relationship between gender and bio-accumulated metals in burrowing crab (*Neohelice granulata*) caught in a coastal lagoon in Argentina [14]. Furthermore, Chen et al. reported that there was no relationship between gender and metal accumulation in muscle tissues of the rock crab (*Thalassidroma crenata*) caught in Dapeng Bay, Taiwan [15]. All of these studies present findings similar to ours.

The annual ranges of metal levels in the LCM of blue swimmer crab were: 0.50-1.01 mg Cd kg⁻¹, 0.37-0.45 mg Cr kg⁻¹, 0.10-0.40 mg Pb kg⁻¹, 9.64-33.85 mg Cu kg⁻¹, 37.80-78.16 mg Zn kg⁻¹, and 9.35-25.51 mg Fe kg⁻¹. The annual ranges of metal levels in the CCM of blue swimmer crabs were: 0.63-1.47 mg Cd kg⁻¹, 0.31-0.53 mg Cr kg⁻¹, 0.12-0.52 mg Pb kg⁻¹, 25.13-72.50 mg Cu kg⁻¹, 89.85-178.61 mg Zn kg⁻¹, and 7.31-20.08 mg Fe kg⁻¹ (Table 2). It can be seen that Cu, and Zn levels in CCM were higher than those in LCM (p < 0.05). Cu and Zn metals specifically accumulated in CCM. Ayas and Ozogul also found out that Cu (22.17-68.09 mg/kg) and Zn (93.92-175.21 mg/kg) in CCM of Atlantic blue crabs (*C. sapidus*) were higher than Cu (9.72-43.70 mg/kg) and Zn (39.52-97.26 mg/kg) in LCM caught in Mersin Bay [3]. In a similar study, Gökoğlu and Yerlikaya also reported that Cu (2.08 mg/kg) and Zn (4.68 mg/kg) in CCM of blue swimmer crab (*P. pelagicus*) were higher than Cu (1.49 mg/kg) and Zn (3.72 mg/kg) in LCM caught in Antalya Bay, Northeastern Mediterranean [12]. Both of these studies showed findings similar to those obtained in our study. The accumulation of Cu and Zn in CCM of portunid crabs may be a general characteristic.

Table 2: The effects of season and sex on metal levels of different muscle tissues of the blue swimmer crab (mg kg⁻¹).

| Metal | Sex | Spring $\bar{X} \pm S_{\bar{X}}$ | Summer $\bar{X} \pm S_{\bar{X}}$ | Autumn $\bar{X} \pm S_{\bar{X}}$ | Winter $\bar{X} \pm S_{\bar{X}}$ | Muscle |
|-------|-----|-------------------------------------|-------------------------------------|-------------------------------------|-------------------------------------|--------|
| Cd | ♀ | 1.01 ± 0.02 ^{a,z} | 0.80 ± 0.02 ^{b,y} | 0.71 ± 0.04 ^{b,x} | 0.87 ± 0.05 ^{a,y} | LCM |
| | | 1.23 ± 0.02 ^{b,y} | 1.17 ± 0.02 ^{c,y} | 1.06 ± 0.01 ^{c,x} | 1.02 ± 0.02 ^{b,x} | CCM |
| | ♂ | 1.01 ± 0.01 ^{a,z} | 0.58 ± 0.01 ^{a,y} | 0.50 ± 0.02 ^{a,x} | 0.97 ± 0.02 ^{b,z} | LCM |
| | | 1.47 ± 0.04 ^{c,z} | 0.63 ± 0.02 ^{a,x} | 0.69 ± 0.04 ^{b,x} | 1.17 ± 0.01 ^{c,y} | CCM |
| Cr | ♀ | 0.40 ± 0.03 ^{a,y} | 0.37 ± 0.02 ^{b,x} | 0.43 ± 0.03 ^{a,y} | 0.45 ± 0.02 ^{b,y} | LCM |
| | | 0.32 ± 0.06 ^{a,x} | 0.53 ± 0.05 ^{d,y} | 0.34 ± 0.02 ^{a,x} | 0.53 ± 0.02 ^{c,y} | CCM |
| | ♂ | 0.38 ± 0.05 ^{a,x} | 0.44 ± 0.01 ^{c,x} | 0.38 ± 0.05 ^{a,x} | 0.44 ± 0.01 ^{b,x} | LCM |
| | | 0.38 ± 0.02 ^{a,x} | 0.31 ± 0.04 ^{a,x} | 0.38 ± 0.02 ^{a,x} | 0.35 ± 0.04 ^{a,x} | CCM |
| Pb | ♀ | 0.29 ± 0.04 ^{a,y} | 0.30 ± 0.02 ^{a,y} | 0.29 ± 0.01 ^{a,y} | 0.10 ± 0.02 ^{a,x} | LCM |
| | | 0.29 ± 0.01 ^{a,y} | 0.52 ± 0.01 ^{c,z} | 0.27 ± 0.01 ^{a,y} | 0.12 ± 0.01 ^{a,x} | CCM |
| | ♂ | 0.30 ± 0.02 ^{a,x} | 0.40 ± 0.03 ^{ab,y} | 0.31 ± 0.04 ^{b,x} | 0.33 ± 0.04 ^{b,x} | LCM |
| | | 0.46 ± 0.01 ^{b,y} | 0.42 ± 0.05 ^{b,y} | 0.34 ± 0.04 ^{b,x} | 0.29 ± 0.04 ^{b,x} | CCM |
| Cu | ♀ | 33.85 ± 0.88 ^{b,z} | 30.91 ± 0.39 ^{b,y} | 12.49 ± 0.43 ^{b,x} | 11.74 ± 0.36 ^{a,x} | LCM |
| | | 50.81 ± 0.98 ^{c,z} | 70.62 ± 1.40 ^{c,q} | 28.32 ± 0.61 ^{d,y} | 25.13 ± 0.35 ^{c,x} | CCM |
| | ♂ | 30.32 ± 0.82 ^{a,q} | 15.56 ± 0.31 ^{a,z} | 9.64 ± 0.26 ^{a,x} | 13.27 ± 0.67 ^{b,y} | LCM |
| | | 50.34 ± 0.95 ^{c,y} | 72.50 ± 1.00 ^{c,z} | 26.74 ± 0.56 ^{c,x} | 28.47 ± 0.45 ^{d,x} | CCM |
| Zn | ♀ | 77.81 ± 0.90 ^{b,y} | 78.16 ± 0.84 ^{b,y} | 39.77 ± 0.78 ^{b,x} | 37.80 ± 0.46 ^{a,x} | LCM |
| | | 178.61 ± 0.92 ^{d,q} | 153.20 ± 1.74 ^{d,z} | 93.99 ± 0.48 ^{d,x} | 97.55 ± 0.39 ^{c,y} | CCM |
| | ♂ | 66.77 ± 0.70 ^{a,y} | 39.46 ± 1.22 ^{a,x} | 37.98 ± 0.27 ^{a,x} | 38.43 ± 0.48 ^{a,x} | LCM |
| | | 150.13 ± 0.99 ^{c,q} | 139.20 ± 0.80 ^{c,z} | 89.85 ± 0.69 ^{c,x} | 92.51 ± 0.38 ^{b,y} | CCM |
| Fe | ♀ | 25.32 ± 0.40 ^{c,q} | 15.87 ± 0.19 ^{b,z} | 9.35 ± 0.14 ^{b,x} | 10.61 ± 0.43 ^{b,y} | LCM |
| | | 17.52 ± 0.44 ^{a,z} | 14.38 ± 0.56 ^{b,y} | 7.76 ± 0.25 ^{a,x} | 8.69 ± 0.46 ^{a,x} | CCM |
| | ♂ | 25.51 ± 0.78 ^{c,z} | 11.67 ± 0.47 ^{a,y} | 10.26 ± 0.17 ^{c,x} | 11.53 ± 0.46 ^{b,y} | LCM |
| | | 20.08 ± 0.60 ^{b,q} | 12.24 ± 0.67 ^{a,z} | 7.31 ± 0.19 ^{a,x} | 8.64 ± 0.13 ^{a,y} | CCM |

Different letters (a, b, c, d) in the same columns for each metal indicate significant differences (p<0.05). Different letters (x, y, z, q) in the same rows for different muscle tissues indicate significant differences (p<0.05). $\bar{X} \pm S_{\bar{X}}$ = mean ± standard error

According to our findings, the levels of Cd, Pb, Cu, Zn, and Fe in *P. pelagicus* showed seasonal variations. Seasonally, Cd and Pb showed very small variations. On the other hand, metals such as Cu, Zn, and Fe exhibited large seasonal variations (Table 2). It was found that Cu, Zn, and Fe levels in muscle tissues of blue swimmer crabs in spring and summer seasons were higher than those in autumn and winter. Ayas and Ozogul also reported that Cd, Cr, Pb, Cu, Zn, and Fe levels in muscle tissues of *C. sapidus* showed seasonal changes [3]. It was particularly stated that Cu, Zn, and Fe levels were higher in spring and summer than in autumn and winter, indicating large seasonal changes. Beltrame et al. also reported that concentrations of some metals (Cd, Cu) in *N. granulata* displayed some seasonal changes. These studies presented findings similar to those obtained in our study [14]. Overall, the studies show conclusively that the levels of Cu, Zn, and Fe in *P. pelagicus* showed seasonal variations (Table 2).

Molting of blue swimmer crabs in Mersin Bay was observed in October-November, and the crabs caught in these months were semi-soft shell crabs. Cu, Zn, and Fe levels in muscle tissues of blue swimmer crabs were found to be higher in spring and summer than in autumn and winter (Table 2). Reductions in feeding in autumn and winter compared with the other seasons and carapace molting in autumn may have played a role in these results. In the present study, molting was associated with differences in metal levels depending

on the type of metal and muscle tissue. Molting caused a reduction in the levels of Cu, Zn, and Fe of blue swimmer crabs by 40-60% in autumn and winter, with the reduction in the CCM being lower than those in LCM. Ayas and Ozogul also reported metal elimination and a reduction in Cu, Zn, and Fe levels by nearly 70% during the molting of carapace in Atlantic blue crabs in Mersin Bay [3]. In a similar study, Bergey and Weis also found metal elimination and a reduction in Cu and Zn levels by 3-12% and 8-22%, respectively, during the molting of carapace in crabs [16]. In a related study carried out by Benjakul and Sutthipan, a reduction of 100% in Cu, Fe, and Zn in soft shell crabs were reported when compared with hard shell crabs [17]. It was reported that the reduction in LCM were higher than those in CCM. The findings of our study are therefore supported by those of Benjakul and Sutthipan [17].

Cd, Cr, and Pb levels in muscle tissue of blue swimmer crabs were determined annually as 0.50-1.47 mg Cd kg⁻¹, 0.31-0.53 mg Cr kg⁻¹, and 0.10-0.52 mg Pb kg⁻¹, respectively (Table 2). The Cd limit levels reported for crabs were 0.5 mg Cd kg⁻¹ [18,19]. It was noted that blue swimmer crabs living in Mersin Bay were contaminated with Cd. The Cr limit level reported by U.S. FDA was 12 mg Cr kg⁻¹ for crabs [20]. Pb limit level for crabs has been set at 0.5 mg Pb kg⁻¹ [18,19]. Thus, it can be stated that blue swimmer crabs living in Mersin Bay were not contaminated with Cr and Pb.

Cu and Zn levels in muscle tissues of blue swimmer crabs were in the range 9.64-72.50 mg Cu kg⁻¹ and 37.80-178.61 mg Zn kg⁻¹, respectively (Table 2). The Cu and Zn limit levels reported for crabs were 20 mg Cu kg⁻¹ and 50 mg Zn kg⁻¹, respectively [19]. In the present study Cu levels in LCM of blue swimmer crabs were only lower than those limit level in autumn and winter. Cu levels in all muscle tissues of blue swimmer crabs were higher than the reported limit level [19], except for Cu levels in LCM of blue swimmer crabs in autumn and winter. Zn levels showed similarities with Cu levels and Zn levels in all muscle tissues of blue swimmer crabs were higher than the reported limit level [19] except for Zn in LCM of blue swimmer crabs in autumn and winter. Similar results were obtained for Cu and Zn levels in the study conducted on Atlantic blue swimmer crab in Mersin Bay by Ayas and Ozogul [3].

Conclusion

The results of metal levels in blue swimmer crab indicated that there was no relationship between gender and metal accumulation. The results also showed that the season specifically affected levels of Cu, Zn, and Fe in muscle tissues. It was found that the Cd, Cu, and Zn levels in blue swimmer crabs caught in Mersin Bay, Turkey, were higher than those that are internationally accepted. All blue swimmer crabs were contaminated with these metals. Molting was found to be a good detoxification mechanism for this crab species. It is recommended that these big sea crabs caught in Mersin Bay should be consumed only when caught in autumn and winter and in the other seasons their harvesting and consumption should be avoided or prohibited.

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Author Affiliation

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¹Department of Seafood Processing Technology, Faculty of Fisheries, Mersin University, Mersin, Turkey

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