

Comparison of Otolith Mass Asymmetry in Two Different *Solea solea* Populations in Mediterranean Sea

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Abstract

In general, water pollution can be called as a kind of pollution resulting from natural and human activities on water resources such as oceans, seas, rivers, lakes and ground waters. Fish species are directly and indirectly adversely affected by water pollution. Like other living things in the aquatic system, fish are confronted with different types of stresses from different pollutants such as pesticides, insecticides, herbicides and heavy metals in their habitats. Otolith mass asymmetry may reflect some developmental disorders of fish caused by environmental stress. Especially high-level otolith mass asymmetry can adversely affect fish life, so the determination of the asymmetry is very important for each species. The aim of the present study to compare the otolith mass asymmetry of *Solea solea* from Mersin and İskenderun populations. A total of 100 fish with 17.5-25.0cm total length and 36.4-126.6g weight were collected from the Mediterranean Sea. The asymmetry was calculated considering the difference between blind and eye sides otolith masses. In the present study, the asymmetry values were found within -0.25483 and +0.41220 in the populations. The otolith mass asymmetry and absolute otolith mass asymmetry of *S. solea* in the Mersin and İskenderun populations are not related to the total length.

Keywords: Otolith; Mass asymmetry, *Solea solea*; Ecological factors; Water pollution

Akdeniz'de İki Farklı *Solea solea* Popülasyonunda Otolit Kütle Asimetrisinin Karşılaştırılması

Öz

Su kirliliği genel olarak okyanuslar, denizler, nehirler, göller ve yeraltı suları gibi su kaynakları üzerinde doğal ve insan faaliyetleri sonucu oluşmuş bir tür kirlilik olarak adlandırılabilir. Su kirliliği balık türlerini doğrudan ve dolaylı olarak olumsuz etkilenmektedir. Sucul sistemdeki diğer canlılar gibi balıklar da habitatlarında bulunan pestisitler, insektisitler ve herbisitler ve ağır metaller gibi farklı kirleticilerden kaynaklı farklı stres türleriyle karşı karşıyadır. Otolit kütle asimetrisinin, balıklarda çevresel stresin neden olduğu bazı gelişimsel bozuklukları yansıtabilir. Özellikle yüksek seviyedeki otolit kütlesi asimetrisi balıkların yaşamını olumsuz yönde etkileyebilir, bu nedenle bu asimetrinin belirlenmesi her balık türü için çok önemlidir. Bu çalışmanın amacı Mersin ve İskenderun popülasyonlarındaki *Solea solea*'nın otolit kütle asimetrisini karşılaştırmaktır. Total boyları 17,5-25,0 cm ve ağırlıkları 36,4-126,6 gr olan toplam 100 adet balık Akdeniz'den yakalanmıştır. Asimetri, kör ve göz bölge otolitlerinin kütleleri arasındaki fark dikkate alınarak hesaplanmıştır. Bu çalışmanın sonucunda, asimetri değerleri, bu iki popülasyonda -0,25483 ve +0,41220 değerleri arasında bulunmuştur. Mersin ve İskenderun popülasyonlarındaki *Solea solea*'nın otolit kütle asimetrisi ve mutlak otolit kütle asimetrisi, balıkların total boyuyla ilişkili değildir.

Anahtar Kelimeler: Otolith; Kütle asimetrisi, *Solea solea*; Ekolojik faktörler; Su kirliliği

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1. Introduction

Ecological factors such as abiotic and biotic factors affect the life of animals in their habitats. Generally, fish species are faced with different types of stress from different pollutants such as pesticides, herbicides, and heavy metals found in their habitats. Otoliths are bony structures were found in the inner ear of the marine and freshwater teleost. Asteriscus, lapillus, and sagitta are three types of otoliths and their growth is continuous throughout the life of the fish (Campana & Thorrold 2001; Campana 2004). Organic and inorganic materials of otoliths are not re-metabolized by fish species; therefore, accumulation in the otoliths can represent an uninterrupted record of the fish life (Arai et al 2007). Otoliths are kind of conserved sensory organs in the fish species. The otoliths play an important role in balance, hearing, gravity sensation and linear acceleration in the species; therefore, they are crucial for the survival of the fish species (Nolf 1985; Poper & Lue 2000). The otoliths do not always have to grow equally in all dimensions, even though they have a three-dimensional structure (Campana 1999; Campana & Thorrold 2001). These otoliths are normally bilaterally expected to be symmetrical in the fish but a weight difference between the left and right otolith masses is observed in some cases, and this is called otolith mass asymmetry.

The asymmetry in fish species is assumed to reflect the developmental disorders of fish caused by different type of stress such as genetic or environmental stress (Valentine et al 1973). Increase or decrease of otolith mass asymmetry can negatively affect other activities necessary for the life of the fish, especially the sense of hearing and balance. The otolith mass asymmetry has been used as a bioindicator to test the condition between different aquatic habitats (GrønkJaer & Sand 2003) and it was also used to test different environmental effects in fish populations. Soleidae is a flatfish family and it contains 32 genera and 174 species distributed in the freshwater, brackish and marine aquatic habitats (Froese & Pauly 2017). *Solea* is one of the genera in Soleidae family and it is commonly found in the Mediterranean Sea, East Atlantic, and Indo-Pacific Oceans. Genus *Solea* is represented by 9 species worldwide such as *S. capensis*, *S. aegyptiaca*, *S. elongate*, *S. ovata*, *S. heinii*, *S. turbynei*, *S. solea*, *S. senegalensis* and *S. stanalandi* (Froese & Pauly 2017). There are two species of these flatfish such as *Solea senegalensis* and *Solea solea* are widely found in Turkish waters.

Solea solea are called several names such as common sole, dover sole, and black sole. They have an oval and compressiform body, and their eyes on the right side. The fish are bilaterally symmetrical when they leave the egg, but after they have metamorphosed. The metamorphosis shifts their left eye to the right side thus they lose these symmetries and gain an asymmetrical structure. They usually live on muddy or sandy floors at depths of 20-40 meters and go down to 100 meters in the winter (Froese & Pauly 2017). *S. solea* feeds on mollusks, soft-shelled bivalves, small fish, and crustaceans. The average size of the species is 30-35 cm, maximum 50 cm and average weight is around 300-350 grams (Froese & Pauly 2017). Their meats are delicious and marketed as fried, frozen, broiled, fresh, microwaved, steamed and baked. Therefore, it is also commercially important species in Turkey (Bingel 1987; Özyurt et al 2008) Although the increase in otolith mass asymmetry studies throughout the world, information of otolith mass asymmetry in *S. solea* remains largely unknown in Turkey. In the current study, the first objective was to compare the otolith mass asymmetry and absolute otolith mass asymmetry of *S. solea* in the Mersin and İskenderun populations. The second objective was to

compare total length-otolith mass asymmetry and total length- absolute otolith mass asymmetry relationships *Solea* in the Mersin and İskenderun populations.

2. Materials and methods

Solea solea samples were collected from commercial anglers in Mersin and İskenderun Bays, Turkey. The total length of the samples was measured to the nearest 0.1 cm and their weight was recorded to the 0.1g for each species. Sagittal pairs were removed, cleaned and undamaged otolith pair (blind and eye sides) was weighted to the nearest 0.0001g for each species. The otolith mass asymmetry (x) was calculated using the formula: $x=(M_R-M_L)/M$, where M_R and M_L are the otolith masses of the right and left paired otoliths and M is the mean mass of the right and left paired otoliths. Theoretically otolith mass asymmetry (x) can change from -2 to +2. While -2 or +2 values indicate maximal asymmetry, '0' value refers to the absence of the mass asymmetry ($M_R=M_L$).

In the present study, the relationship between otolith mass asymmetry (x) and total length was calculated using $x=a.TL+b$ formula and the same formula also used for determination of the relationship between absolute otolith mass asymmetry ($|x|$) and total length. In this formula, TL is the total length of the fish, “ a ” is the coefficient characterizing the growth rate of the otolith, and “ b ” is a constant for the given species. The relationships were estimated using the linear regression methods in Excel software (Ver. 2016). In addition, the left and right otolith were compared and the differences between variables of pairs were investigated using the paired t-test for all individuals. A MINITAB (Ver. 16.0) software statistical analysis program was used for calculations and statistical analysis.

3. Results

Solea solea were collected from Mersin and İskenderun Bays in the Mediterranean Sea (Fig. 1). A total of 200 sagittal otoliths of 100 samples were removed in Mersin Bay and İskenderun Bay. Their total length ranged from 17.5 to 25.0 cm and weight range of the samples was to 36.4-126.6 g.



Figure 1. Two sampling habitats for *Solea solea* in the Mediterranean Sea; İskenderun Bay, Mersin Bay.

It is determined that the blind side otoliths of *S. solea* in the İskenderun and Mersin populations are heavier than the eye side otoliths. The blind and eye side otolith weight mean values are 0.013793 ± 0.000217 and 0.013381 ± 0.000219 for İskenderun population, respectively and 0.015822 ± 0.000422 and 0.015420 ± 0.000376 for Mersin population, respectively (Table 1). There is a statistical difference between blind and eye side otolith weights of *S. solea* for Mersin Bay (Paired-t; *P*-value= 0.019) and İskenderun Bay (Paired-t; *P*-value= 0.017) (Table 1).

Table 1. Descriptive statistics of *Solea solea* blind and eye side otolith weights in İskederun and Mersin Bays.

Population	Side	n	Mean	SE Mean	Minimum	Maximum	T-values	P-values
İskederun	blind	50	0.013793	0.000217	0.010600	0.017000	2.47	0.017*
	eye	50	0.013381	0.000219	0.010900	0.017200		
Mersin	blind	50	0.015822	0.000422	0.009600	0.024300	2.42	0.019*
	eye	50	0.015420	0.000376	0.009100	0.020400		

While the otolith asymmetry was calculated within the range of $-0.25483 \leq x \leq +0.31077$ for İskederun population, it was also calculated within the range of $-0.06620 \leq x \leq +0.41220$ for Mersin population (Table 2). Absolute otolith mass asymmetry was calculated as $0.00000 \leq |x| \leq +0.31077$ for İskederun population $0.00000 \leq |x| \leq +0.41220$ for Mersin population (Table 2). The mean values of *x* are 0.01391 ± 0.00667 and 0.03030 ± 0.01220 for İskederun and Mersin populations (Table 2). Furthermore, the mean values of *|x|* are 0.04301 ± 0.00505 and 0.04558 ± 0.00795 for İskederun and Mersin populations (Table 2). According to *Solea solea* otolith mass asymmetry and absolute otolith mass asymmetry results, there were no differences between İskederun and Mersin populations (*P*>0.05).

Table 2. Descriptive statistics of *Solea solea* otolith mass asymmetry (*x*) and absolute otolith mass asymmetry (*|x|*) in İskederun and Mersin Bays.

Population	Variable	n	Mean	SE Mean	Minimum	Maximum	T-values	P-values
İskederun	<i>x</i>	50	0.01391	0.00667	-0.25483	0.31077	1.06	0.291
Mersin		50	0.03030	0.01220	-0.06620	0.41220		
İskederun	<i> x </i>	50	0.04301	0.00505	0.00000	0.31077	0.15	0.884
Mersin		50	0.04558	0.00795	0.00000	0.41220		

There were no relationships between total length and otolith mass asymmetry for Mersin and İskederun populations, and their correlation coefficients and regression equations were $y=0.0204x-0.3923$; $R^2=0.0725$ and $y=0.0017x-0.0237$; $R^2=0.0013$, respectively (Figure 2A-D). According to regression analysis of total length-absolute otolith mass asymmetry results, the correlation coefficients and regression equations were calculated as $y=0.0209x-0.3828$; $R^2=0.0977$ and $y=0.0052x-0.074$; $R^2=0.0204$ for Mersin and İskederun populations respectively (Figure 2C-D).

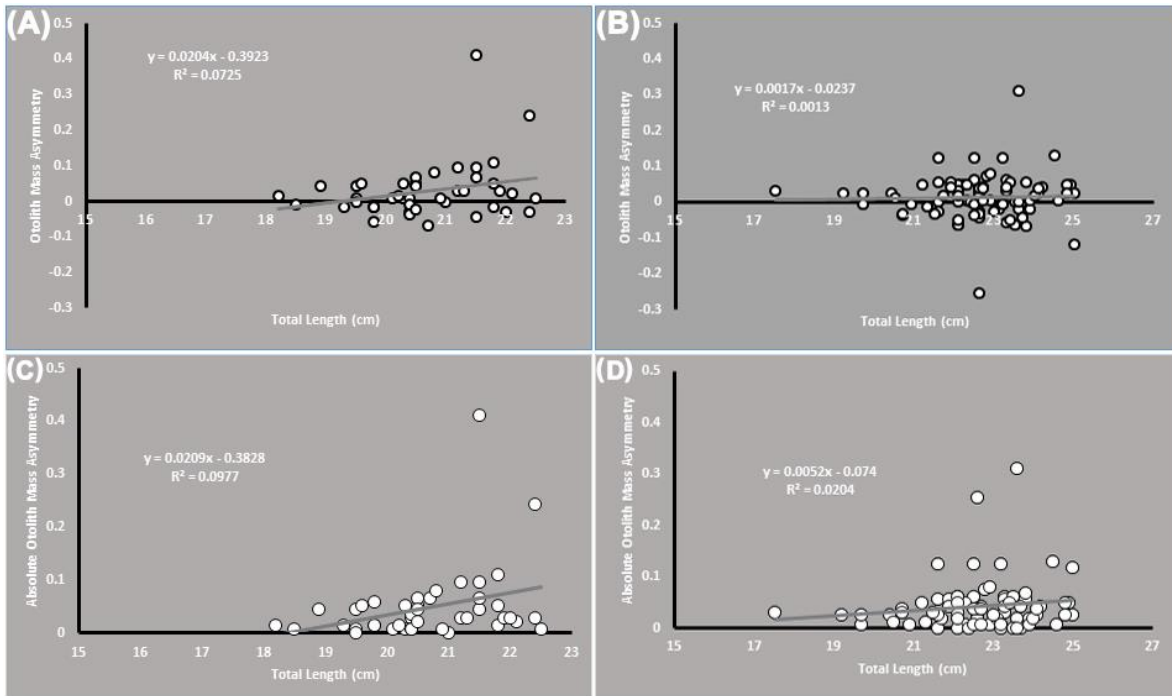


Figure 2. *Solea solea* otolith mass asymmetry (x) and absolute otolith mass asymmetry (|x|) as a function of total length in (A-C) Mersin Bay and (B-D) İskenderun Bay.

4. Discussion and Conclusion

Pollution in Turkey as well as all over the world comes at the beginning of the most important environmental problems. Some of the most common types of pollution are air pollution, water pollution, thermal pollution, soil pollution, radioactive pollution, noise pollution, light and visual pollutions. Some or all of these pollutions are affecting the life of the creatures, either separately or together. When considering the importance of water in living organisms, water pollution is a quite different position in other pollutions. Whether it is aquatic or terrestrial, there is only one purpose of investigating contamination; it is to examine the direct or indirect effects of pollution on living things or living resources and to take necessary measures according to the results obtained. These influences are also known to determine the physiology, histology and anatomy, behavioral patterns and nutritional habits of living things.

İskenderun and Mersin Bays where fish areas are caught in the present study are quite concerned with their pollution in the Mediterranean. Gündoğdu & Çevik (2017) reported that Mediterranean coast of Turkey, especially these areas, are very significantly polluted because of the oil storage stations, iron-steel industry, high maritime traffic, fertilizer, power plants and tourism activities.

The aquatic environment is constantly polluted by exposure to domestic, industrial and agricultural wastes and the negative impact of pollution on the ecosystem is increasing. This causes the degradation of the quality of water resources and the constant change of the aquatic

ecosystem (Turgut & Özgül 2009). The pollution in the living environment causes stress in the aquatic animals. This stress can cause developmental instabilities in fish. The fact remains that, based on previous studies in this area, there is a direct correlation between environmental stress and asymmetry resulting from pollutions (Jawad et al 2012).

Otolith mass asymmetry may have been a consequence of environmental stress caused by pollution, pesticides, insecticides, herbicides, predator pressure, lack of food and heavy metals, ion exchange in the water, accumulation problems in otoliths, crystal structures of otoliths (e.g. Aragonitic, Vateritic), and disease, genetic predisposition and even combination of some or all of these (Bostanci et al 2017).

In fisheries, there are several studies on otolith mass asymmetry were conducted and the otolith mass asymmetry values were ranged $-0.2 < x < +0.2$ for several marine and freshwater species (Lychakov et al 1988; Lychakov 1992; Takabayashi & Ohmura-Iwasaki 2003; Lychakov & Rebane 2004; Lychakov & Rebane 2005). As a result of the present study, this value was ranged $-0.25483 \leq x \leq +0.41220$ in these values for both İskederun and Mersin populations of *Solea solea*. The data obtained in current study indicate that the otolith mass asymmetry and absolute otolith mass asymmetry values of the *S. solea* are above the critical values in both the İskenderun and the Mersin populations.

However, Lychakov et al (2008) examined the otolith asymmetry of different types of flatfish and they determined the mean absolute otolith mass asymmetry value for *Solea solea* as 0.075 ± 0.012 in Catalan coast. This value was calculated as 0.04301 ± 0.00505 and 0.04558 ± 0.00795 for in both the İskenderun and the Mersin populations, respectively. This difference in the same species from İskenderun, Mersin and Catalan coasts can have several reasons. The main reasons are sampling size, size distributions of *S. solea* in this samples and gender differences. Another reason may be the differences in biotic and abiotic factors in the sampling areas. However, no statistical difference was found both the populations and species in terms of otolith mass asymmetry and absolute mass asymmetry in the current study ($P > 0.05$). High-level otolith mass asymmetry can adversely affect fish life, so the determination of the asymmetry is very important for each species. In the current study, the average otolith mass asymmetry of both populations of *Solea solea* was low, whereas the mass asymmetry of individuals with very high otolith mass asymmetry was determined when they were examined on an individual basis. These results show us that both populations are under environmental stress. It has been found that the otolith mass asymmetry of the *S. solea* individuals in the Mersin population is higher than those in the İskenderun population. In this case, it may be described that Mersin population is more exposed to pollution or other factors originating from the stress, and this is reflected by the otolith mass asymmetry of the fish.

On which side of the fish's eyes the blind or eye sides can be a cause for the weight of the otoliths to be greater. Otolith mass asymmetry value of *Pleuronectes platessa* which is right-eyed flatfish is around the zero and there is no significant difference between otolith pair (Helling et al 2005). However, in our study, average blind side otolith mass of *Solea solea* which is right side flatfish, is heavier than eye side otolith. Furthermore, *Psetta maxima* which is left-eyed flatfish, eye side otoliths are lighter than blind side otoliths (Helling et al 2005).

In several studies, the relationships between otolith mass asymmetry and total length and between absolute mass asymmetry and total length have been examined in several roundfishes and flatfishes (Lychakov et al 2006; Mille et al 2015). In the present study, we have investigated the relationships which are total length-otolith mass asymmetry and total length-absolute otolith mass asymmetry in *S. solea* individuals in both İskenderun and Mersin populations. According to present study results, the otolith mass asymmetry and absolute otolith mass asymmetry did not depend on total length for both İskenderun and Mersin populations. These results are correlated with other researchers results in several freshwater and marine species (Lychakov et al 2006, Jawad et al 2011; Jawad & Sadighzadeh 2013; Jawad 2013; Lychakov 2013; Jawad et al 2017).

In Turkey, otolith mass asymmetry studies on both flatfish and round fish are limited and there is not even a comparison study between populations. This study is expected to provide as a roadmap for future otolith mass asymmetry studies in marine and freshwater fish species and will allow researchers which will work on otolith mass asymmetry of *Solea solea* from both Turkey and abroad, to make comparisons between *S. solea* populations.

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