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# MARINE SCIENCE AND TECHNOLOGY BULLETIN

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#### MARINE SCIENCE AND TECHNOLOGY BULLETIN

VOLUME 9 • ISSUE 1 • JUNE 2020

# TABLE OF CONTENTS

RESEARCH ARTICLES	
<b>Public Aquariums in Turkey</b> Pınar ÇELİK, Ebru YALÇIN ÜLGER	1-6
<b>Determination of Fish Consumption in Çanakkale</b> Fahri SAKA, Musa BULUT	7-14
Characterization, Identification and Phylogeny of the Creatine Kinase (ckma) Gene in Medaka (Oryzias latipes) Mehtap BAYIR, Gökhan ARSLAN, Pınar OĞUZHAN YILDIZ	15-2
First Molecular Record of the Pacific Oyster (Crassostrea gigas, Thunberg 1793) in the Marmara Sea, Turkey Emel ÖZCAN GÖKÇEK, Bilge KARAHAN, Pervin VURAL, Evren KOBAN BAŞTANLAR, Sefa ACARLI	23-3
Length-Weight Relationship of the Most Landed Pelagic Fish Species European Pilchard ( <i>Sardina pilchardus</i> Walbaum, 1792) and European Anchovy ( <i>Engraulis</i> <i>encrasicolus</i> Linnaeus, 1758) in the İzmir Bay (Aegean Sea, Turkey) Purse Seine Fishery Ahmet Mert ŞENBAHAR, Özlem GÜLEÇ, Zafer TOSUNOĞLU, Okan ÖZAYDIN	32-3
The Presence of Bristlemouth, <i>Gonostoma denudatum</i> (Rafinesque 1810), From the Coast of Northern Cyprus (Northeastern Mediterranean) <sup>Hasan</sup> Deniz AKBORA, Deniz AYAS, Nuray ÇIFTÇI	42-4
Diet Composition of Bluefish <i>Pomatomus saltatrix</i> (Linnaeus, 1766) in the Sea of Marmara Habib BAL, Telat YANIK, Dilek TÜRKER	46-5
Investigation of Active Tectonics of Edremit Gulf, Western Anatolia (Turkey), Using High-Resolution Multi-Channel Marine Seismic Data Can EYTEMIZ, Faik Erdeniz ÖZEL	51-5
A Study on Maximum Length Record of Saddled Seabream ( <i>Oblada melanura</i> Linnaeus, 1758) Caught Off Gökçeada Island (Northern Aegean Sea, Turkey <sub>Özgür CENGIZ</sub>	58-6
Preliminary Results on the Growth and Survival of Larval European Lobster (Homarus gammarus (Linneaus, 1758)) in Turkey Emre ÖZER, Sefa ACARLI, Selçuk BERBER	62-7



#### MARINE SCIENCE AND TECHNOLOGY BULLETIN

VOLUME 9 • ISSUE 1 • JUNE 2020

# T A B L E O F C O N T E N T S

# SHORT COMMUNICATIONS

Additional Record of <i>Hemiramphus far</i> (Hemiramphidae) in Northern Aegean Sea (İzmir Bay, Turkey) Okan AKYOL, Zafer TOSUNOĞLU	38-41
An Observation About Maximum Size Record of Blotched Picarel ( <i>Spicara maena</i> Linnaeus, 1758) from Northern Aegean Coasts of Turkey <sup>Özgür</sup> CENGİZ	71-74



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# Marine Science and Technology Bulletin

# **RESEARCH ARTICLE**

# **Public Aquariums in Turkey**

# Pınar Çelik<sup>1\*</sup> 🕩 • Ebru Yalçın Ülger<sup>2</sup> 🕩

<sup>1</sup> Çanakkale Onsekiz Mart University, Faculty of Marine Sciences and Technology, Department of Aquaculture, Çanakkale, Turkey <sup>2</sup> Bursa Uludağ University, Faculty of Veterinary Medicine, Department of Physiology, Bursa, Turkey

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# ABSTRACT

This study has presented information in order to reveal the general condition of the activities of public aquariums in Turkey. Firstly, the location and number of public aquariums in Turkey are determined. Afterward, survey questions were prepared, which could show these business profiles. After the surveys have been prepared, some businesses were visited and the authorities were talked face to face. The business officials whom we could not visit were contacted by telephone and e-mail. In the survey, care has been taken not to ask for information that may transgress the public disclosure policy. The right to answer every question is left to the authorities' preference. For these reasons, the information provided about the enterprises can only be sufficient to reveal the general profile of the enterprises. According to this survey, a total of 13 large public aquariums in Turkey have been identified in 2019 and there is a large tunnel aquarium under construction. While 5 of the public aquariums of Turkey are in İstanbul, and 3 of them are located in Ankara, there is one public aquarium in Antalya, Bursa, Diyarbakır, Eskişehir and İzmir provinces. The majority of these aquariums (8) are located in or near shopping centers, the others (5) were located in the parking areas. In aquariums, whereas sea creatures are allocated more space, the areas reserved for freshwater creatures are less. Most of the aquariums have educational activities as well as entertaining activities. Aquaculture engineers, aquaculture technicians, aquanauts, veterinarians, biologists, graduates of fisheries technology and underwater technologies are the occupational groups that are employed in public aquariums.

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#### Introduction

\* Corresponding author

The history of public aquariums dates back to the 19th century (Karydis, 2011). These aquariums, which consist of large volume or a large number of small volume aquariums made of various materials, are called Public Aquarium or City Aquarium. The first large public aquarium in the world, known as the Fish House, was built in 1853 at the London Zoo with the name The London Zoo (Brunner, 2003). The second-largest aquarium was opened in Europe, Berlin (Karydis, 2011). Following the aquarium in Berlin, a large public aquarium was opened in Paris. Since the second half of the 1800s, people's interest in aquariums has gradually increased. Aquarium magazines began to be published since 1876 as The New York Aquarium Journal. In 1893, the first aquarist community was established in New York, The United



States of America (Karydis, 2011). Today, large public aquariums continue to operate in many countries, notably Europe, the United States, Canada, and the Far East. In the last 30 years, technological developments in fish farming have contributed to the development of technical equipment used in aquariums (Barnabe, 1989; Huguenin and Colt, 1992). The fact that the transfer of living beings between countries has become easier and has accelerated the development of aquariums all over the world. Nowadays, most of the large public aquariums operating around the world have been built in the last 30 years (Karydis, 2011). In Turkey, the first large public aquarium began to operate in İstanbul in 2009. The number of public aquariums increased one by one in the following years reached to 13 in 2019. In this study, detailed information about the large public aquariums operating in Turkey is presented. Identification of the current status of such public aquarium establishments and publications of the data obtained are important to be the source of new studies in this field.

#### **Material and Methods**

In the study, 13 public aquariums operating in Turkey were examined. The data presented in the study were collected in 2018. Firstly, current enterprises have been identified. Then, information was obtained from the enterprises with the questionnaires prepared in advance. The questionnaires were prepared in order to reveal the general description of the companies. For this purpose, questions were asked to describe the general status of the enterprise such as total area occupied, total aquarium volumes, aquarium sizes/numbers, the species they contain and the number of animals. The data obtained are based on oral and written information received from business authorities.

#### Results

According to the results of survey, the numbers of public/city aquariums operating in Turkey are 13 as of November 2019 (Table 1). One tunnel aquarium is also under construction. The first public aquarium open to the public was opened in 2009 in Bayrampaşa, İstanbul. This aquarium, which was built in a shopping center, was opened with the name Turkuazoo. This facility, which has a total water capacity of 5000 m<sup>3</sup>, currently operates under the name of SEA LIFE İstanbul Aquarium. The first large public aquarium in İstanbul attracted great attention. Then, the second-largest public aquarium in İstanbul entitled İstanbul Akvaryum was opened two years later. The water capacity of İstanbul Akvaryum is around 6800 m<sup>3</sup>. During these two years, two large public aquariums were also opened in Ankara with the initiative of the local municipalities. Although, these aquariums, which started to operate in Ankara, are much smaller than the two large aquariums in İstanbul, and continue to attract a large number of visitors. After the first large aquarium was founded in 2009, the total number of large aquarium enterprises in Turkey has risen to 13. So, all of the major public aquariums in Turkey were established in the last 10 years. All of these enterprises are located in metropolitan cities such as İstanbul (5 units), Ankara (3 units), Antalya (1 unit), Bursa (1 unit), Diyarbakır (1 unit), Eskişehir (1 unit) and İzmir (1 unit) (Table 1). The vast majority (60%) of large public aquariums in Turkey are located in İstanbul and Ankara which are the most crowded and the capital city

of Turkey, respectively. Most of enterprises were established by supporting of local municipal investments. The greater part of them is operating in or near shopping centers. Among these aquariums, Antalya and İstanbul come to the forefront with respect to the criteria such as water capacity, the number of themes contained the tank presentations, animal variety, and the total area occupied.

#### SEA LIFE İstanbul Aquarium (Turkuazoo)

SEA LIFE İstanbul Aquarium is the first and the largest public aquarium in Turkey and it was established in 2009. This business was established under the name of Turkuazoo and then it was entitled as SEA LIFE İstanbul Aquarium because of the management change. This aquarium was built into the shopping mall called Forum İstanbul in Bayrampaşa, İstanbul. Its original investment is around 17 thousand Euros. By the representative of Indonesian company Global Aquarium in Turkey, İstanbul Underwater World Tourism Trade Inc. was established as Turkuazoo, and later passed into UK company Merlin Entertainments. When it was opened, its visitor numbers reached to 2500 at the end of the first week. Initially, it was reported that the number of visitors reached close to 1 million per year. It is an enterprise that could use the advantage of being the first public aquarium established in Turkey in terms of visitor numbers. According to the information provided by the establishment, approximately 320000 people visited this aquarium in 2018.

More than 50 people are employed including aquaculture engineers, aquaculture technicians, aquanauts, biologists, visitor guides, and management staff in SEA LIFE İstanbul Aquarium.

The total water capacity in the aquarium is around 5000 m<sup>3</sup>. Totally 30 tons of sand was used in aquariums. There are 45 tanks in the facility, 8 of them are freshwater aquariums, and others are marine aquariums. The facility hosts 15478 nektons of 500 species. There is one large tunnel aquarium. In addition to one large main tank, other tanks are of various sizes, large and small.

The main species exhibited in the establishment are composed of species such as Bowmouth shark, sand tiger shark, Bonnethead shark, blackfin sharks, zebra shark, guitar shark, giant grouper, clownfish, reef flying gurnard, blue-faced African threadfish, seahorse, jellyfish, starfish, giant moray eel, cow nosed stingray, spotted freshwater stingray, black stingray, spotted common eagle ray, leopard stingray, bug-eyed soldierfish, nurse shark, octopus, brown crab, blue crab, stickleback bubble fish, lobster, queen triggerfish, batfish, lionfish, croc hunter chelonian, mata mata turtle, Danube sturgeon, red-bellied piranhas, discus, boxfish, long-horned cowfish, dragon eels, knife fish, horseshoe crab, sanitary shrimp, hermit crab, rainbow crab, tropical corals and stonefish. There is also SEA LIFE İstanbul Sea Turtle Rehabilitation Center in the facility.

#### Keçiören Outdoor Aquarium

This aquarium was established in 2010 by Keçiören Municipality of Ankara as an open-air aquarium in Fatih Sultan Mehmet Park in Etlik, Ankara. The aquarium is a marine aquarium with a length of 35 meters and a capacity of approximately 300 tons of water. This aquarium is introduced as the first and the only open-air aquarium established in Turkey.





Name of the Aquarium	Year of Establishment	Current Location	Water Capacity (m <sup>3</sup> )	Number of Tanks (pcs)	Number / Species of Animals
SEA LIFE İstanbul Aquarium (Turkuazoo)	2009	Bayrampaşa, İstanbul	5000	45	It is home to 15478 nektons of 500 species
Keçiören Outdoor Aquarium	2010	Etlik, Ankara (Fatih Sultan Mehmet Park)	300		It was opened with 2230 invertebrates together with 671 sea fish in 13 species
Deniz Dünyası	2010	Keçioren, Ankara	1000	18	It accommodates 4000 marine and freshwater species of 150 species
İstanbul Akvaryum	2011	Florya, İstanbul	6800	64	There are 17000 land and sea creatures
Kaplıkaya Cazibe Merkezi	2011	Yıldırım, Bursa	3000	17	There are approximately 5000 fish in tunnel aquarium and 150 fish in other aquariums
Aqua Vega Aquarium	2012	Ankara	4500	24	There are 12000 marine species
Antalya Aquarium	2012	Konyaaltı, Antalya	7500	64	It hosts roughly 10 thousand species
ETI Underwater World	2014	Sazova Park, Eskişehir	1400		There are a total of 2150 living creatures in 84 species
Viasea Aquarium	2015	Tuzla, İstanbul	5200		It has over 12000 marine creatures in 47 different themed exhibitions
Aqua Diyarbakır	2015	Diyarbakır	1700	31	It hosts 2500 different marine creatures of 150 species
Jungle İstanbul	2015	Eyüp - İstanbul			
Emaar Aquarium & Underwater Zoo	2017	Üsküdar, İstanbul		48	It hosts over 20000 nektons and amphibians of 200 species
Funtastic Aquarium İzmir	2018	İzmir	2000	70+	
Aquarium Ortahisar	Under construction	Ortahisar, Trabzon		1 Tunnel Aquarium	It is expected to be the longest (180 m) underground tunnel aquarium in the world

Table 1. List of	public aquariums	operating in 7	Furkey ()	As of November 2019).
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When the aquarium was firstly opened, a total of 671 marine fish belong 13 species and 2230 invertebrates were exhibited. This aquarium serves in a way that people can visit for free.

#### Deniz Dünyası

It was established by Keçiören Municipality of Ankara was opened with the name of *Deniz Dünyası* (which is *Marine World* in Turkish) in 2010. It has a closed area of 2700 m<sup>2</sup> and a total area of 4000 m<sup>2</sup>. 140 thousand people were reported to have visited during the first two months of its opening. This facility consists of 12 tunnel aquariums, 7 special aquariums, a cylinder aquarium, a touch aquarium and a diving helmet aquarium. In addition, it has a 1000 m<sup>3</sup> water capacity in total. In the facility, which hosts 4000 sea and freshwater fish and 150 turtles species, there are also 2 African crocodiles with a length of 1.5 m. The aquarium prioritizes training programs, particularly, elementary school students are provided with educational information about marine species and marine life.

#### İstanbul Akvaryum

İstanbul Akvaryum (*in Turkish*; aquarium) is the fourth public aquariums after aquariums of Turkuazoo, SEA LIFE İstanbul Aquarium, and Deniz Dünyası according to the establishment date. However, it is the second public aquarium in terms of establishment concept and theme content. In addition, it is the largest aquarium in Turkey according to the water capacity at the establishment date, 2011. It was opened with an investment budget of approximately 168 million TRY with the initiatives of the İstanbul Metropolitan Municipality. This aquarium, which was initially operated by the İstanbul Metropolitan Municipality, was transferred to a private company in 2013. The number of visitors who come to the aquarium in a year is reported to be approximately 1.2 million people. The enterprise employs approximately 200 people and approximately 500 people with its subcontractors and service units.

İstanbul Akvaryum has a thematic aquarium concept with 64 tanks of various sizes and has a total of 6800 m<sup>3</sup> water capacity. According to the information given by the company, 17000 land and marine animals are exhibited. Its biggest living nekton is the lemon shark. Red-bellied piranhas, Russian sturgeon, anemones, clownfish, groupers, Gentoo penguins, stingrays, and anaconda are counted among the marine animals that it hosts.

It is a large thematic public aquarium. It consists of 17 themes and 1 rain forest following a geographical structure extending from the Black Sea to the Pacific. It is the first aquarium where all the seas are together.

#### Kaplıkaya Cazibe Merkezi

It was opened in 2011 as *Kaplıkaya Cazibe Merkezi* (*in Turkish*, Kaplıkaya Attraction Center) in Yıldırım, Bursa with the initiatives of Yıldırım Municipality of Bursa. This aquarium is designed in a slightly different way from the well-known public aquariums, has succeeded



in attracting public attention. There are totally 5 employees; 2 divers, 1 machine technician and 2 aquaculture engineers in the aquarium. It has a tunnel aquarium and the tunnel aquarium is 25 meters long with a water capacity of 3000 tons. In addition, there are 2 aquariums with a volume of 50 tonnes, 4 aquariums with a volume of 6 tons, 10 aquariums with a volume of 1.5-2 tons. Two of these aquariums are marine aquarium while others are freshwater aquarium. Moreover, there are carp and koi fishes in the tunnel aquarium since it has an open water circulation system. There are 2 eels and clownfish in the marine aquarium, and various species of cichlid, catfish variety, gourami and crocodile fish in other aquariums. There are approximately 5000 fish in tunnel aquarium and 150 fish in other aquariums. These species are procured from within the country.

#### Aqua Vega Aquarium

Aqua Vega Aquarium was opened in 2012 within a shopping center in Ankara. It was established by the private sector with an investment of 17 million Euros. In the first year of its establishment, it hosted 500000 visitors. The main theme of this facility is a large tunnel aquarium with a length of 98 m. 24 different aquarium components were used in the facility which has a water capacity of 4500 m<sup>3</sup>. It is stated that there are approximately 12000 marine animals in the aquarium, it is possible to come across many nektons such as sharks, Koi fish, Napoleon fish, and clownfish. In the Wildlife Section, serval, Flemish giant rabbit, domestic ferret, marmoset, golden pheasant, African crocodile, mephitis, cotton-headed tamarin, a red-cheeked water turtle, and helmeted Guineafowl are also demonstrated.

#### Antalya Aquarium

It is the biggest tunnel aquarium of the world with a length of 131 meters and a width of 3 meters. Antalya Aquarium was opened in 2012 with the initiatives of the Antalya Metropolitan Municipality. It has been established in the Konyaaltı region of Antalya where the number of domestic and foreign tourists is quite high. It was established with an investment fund of 80 million TRY. This facility is located in the tourism region and therefore has a large visitor portfolio. Most of the foreign visitors coming to Antalya from other countries visit this aquarium. In this respect, Antalya Aquarium also plays an important mission for the presentation of Turkey. Since this company was established with the build-operate-transfer logic of the Antalya Metropolitan Municipality, it was transferred to a private company after a while. The number of visitors reached 1 million at the end of the first year, and reached 5 million within 7 years. Antalya Aquarium has nearly 50 employees, 24 of which are life support teams.

There are 64 aquariums of different sizes in the facility and 40 different themes are displayed. Total water volume of the aquariums is approximately 7500 m<sup>3</sup>. The water capacity of its main tanks is approximately 5000 m<sup>3</sup>. Although there are about 10 thousand living animals in the aquarium, most of these species are marine animals.

#### ETİ Underwater World

ETİ Underwater World, opened in 2012 in Sazova Park, Eskişehir, is an enterprise established in cooperation with Eskişehir Metropolitan Municipality and ETİ Company. This aquarium was launched with an investment of approximately 6.5 million TRY. The aquarium is capable of hosting 400 visitors at a session, and 3500 visitors in a day. The entrance fees have been kept at very reasonable levels. In this way, it was aimed to be able to host as many visitors as possible. It accommodated 2500 people on the first day of its opening and 100000 people in 24 days. ETI Underwater World was established on an area of 2350 m<sup>2</sup>. It consists of more than 30 thematic aquariums, a 19-meter aquarium tunnel, and a tropical aquarium with poisonous and tropical amphibian species, Amazon River and sturgeon aquarium, terrarium, touch aquarium. A total of 2150 living animals belong to 84 species are demonstrated in the aquarium, which is a public aquarium rich in diversity.

#### Viasea Aquarium

This aquarium was established in 2015 by a private company within a shopping center in Tuzla, İstanbul. It has an investment value of approximately 1 billion TRY and is located in a theme park. In the first two days of its opening, it hosted 10000 visitors. The water capacity of this facility is approximately 5200 m<sup>3</sup>. 47 different themed aquariums display over 12000 marine animals. This aquarium is the first largest public aquarium on the Asian side of İstanbul. It has the largest capacity among public aquariums in Turkey and also a rehabilitation center (quarantine area).

Visitors are also given the opportunity to walk around life support sections to show how a public aquarium is managed.

#### Aqua Diyarbakır

Aqua Diyarbakır is the first largest public aquarium launched in the Eastern Anatolia Region of Turkey. This company was also established by a private company in a shopping center with an investment of approximately 30 million TRY. The aquarium hosted 696 thousand people in the first 5 days of its opening. There is a team of 40 experts working in the facility including veterinarians, biologists, aquanauts, and aquaculture engineers. It has a water capacity of 1700 m<sup>3</sup> with thematic aquariums, touch ponds, main tank (3 different concepts), and a 55-meter long tunnel aquarium with a total of 31 aquariums. It hosts 2500 different sea marine animals belong to 150 species, including sharks, piranha, lobster, and octopus.

#### Jungle İstanbul

This aquarium is also a thematic aquarium established by a private company in a shopping center. Jungle İstanbul was opened in 2015 in Eyüp district of İstanbul. The aquarium, which was established with an investment of approximately 650 million TRY, serves the visitors coming to the shopping center. In addition to aquariums, the property of the business is designed in a large theme park concept, where various tropical animals such as snakes, spiders, chameleons, crocodiles, frogs and exotic birds are on appearance.

#### Emaar Aquarium & Underwater Zoo

This establishment was opened in a shopping mall in Üsküdar district of İstanbul. One veterinarian and 8 aquaculture engineers are employed in the aquarium. There are 48 tanks in various sizes in the facility. It hosts over 20000 marine and amphibian animals belong to 200 species such as shark and stingray species, shrimps, shellfish, coral reefs, jellyfish, snakes, spiders, iguanas, chameleons, wild piranhas, giant water rats, otters, red-cheeked turtles, arawanas, herbivore piranhas, thornback rays, Humbolt penguins, Macaw parrots, naked mole-rat, Cayman lizard), mini manta, giant spider crab, king crocodile.

It consists of 7 different thematic sections including rocky shores, main tank with tunnel aquarium (glass-like underwater tunnel with a 270-degree view of coral reefs 3.5 meters below the surface), jellyfish gallery, forests, rivers and waterfalls, penguin island, crocodile zone.

#### Funtastic Aquarium

Funtastic Aquarium is a medium-sized public aquarium that established in 2018 within a shopping center in İzmir. Its water capacity is approximately 2500 m<sup>3</sup>. In this facility, species such as sand tiger shark, stingray, blowfish, red-bellied piranha, moray eel, archerfish, clownfish, octopus, red arowana are exhibited. This aquarium is the first aquarium in İzmir.

#### Aquarium Ortahisar

It is still under construction. The aquarium planned to be built in Ortahisar, Trabzon. It is expected to be the longest (180 meters) underground tunnel aquarium in the world. This aquarium is established by the initiatives of the Ortahisar Municipality of Trabzon.

#### Discussion

Mankind's interest in caring for and protecting wild and domestic animals goes back to ancient times. So much so that Mesopotamia, Egypt, China and possibly India between 3000 BC and 1456 AD were the first known communities to have animal collections (Kisling, 2000). People's interest in animals led them to have animal collections. The epicenter of such animal collecting activities later spread to the Greco-Roman regions, the Persians and the Arab regions (Kisling, 2000). People's interest in animals first started with collecting activities, later on, there was development towards establishing zoos. The first animal species exhibited in zoos are of course were land animals. It has become a very popular field of activity to present wild species to people's tastes. After land animals, aquatic animals were also exhibited in zoos. This is how public aquariums were formed. The first large public aquarium known in the world was opened in 1853 at the London Zoo (Brunner, 2003). Since then, public aquariums have become a well-known and widespread activity all over the world.

Zoos and aquariums can be defined as exhibition spaces that offer thousands of different species to people's tastes. However, nowadays, zoos and public aquariums have social responsibility areas other than just exhibiting animals. These structures were also part of wildlife conservation activities. On the other hand, it has multifaceted positive outcomes that emphasize development of veterinary medicine, technology, education, park and recreation development, human sensitivities to nature and cultural change (Kisling, 2000). From this point of view, public aquariums have the power of raising awareness on various fields ranging from education to nature conservation besides creating a good time and entertaining people.

Large public aquariums, which have begun to spread between the second half of the 18th century and the first half of the 19th century

(Karydis, 2011), began to be established in Turkey after 2009 for the first time. As it is indicated in the present paper, the number of public aquariums operating in Turkey, having a population of around 82 million in 2019, is 13. All of these aquariums are located in provinces with the highest population density such as İstanbul, Ankara, İzmir, Bursa, Antalya, Eskişehir and Diyarbakır in Turkey. Predominantly, İstanbul is the province that has the most public aquariums. Almost half of the current aquariums in Turkey (5 aquariums) are operating in İstanbul. The largest two public aquariums in Turkey were established in İstanbul. People's interest in these two aquariums triggered the opening of public aquariums in İstanbul and other regions of Turkey.

The installation and operation costs of public aquariums are very high. For this reason, in order to sustain efficiently business, the income obtained from the visitors must be continuous. This is one of the primary reasons for the establishment of these aquariums in İstanbul, İzmir, Ankara and other metropolitan cities. There is a direct correlation between the sustainability of the business and the number of visitors to the aquarium. Since the investment costs are high, the local municipalities undertook these investments in many provinces. In addition to the economic gains of these structures built for the benefit of the public, their social contributions are very valuable.

Dozens of different tropical marine and freshwater species are on display in public aquariums in Turkey. However, it exhibits the most popular animals in demand such as sharks, stingray fish, tropical coral ecosystems, tropical marine fish, herd species, crabs, seahorses, piranhas, moray eel, which are the species more attracting the attention of people, as well as exotic animals such as crocodiles, penguins, water snakes. Species living in the seas of Turkey are exhibited as well as tropical species. A total of 67 fish species including 8 species belong to 4 ordo and 8 families from Chondrichthyes (cartilaginous fish) group, and 59 species belonging to 9 ordo and 24 families from Osteichthyes (bony fish) group have been identified in the waters of Turkey were reported in a study examining the species exhibited in public aquariums operating in İstanbul, Ankara, Bursa and Antalya (Gültekin et al. 2014). When the habitats of the detected species exhibited in aquariums are examined, it is observed that mostly benthic species (57 species) are preferred in the public aquariums (Gültekin et al., 2014). In addition, 6 semi-pelagic, 3 epipelagic and 1 pelagic species have been reported (Gültekin et al., 2014). In public aquariums in Turkey, 40 species living in the Black Sea, 48 species living in the Sea of Marmara, 62 species living in the Aegean Sea and in the Mediterranean Sea are known to be widespread (Gültekin et al., 2014). People also have the chance to see many aquatic organisms such as sharks, stingrays, eels, seahorses, crabs, lobsters, octopuses in public aquariums, which they cannot see in the regions where they live (Avcı, 2016).

#### Conclusion

Public aquariums significantly contribute to the educational activities of children, principally for children of small age groups. They can provide opportunities for people of all ages to have a good time and have fun with their families in these facilities. Public aquariums are also profitable commercial areas for operators. The number of visitors coming to some aquariums shows that public aquariums are also important commercial areas. Public aquariums are important for their economic and social contributions. In addition, they provide employment opportunities to many people who have been trained as aquaculture engineers, aquaculture technicians, biologists, veterinarians, and fisheries technology graduates. Given all these contributions, there is no reason why public aquariums should not continue their activities in the future. The total number of public aquariums in Turkey, which are currently 13 nowadays, can be expected to increase in the future.

#### **Conflict of Interest**

The authors declare that there is no conflict of interest.

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# Marine Science and Technology Bulletin

# **RESEARCH ARTICLE**

# Determination of fish consumption in Çanakkale

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Çanakkale

# ABSTRACT

Fish plays a key role in human consumption in terms of protein, mineral, and essential fatty acid contents. Unfortunately, despite its importance for the human health, there is lack interest on the fish consumption in Turkey. In this context, this paper aimed to determine the fish consumption habits in Canakkale. It is estimated that fish consumption could be higher in the locations along the coasts of marine and inland waters. Therefore, consumers living in all districts of the city were surveyed to test this hypothesis. The questionnaire was carried out to provide an insight into the fish consumption habits of randomly selected 1056 consumers in Çanakkale. Socio-economic and demographic structures such as age, gender, educational status, profession, income level of the consumers were determined. Responses of the consumers were arranged and analysed by using SPSS and MS-Excel software. Moreover, fish consumption amount, consumption frequency, preferred fish species, most consumed fish species were also determined. The results give an excellent snapshot of fish consumption habits in Çanakkale. Both the most consumed and the most favourite fish species are identified as Sarda sarda. Fish consumption frequency was described as 33.3% (352 individuals) for consuming fish one a week followed by bimonthly frequency (21.9%, 231 individuals) and monthly frequency (21.5%, 227 individuals). 2.3% (24 individuals) of the participants noted that they never consume fish. Moreover, the majority of consumers specified that they consume fish 1-2 kg (39.0%, 312 individuals) and 27.3% (288 individuals) consume fish 0.5-1 kg. Socio-economic and demographic characteristics of consumers are affecting the fish consumption habits. In the present study, season, income level, and freshness of fish are found to be driving force for fish consumption in Çanakkale. Therefore, fish farmers and sellers are recommended to remain the freshness of fish and to follow the appropriate fishing season for providing fish to consumers.

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# Introduction

Fish has a great importance for human health since its content including protein, mineral, vitamin and essential fatty acids. Therefore, fish consumption is vital for healthy life. Global fish consumption has reached 20.3 kg/year per capita in 2016 (FAO, 2018). On the other hand, fish consumption was 5.4 kg per capita in 2016 for Turkey which is the least consumption amount per capita since 2000 (GDFA, 2019). Recently, it increased to 6.14 kg/year per head in 2018 (TurkStat, 2019). Moreover, in 2018, aquaculture has provided more fish for human consumption than capture fisheries in Turkey.

The expansion in fish consumption has been driven not only by enlarged production, but also by a grouping of several other dynamics, containing better utilization, growing demand, reduced wastage, and developed distribution networks, connected with rising incomes, population growth, and urbanization (FAO, 2018). Moreover, increasing interest on dietetic aspects, waste reduction, food safety, and food quality has also supplemented the increase of the fish consumption.

FAO and WHO (2011) indicated that fish consumption has positive effects on mental health, age related macular degeneration, and inhibiting cardiovascular diseases. In case of low per capita consumption of fish, even slight amounts of fish are able to supply essential fats, amino acids, and micronutrients (e.g., calcium, iodine, iron, and vitamin D) which are not originate in plant-based diets (FAO, 2018). Authorities come to an agreement that the beneficial effects of high fish consumption mainly compensate the possible undesirable effects associated with contamination or further safety risks (FAO and WHO, 2011).

Average per capita fish consumption differs meaningfully within and across regions and countries due to the effects of geographic, economic, demographic and cultural factors. In the present study, it is aimed to determine the fish consumption in Çanakkale. This study investigated fish consumption behaviour of the consumers living in all districts of the city.

#### **Material and Methods**

The core material of the study is the original data recently collected through questionnaires from the participants living in Çanakkale. Questionnaire survey was conducted between February 2019 and December 2019. A total of 1056 people were surveyed in all districts of Çanakkale. The targeted consumers were requested permission to fetch data, and the data were obtained from the enthusiastic consumers within 5-10 min.

Total population of Çanakkale is reported as 540662 by TurkStat (2019). Required minimum sampling size was determined with equation (1) according to the random sampling method suggested by Collins (1986). The population, required minimum sampling size and applied sampling size for all districts are tabulated in Table 1.

$$n = \frac{N \times P \times Q \times Z_{\alpha}^2}{d^2} \tag{1}$$

In this equation, *n* is the sample size, *N* is the population of district, *P* is the probability of occurrence (assumed as 0.05), *Q* is the unoccurrence probability (Q=1-P),  $Z_a$  is the confidence coefficient (accepted as 2.58 for 0.01 error margin), *d* is the sampling error that is accepted according to the incidence of the event.

**Table 1.** The population, required minimum sampling size andapplied sampling size for all districts

District	Population	Required Minimum Sampling Size	Applied Sampling Size
Ayvacık	33568	50	30
Bayramiç	29716	45	30
Biga	90576	136	48
Bozcaada	3023	5	30
Çan	48215	72	36
Eceabat	8912	13	57
Ezine	32003	48	44
Gelibolu	44809	67	110
Gökçeada	9783	15	72
Lapseki	27327	41	122
Merkez	180823	272	400
Yenice	31907	48	77

The data acquired from the questionnaire were statistically assessed by using SPSS v23.0 statistical package program. Frequency tables, distribution charts, Chi-square test, and oneway analysis of variance (ANOVA) were used. The statistical significance of the relationships between the variables was accepted as p <0.05.

The statistical relationship between the frequency of fish consumption of consumers and their socio-economic, demographic and behavioural characteristics were evaluated using the Chi square test. Moreover, the effects of the socio-



economic, demographic and behavioural characteristics of the habits on the fish consumption frequency were also assessed.

#### Results

The socio-economic and demographic status of the consumers is presented in Table 2. The distribution of the participants according to the district of residence was presented in Figure 1. 57.6% of the respondents were male and 42.4% were female. 57.5% of the participants are married and 42.5% are single. When the ages of the participants were examined, 30.8% were in the 19-29 age range and 27.8% were in the 30-49 age range. When the educational status was evaluated, it was determined that 33.4% graduated from high school or equivalent schools and 19.1% graduated from undergraduate programs. When the professions of the participants were examined, 27% were students, 20% were self-employed, 18.6% were workers, 13.6% were homemakers, 13.3% were public officers, and 7% were retired. When the income levels are analysed, it is determined that 41.9% of the monthly income is 2020 TRY or less, which is the minimum wage for Turkey in 2019, and 24.5% is between 3001-4000 TRY. The majority of the participants have a minimum wage or less monthly income. The household size and the preference status for fish consumption of the participants are given in Table 2.

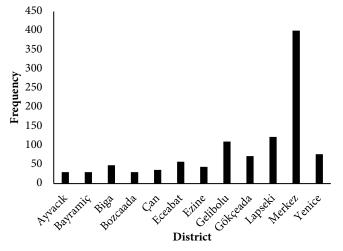
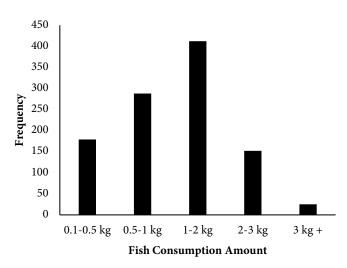


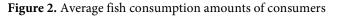
Figure 1. Distribution of the participants according to the district of residence

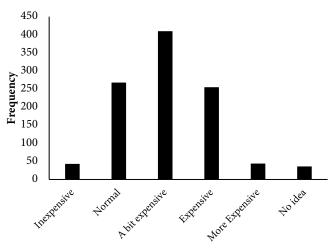
When the amount of fish consumption is analysed, 39% of the respondents stated that they consumed 1-2 kg, 27.3% consumed 0.5-1 kg, 17% consumed 0.1-0.5 kg, 14.4% consumed 2-3 kg, and 2.4% consumed 3 kg or more fish (Figure 2).

While 95.4% of the respondents stated that they bought fish instead of fishing (Table 2), 38.8% thought that fish prices were a bit expensive (Figure 3). 21.2% of consumers preferred peddlers for fish buying while 27.1% preferred fish stalls and 21.8% of consumers preferred fish markets. A total of 30.6% of

the participants preferred more than one place to buy fish (Figure 4). 59.1% of the respondents preferred to fish consumption according to the season when buying fish (Table 2).

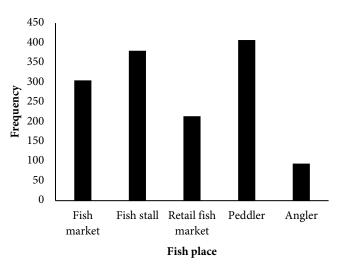


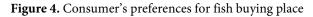




Consumer's opinion about fish price

Figure 3. Consumer's opinion about fish price







With regard to fish consumption according to season, 33.1% of consumers stated that they consumed mostly in winter season while 43.5% of the respondents stated that they consumed fish in more than one season (Figure 5). The most fish consumed season was described as the winter followed by spring, autumn, and summer, respectively.

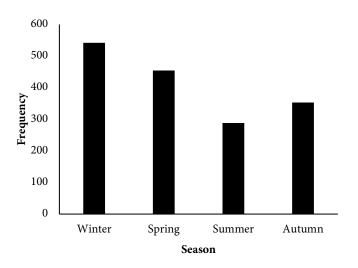


Figure 5. Seasonal preference of consumers for fish consumption

The majority of participants have preferred fresh fish for consuming (87.6%, 925 individuals). Moreover, consumers pay attention to the freshness of the fish during buying fish (26.0%, 746 individuals) while 15.9% of consumers take care to be appropriate to the season. The huge portion of the participants (50.0%, 528 individuals) shows ultimate attention for buying fish and checks more than one criterion (Figure 6).

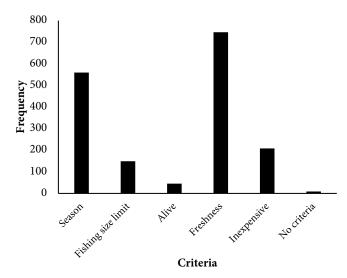


Figure 6. Main criteria that consumers take care of when buying fish

The most favourite fish species was determined to be Atlantic bonito (*Sarda sarda*) by 51% (538 individuals) of the consumers. After anchovy, the most favourite fish species was

determined to be anchovy (*Engraulis encrasicolus*) by 47% (496 individuals) and bluefish (*Pomatomus saltatrix*) by 37% (391 individuals) of the consumers (Figure 7).

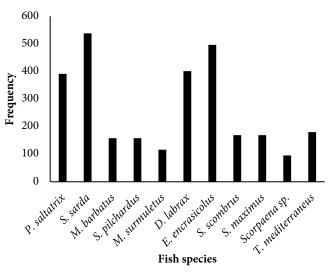


Figure 7. The most favourite fish species of consumers

It was determined that the most consumed fish species was Atlantic bonito (65%, 686 individuals). The most consumed fish species after anchovy were found as anchovy (53%, 560 individuals), sea bass (*Dicentrarchus labrax*) (43%, 454 individuals), and bluefish (27%, 285 individuals) (Figure 8).

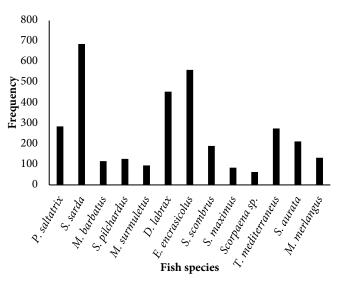


Figure 8. The most consumed fish species of consumers

When the frequency of fish consumption of the participants was examined, it was determined that 33.3% of the respondents consume fish once a week, 21.5% of the participants consume fish once a month and 21.9% of the individuals consume fish bimonthly. However, 2.3% of the participants stated that they never consume fish (Figure 9). When the fish consumption pattern of the participants is examined, it is understood that 87.6% of the participants prefer fresh fish consumption. While 31.6% of the respondents preferred to fry as cooking method,



Table 2. The socio-economic an	d demographic structures of
consumers	

26.2% preferred grilling method. However, 57.5% of the participants used more than one cooking method (Figure 10).

Characteristics	Frequency	Ratio (%)
Gender		
Male	608	57.6
Female	448	42.4
Age		
18	149	14.1
19-29	325	30.8
30-49	294	27.8
50-49	168	15.9
60+	120	11.4
Marital Status		
Single	449	42.5
Married	607	57.5
Education Level		
Not graduated	5	0.5
Primary school	115	10.9
Secondary school	174	16.5
High school	353	33.4
Associate degree	148	14.0
Bachelor's degree	202	19.1
Master's degree	46	4.4
Doctoral degree	13	1.2
Profession		
Public officer	140	13.3
Worker	196	18.6
Student	285	27.0
Retired	80	7.6
Homemaker	144	13.6
Self-employed	211	20.0
Income Level (TRY/month)		
< 2020 TRY	442	41.9
2021-3000 TRY	224	21.2
3001-4000 TRY	259	24.5
4001-5000 TRY	103	9.8
> 5001 TRY	28	2.7
Household Size		
1	99	9.4
2	189	17.9
3	387	36.6
4	320	30.3
5+	61	5.8
Preference		5.0
Optional	432	40.9
Seasonal	432 624	40.J 59.1
Fish Providing Method	024	57.1
Buying	1007	95.4
Fishing	38	3.6
1 10111118	50	5.0

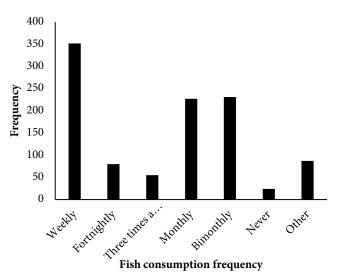


Figure 9. Fish consumption frequency of consumers

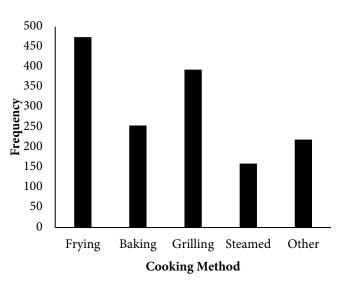


Figure 10. Cooking preferences of consumers

#### Discussion

The questionnaires are main methods to collect data and information about an issue. Therefore, this method was applied to the people living in Çanakkale and fish consumption habits were assessed. Socio-economic and demographic information about people could help to predict possible fish consumption prefers of people. In this context, fish consumption habits were evaluated with regard to socio-economic structures and demographic characteristics of the participants.

The results of the present study revealed that 39% of respondents consume fish 1-2 kg per consumption. On the other Bayraktar et al. (2019) reported that 47% of participants consumed fish less than 1 kg. Arık Çolakoğlu et al. (2006) stated that 87.46% of respondents consumed 1-6 kg fish monthly. This wide range could not help to preciously determine the fish



consumption of surveyed population. Therefore, more narrow range should be provided in the questionnaire survey. However, this study revealed that fish consumption amount was higher compared to both studies of Bayraktar et al. (2019) and Arık Çolakoğlu et al. (2006). Moreover, fish consumption amounts per capita were also determined as 13 kg for Tokat (Erdal and Esengül, 2008), 14.16 kg (Abdikoğlu et al., 2015) and 14.69 kg (Abdikoğlu and Unakıtan, 2019) for Tekirdağ, 12.2 kg for Diyarbakır (Aydın and Odabaşı, 2017), 13.28 kg for Çan (district of Çanakkale) (Selvi et al., 2019). Annual fish consumption per capita were also identified 3.4 kg for Ankara (Özer et al., 2016), 3.8 kg for Niğde (Bashimov, 2017), 5.06 kg for Amasya (Kızılaslan and Nalinci, 2013), 2.98 kg for Antakya (Can et al., 2015), 8 kg for Adana and Mersin (Cengiz and Özoğul, 2019), 29.59 kg for Giresun and Trabzon (Aydın and Karadurmuş, 2013), 26.3 kg for Ordu (Aydın and Karadurmuş, 2013).

Çanakkale has advantages in terms of fish consumption due to its location along the coasts of Marmara Sea, Çanakkale Strait, and Aegean Sea. Therefore, fish consumption might be supported by recreational fisheries as described by Ünal et al. (2010). Authors reported that shore-based fishing (68%) was the most popular fishing type for the respondents. In the present study, 6.4% (68 individuals) of the participants indicated that they got their fish by fishing instead of buying. Similarly, 0.81% (55 individuals) of respondents obtained their fish by hand-line fishing from the shore.

Some authors reported that the price of fish is the most imperative factor for fish consumption (Boughanmi et al., 2007; Akpınar et al., 2009; Claret et al., 2012; Hanis et al., 2013; Geslani et al., 2015; Abdikoğlu and Unakıtan, 2019). However, in the present study, price is not affecting the fish consumption for consumers in Çanakkale. Income level, season, and freshness of fish are affecting the fish consumption for respondents. Similarly, Dal et al. (2019) noted that freshness of fish was the most important factor determining of criteria that affecting fish consumption. Arslan (2019) indicated that income level was one of the most important factor affecting the fish consumption in Erzurum. Lee and Nam (2019) put forward that fish consumption frequency is affected by some factors including residential area, household income, preference, price, safety, and favourable fish species. Moreover, wild caught/cultured status of fish species, the age, marital status and number of family members of consumers have no impact on the determining of fish consumption frequency. Similarly, there is no significantly relationship was found between the fish consumption frequency and marital status/age, number of family members/wild-cultured status in the present study.

Kale (2017a) reported that the increase in temperature trends caused to climate change. Author stated that Çanakkale city will be affected by global warming and the climate change, and also will have a warmer climate in the future. Similarly, Kale (2017b) also reported that annual evaporation will increase in the future similar to temperature. Climatic factors are affecting the production of food and agricultural products. Thus, potential impacts of the climate change could have adverse effects on the fisheries and aquaculture sectors. Therefore, the amount of fish consumption per capita should be increased for healthy life without affecting by the adverse impacts of the climate change on fish resources.

Fish consumption could be increased by improving the awareness of consumers about benefits of fish for health due to its high nutritional content. Global health organizations also recommend to the consumption nearly 300 grams of fish per capita once a week to live a healthy life. Likewise, increasing interest on dietetic aspects, waste reduction, food safety, and food quality has also supplemented the increase of the fish consumption (FAO, 2018). Therefore, consumers should be learnt about the vitality and benefits of fish consumption to increase the consumption amount.

# Conclusion

This paper determined the fish consumption habits in all districts of Canakkale. Both the most consumed and the most favourite fish species are identified as Sarda sarda. Fish consumption frequency was described as 33.3% for consuming fish one a week followed by bimonthly frequency (21.9%) and monthly frequency (21.5%). 2.3% of the participants noted that they never consume fish. Moreover, the majority of consumers specified that they consume fish 1-2 kg (39.0%) and 27.3% consume fish 0.5-1 kg. Socio-economic and demographic characteristics of consumers are affecting the fish consumption habits. In the present study, season, income level, and freshness of fish are found to be driving force for fish consumption in Çanakkale. Therefore, fish farmers and sellers recommended to remain the freshness of fish and to follow the appropriate fishing season for providing fish to consumers.

# Acknowledgements

This paper includes a part of M.Sc. thesis of Fahri Saka. Authors would also like to thank Semih Kale for his contributions and supports during the study.

# **Conflict of Interest**

Authors declare that there is no conflict of interest.



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# Marine Science and Technology Bulletin

# **RESEARCH ARTICLE**

# Characterization, identification and phylogeny of the creatine kinase (*ckma*) gene in medaka (*Oryzias latipes*)

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#### ARTICLE INFO

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# ABSTRACT

Creatine kinase (*ckma*) has been characterized and described in the medaka (*Oryzias latipes*), an aquatic model organism and the gene structure has been designed using the exons, introns, produced amino acids of the gene, TATA box, poly A tail and 5' UTR and 3' UTR regions of the *ckma* gene. In another step, firstly, the chromosome region of the *ckma* gene was determined in medaka and then the other genes which placed in the same region were determined. Then the locations of these genes were determined in zebrafish and human which are the orthologs of medaka. Finally, the conserved gene synteny was designed manually, using these data. However, genetic identity and similarity ratio between medaka and its orthologs were calculated. In this study, characterization and identification, phylogenetic relationship, conserved gene synteny of *ckma* gene in medaka (*O. latipes*) which is an important model organism were analyzed by using bioinformatics tools (NCBI database, Ensembl genomic database, Expasy, Reverse Complementary and some programs such as MEGA6 program, BLOSUM62 matrix program and BioEdit software). All these data will be used in future studies on molecular stress response in fish and they were presented to the scientific world with this study.

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Bayır, M., Arslan, G., Oğuzhan Yıldız, P. (2020). Characterization, Identification and Phylogeny of the Creatine Kinase (*ckma*) Gene in Medaka (*Oryzias latipes*). *Marine Science and Technology Bulletin*, 9(1): 15-22.

#### Introduction

**Bioinformatics** 

Medaka (*Oryzias latipes*) is a small freshwater fish lives in East Asia. It is an omnivore fish which feeds on vegetable animal foods such as phytoplankton and zooplankton (Hori, 2011). The male medaka can be easily distinguished from the female by its external morphology. Embryos are transparent. Medaka is the first vertebrate in which Mendel inheritance is also exhibited (Ishikawa, 2000; Jacquet et al., 2004; Shima and Mitani, 2004). Although the physiology, embryology and genetics of medaka (*Oryzias latipes*) have been extensively studied for the last 100 years, the studies carried out in this organism have focused on the use of genetic model systems for early development, pigmentation, sex determination and human diseases and the biological history of this fish in the recent years (Naruse et al., 2011). Medeka embryos are used



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especially in transplantation, microinjection, transgenesis and gene expression studies. Medaka has contributed to important steps in the studies on oncology, ecotoxicology, endocrinology and determination of conserved gene structure (Shima and Shimada, 1991, 2001).

Quantification of fish muscle protein levels indicates that creatine kinase is one of the most highly expressed proteins in fish muscle. This has both cytosolic and mitochondrial forms of regulation of energy production (mitochondria) and use (cytosol) through actions related to adenosine triphosphate (ATP) (McLean et al., 2007).

There is a chemical cycle in the muscle of alive fish. These chemical events provide energy to the muscle during the swimming of the fish and provide the substances necessary for growth and regeneration of dead tissues. Enzymes are substances that create and control chemical reactions in living muscle. Chemical energy is converted to mechanical energy for ATP production which provides the necessary energy. While ATP consumption regeneration and contraction-relaxation events are continuous in living tissue, the amount of ATP decreases rapidly after blood circulation and oxygen supply is cut off in post mortem tissue and contraction and relaxation events continue to be limited during this decrease. The energy required for muscle contraction in live fish is provided by ATP formed during glycolysis. ATP breaks down into adenosine diphosphate (ADP) and inorganic phosphate (P) by the ATPase enzyme, and the energy is used for muscle contraction. ADP and creatine are catalyzed by the creatine kinase enzyme to regenerate ATP from phosphate (Stryer, 1995).

Genetic similarities among species present in all organisms mean that studies on one organism can be used as a data source for other species (Collins et al., 1998). Therefore, in this study, the bioinformatics of ckma gene in aquatic model organism, medaka (O. latipes) will be completed and the leading data will be provided for molecular studies in other fish.

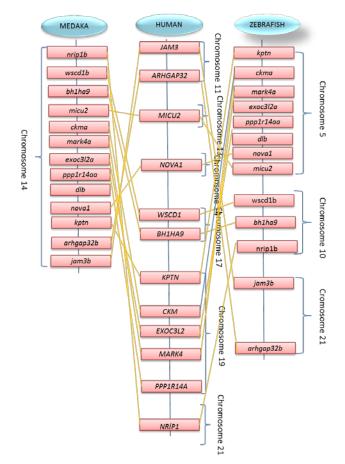
# Material and Methods

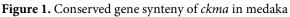
# Bioinformatics of ckma gene in medaka (O. latipes)

In this study, firstly The National Center for Biotechnology Information (NCBI) (http://www.ncbi.nlm.nih.gov/) was used to investigate whether the creatine kinase (ckma) gene functional in medaka (O. latipes) and then its cDNA sequence was obtained from ENSEMBL. However, ensembl database was used to characterize the *ckma* gene in medaka (O. latipes).

We determined that this gene encode a 381 amino acid protein and has isoform а single (https://www.ensembl.org/Oryzias latipes/Info/Index) and its ENSEMBL ID and UNIPROT ID have been found as ENSORLT00000033423.1 and A0A3B3I369, respectively.

In the next step, location and chromosome of these genes in zebrafish (Danio rerio) and human (Homo sapiens) were determined (Table 1) and manually conserved gene synteny was designed (Figure 1) in order to prove the conservation of these genes in these two orthologs of medaka.



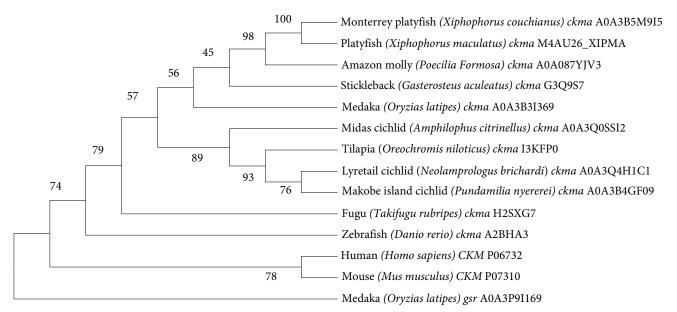


For the designing of phylogenetic tree among medaka (Oryzias *latipes*), Monterrey platyfish (Xiphophorus couchianus), platyfish (Xiphophorus maculatus), Amazon molly (Poecilia formosa), stickleback (Gasterosteus aculeatus), Midas cichlid (Amphilophus citrinellus), tilapia (Oreochromis niloticus), lyretail cichlid (Neolamprologus brichardi), Makobe island cichlid (Pundamilia nyererei), fugu (Takifugu rubripes), zebrafish (Danio rerio), human (Homo sapiens), mouse (Mus musculus) ckma/CKM gene sequences aligned by BioEdit (http://www.mbio.ncsu.edu/bioedit/page2.html) using CLUSTALW (Thompson et al., 1994) and then MEGA6 (Tamura et. al., 2013) program was used according to the maximum likelihood method (Kell et al., 2018) (Figure 2). glutathione reductase (gsr) Medaka (Oryzias latipes) (A0A3P9I169) was chosen as an external group.









**Figure 2**. Phylogenetic tree of *ckma* in medaka (*O. latipes*). Phylogenetic relationships between *ckma* sequence from medaka and the other vertebrates. Tree was produced using Maximum Likelihood method (Felsenstein, 1989). Accession numbers (UNIPROT) of the sequences used for phylogenetic tree are shown in phylogenetic tree.

Come	Come and al	Medaka		Zebrafish		Human	
Gene	Gene symbol	Chromosome	Location	Chromosome	Location	Chromosome	Location
Creatine kinase, muscle a	ckma	14	2.16	5	36.83	19	45.30
Junctional adhesion molecule 3b	jam3b	14	1.79	21	24.98	11	134.06
Rho GTPase activating protein 32b	arhgap32b	14	1.88	21	24.53	11	128.96
Neuro-oncological ventral antigen 1	nova1	14	1.98	5	36.61	14	26.44
Kaptin, actin binding protein	kptn	14	1.96	5	36.91	19	47.47
DeltaB	dlb	14					
Exocyst complex component 3-like 2a	exoc3l2a	14	2.41	5	3.67	19	45.21
Protein phosphatase 1 regulatory inhibitor subunit 14A	ppp1r14aa	14	2.10	5	36.73	19	38.51
Microtubule affinity regulating kinase 4a	mark4a	14	2.14	5	36.76	19	45.07
Mitochondrial calcium uptake 2		14	2.26	5	36.59	13	21.49
Basic helix-loop-helix family member a9		14	2.35	10	37.92	17	1.27
WSC domain containing 1b	wscd1b	14	2.40	10	37.98	17	6.05
Nuclear receptor interacting protein 1	nrip1b	14	2.65	10	8.25	21	14.96

For the design of gene structure, ENSORLT00000033423.1 cDNA transcript of medaka (*O. latipes*) *ckma* gene was used. exon-intron organization of the medaka (*O. latipes*) *ckma* gene

and the amino acids produced by the exons, the 5' UTR and 3' UTR regions of the *ckma* gene, the TATA box, the poly A tail, and the starting point of transcription (+1) were showed in the



gene structure (Table 2). Zebrafish (*Danio rerio*), Nile tilapia (*Oreochromis niloticus*), fugu (*Fugu rupripes*), human (*Homo sapiens*) and mouse (*Mus musculus*) ckma/CKM proteins were used in Bioedit program, CLUSTALW (Thompson et al., 1994) for analyzing the similarity-identity ratios (Table 3).

# **Results and Discussion**

# Bioinformatics of ckma gene in medaka (O. latipes)

Oxygen deficiency is a major factor in creatine increasing in fish, besides the impact of industrial enterprises' waste (Arslan, 2015). Stress responses of vertebrates include different interactions between physiological pathways that can be characterized in both acute and chronic conditions. Creatine kinase (CK) is an important enzyme used in the detection of damage to tissues and organs such as glutamic-pyruvic acid transaminase (GPT), glutamic-oxaloacetic acid transaminase (GOT), alkaline phosphatase (ALP) and lactic dehydrogenase (LDH) enzymes. These enzymes, except from CK, are liver enzymes and those are also used to understand liver problems.

CK and GOT enzymes tend to increase in wounds on fish skin and in case of damage to muscle tissue and brain. In addition, the CK enzyme allows the regeneration of ATP in contraction or delivery systems. Therefore, the completion of the detailed bioinformatics study of the creatine kinase (*ckma*) gene, which is one of the stress markers, in the medaka (*O. latipes*) (Iwama et al., 1999) is important. Therefore, it is of great importance to complete detailed bioinformatics study of the creatine kinase (*ckma*) gene which is one of the stress markers in medaka (aquatic model organism) has great importance, because acute or chronic stress responses of fish change with environmental differences.

Because fish are aquatic organisms, changes in both qualitative and quantitative properties of water can lead to changes in the functional structures of these organisms, resulting in unfolding of protein folds from time to time, and these proteins can combine with other proteins in the cell to form clusters. Consequently, proteins may lose their functions due to conformation deformation (Basu et al., 2000). However, in this research, firstly, ckma gene was determined to be a functional gene in medaka (O. latipes) by using of bioinformatics tools, and then the other bioinformatics studies were carried out such as gene structure determination, phylogenetic tree design, conserved gene synteny and calculation of the identity-similarity rates between medaka (O. latipes) and its orthologs. When a molecular study is planned, firstly bioinformatics studies should be completed before experimental studies to understand how the expression of genes

changes with various stress factors. Therefore, this study will provide important bioinformatics data both for fish physiology studies and for the other studies on vertebrates because medaka (*O. latipes*) is an aquatic model organism.

In this study, ENSEMBL, UNIPROT, NCBI databases and BioEdit software, BLOSUM62 matrix program and MEGA6 program were used to reach some knowledge such as the cDNA, exons and introns of the *ckma* gene, the amino acids produced by this gene, the 5' UTR and 3' UTR regions, the chromosome and location where the gene is positioned, and the protein sequences necessary to determine the phylogenetic relationship to other vertebrates. The cDNA sequence of the medaka (*O. latipes*) *ckma* gene was obtained from the Ensembl database (Ensemble number ENSORLT00000033423.1) and it was found that this gene has a single isoform, which encoded a protein of 381 amino acids. Medaka *ckma* gene has 7 exons and 6 introns located between these exons. The amino acids produced by the exons and the 5' and 3' ends of the gene, TATA box and Poly A tail are given in detail in Table 2.

The sequence identity-similarity ratio was calculated to investigate the orthology between the medaka (*O. latipes*) and zebrafish *ckma* gene. For this purpose, medaka (*O. latipes*), zebrafish (*Danio rerio*), fugu (*Fugu rupripes*), Nile tilapia (*Oreochromis niloticus*) protein sequence produced by *ckma* gene and mouse (*Mus musculus*) and human (*Homo sapiens*) protein sequences produced by CKM gene were aligned using the BioEdit program in the BLOSUM62 matrix algorithm, and the similarity-identity ratios of these organisms were calculated (Gromiha, 2010) and the results were given in Table 3. According to the table, the identity and similarity percentage of medaka (*O. latipes*) *ckma* gene was 98-94% with Nile tilapia, 97-93% with zebrafish, 96-91% with fugu, 93-87% with human, and 92-87% with mouse (Table 3).

In order to define the conserved genes in both medaka and zebrafish and human, the location of *ckma* gene was determined on the 14th chromosome in medaka. Then the other genes and their locations were determined in this chromosome using the Ensemble genome database (Table 1). Conserved gene synteny was determined by detecting the chromosomes and regions of these detected genes (*ckma*, *jam3b*, *arhgap32b*, *nova1*, *kptn*, *dlb*, *exoc3l2a*, *ppp1r14aa*, *mark4a*, *wscd1b*, *nrip1b*) found in human and zebrafish (Figure 1). These genes on chromosome 14 in medaka (*O. latipes*) are also conserved in humans (chromosomes 11, 13, 14, 19 and 20) and zebrafish (chromosomes 5, 10 and 21). It is known that teleost fish have evolutionary conserved regions in the same gene family, and the designed conserved gene synteny clearly demonstrates it. In addition, it is thought that the *ckma* gene of **Table 2.** Gene structure of *ckma* in medaka (*Oryzias latipes*)

Table 2. Gene structure of ckmu in medaka (Oryzius unipes)				
5'taaactgcaaggacttgaagggtaaaaggccagatattctggggctaaaaatacccgg	-299			
agagcaggctctccacccctgctcaatttcaactggacatctgagccactggaaactgag	-239			
	-179			
cgacacttgttaccaagaatctgcggacagcaccgtttgaaatttgcagctgcccaaaat				
gtcatatgctcaaagaaggaaaaagcatcatttgcagcgtccttgctcctctttatgaa				
tgaggctgcaatgacctgtcttcattgtatt <mark>ATATA</mark> gcctaagcttgttgtgtttttcag	- 5 9			
+1				
TGTTAGAAAGCAATC <u>ATGCCTTTCGGAAACACCCCACAACAACTTCAAGCTCAACTACTCCA</u>	60			
-MPFGNTHNNFKLNYS-				
<u>GTTGACGATGAGTTCCCAGACCTGTCCAAGCACAACAACCACATGGCCAAAGTCCTGACT</u>	120			
-VDDEFPDLSKHNNHMAKVLT-				
<u>AAAGAGCTGTATGGTAAGATGAGGGACAAGCAGACGCCCACTGGATTCACTCTGGATGAC</u>	180			
-KELYGKMRDKQTPTGFTLDD-				
GTGATCCAGACCGGCATCGACAACCCTG gtgagacttcaagcaacatttcttctttttc	240			
-VIQTGIDNP				
caacagaatccaagatagtaaaagacaagaaacaagtgttagggtcaattcataaccccc	300			
acctttgttatcagGTCACCCCTTCATCATGACTGTTGGCTGTGTCGCTGGTGACGAGGA	360			
GHPFIMTVGCVAGDEE				
GTCTTATGAGGTCTTCAAAGACCTGCTTGACCCCGTCATCTCTGACCGTCATGGTGGATA	420			
SYEVFKDLLPVISDRHGGY				
TAAGCCCACTGACAAGCACAAGACTGACCTCCAACTTCGAGAACTTGAAGqtqcaatacaq	480			
KPTDKHKTDLNFENLK-	100			
cttctttagagagcagagttacacctagccctttctaatgttcctcacggcccaatctaa	540			
ctqtqtctqtaq <b>GGAGGTGATGACCTGGACCCCAACTACGTTTTGTCCAGCCGTGTTCGT</b>	600			
-GGDDLPNYVLSSRVR-	000			
	660			
ACCGGTCGCAGCATCAAGGGATACGCCCTGCCCCCCCACAACAGCCGTGGCGAGCGCAGA	660			
-TGRSIKGYALPPHNSRGERR-	=			
GCTATTGAGAAGCTGTCCATTGAGGgtaagttttcttgattttggggatttccacaggtc	720			
-AI-EKLSIE				
aagagtatctgatacccaggtttctgtggtcagtcataaaccagactgaatccaggcttt	780			
ctgctctagcaggtcttctaaatcatcatgcaatgcctaatgcatcgatgtatgaaataa	840			
agaagtgttctgttttttggtggatgctgacctaacagtgagcctcttcctgcag <mark>CTCTG</mark>	900			
A L -				
<u>TCCAGCCTTGATGGTGAGTTCAAAGGAAAGTACTATCCCCTGAAGTCAATGACTGATGCT</u>	960			
-SSLDGEFKGKYYPLKSMTDA-				
GAGCAGGAGCAGCTGATCAGTGATCACTTCCTGTTTGACAAACCTGTGTCCCCCCTGTTG	1020			
-EQEQLISDHFLFDKPVSPLL-				
ACCTGCGCCGGTATGGCCCGTGACTGGCCTGACGGCAGAGGCATTTGgtaagtgcagtta	1080			
-T-C-A-G-M-A-R-D-W-P-D-G-R-G-I-W				
ggaatggtcatcctctgtaaatacaccaaacactcagcttgtatagattcatcaggatta	1140			
atcactgacctgcgtagtgctgtccatggtcagtgtccataaatcaagcaag	1200			
tgtctgagcagtcagagtacaactggaaaacatccacaaatgagtcctcaaggatttcct	1260			
ggcagggaaatcatgatggcagtagatacattgggctctgagcttaaattctcattggtc	1320			
tgcaagatattgcacattgtccaaatctgtgcccgttggcatctctacatccag <b>GCACAA</b>	1380			
-HN	1000			
	1440			
CGACAACAAGACCTTCCTGGTGTGGGGTGAATGAGGAGGATCACCTGCGTGTCATCTCCAT	1440			
	1 5 0 0			
GCAGAAGGGTGGCAACATGAGGGAGGTCTTCAGGCGTTTCTGCGTGGGCTTGCAGAAGgt	1500			
QKGGNMREVFRRFCVGLQK-				
gcatgaagaccgcagatcaaatctgctcagcctgtttaaccaagtcaaacctaaagcagc	1560			
tgtgatcctgacccttcttttatgactctcag <mark>ATTGAGGAGATCTTCAAGAAGCACAACC</mark>	1620			
- I E E I F K H N				
<u>ACGGCTTCATGTGGAATGAGCATCTCGGCTACATTCTGACCTGCCCCTCCAACCTGGGAA</u>	1680			
HGFMWNEHLGYILTCPSNLG				
<u>CTGGTCTGCGTGGGGGTGTCCACGTCAAGCTGCCCAAGCTGAGCACACCCCCAAGTTTG</u>	1740			
TGLRGGVHVKLPKLSTHPKF				
AGGAGATCCTCACCAGGTTGCGCCTGCAGAAGCGTGGCACAG gtatggatgtgctccatc	1800			
EEILTRLRLQKRGT				
tgtgggacctctacagaggctctgtggacgctcgtatgaggtgttatgtcatgccacatc	1860			
ctttctctccagGTGGTGTGGACACTGCATCTGTGGGTGGTGTGTTTGACATCTCCAATG	1920			
GGVDTASVGGVFDISN				
CCGACCGTCTTGGATCCTCCGAGGTGGCGCAGGTCCAGTTGGTGGTTGATGGCGTCAAGC	1980			
ADRLGSSEVAQVQLVVDGVK				
TGATGGTTGAGATGGAGAAGAAGCTCGAGAAGGGAGAAGCCATCGACAGCATGATCCCCG	2040			
LMVEMEKKLEKGEAIDSMIP				
<b>CCCAGAAGTGA</b> ggagggacaatctggcattttccttgtgaccttttatgtgcagtcgagc	2100			
A - O - K - * -				
cagctgacagcgtgcctgcagagaaaacagccgctcacctagagactcttgactctgcta	2160			
actcotttoottoottocagotttgttttttottttotoottoottgtogtttttttoacg	2220			
ttcccctgcgttggtcagtaacatccaggggggcagcctcactgagcggggcttgcctagc	2220			
	2340			
gttcAATAAAAcagcgtcccctgaacacgtctgggtcatccctgtctttctt	2400			
Note: The evens of the dama are shown in conital latters and the nucleotide positions are numbered a	4 4 4			

*Note:* The exons of the *ckma* are shown in capital letters and the nucleotide positions are numbered at the end of the each line. The starting site of transcription is +1,5' upstream sequence, 3' downstream sequence and introns are shown in lower case. The TATA box and the poly adenylation signal (AATAAAA) are shown in capital letters and painted in yellow. Amino acids are shown in capital letters which are placed under exons. Stop codon (TGA) is specified asterisk.





Tat	<b>ble 3.</b> Ide	ntity a	nd similarity rate between medaka (Me) and Nile tilapia (Nt), zebrafish (Zf), fugu (Fu), humar	
Me	ckma	1	${\tt MPFGNTHNNFKLNYSVDDEFPDLSKHNNHMAKVLTKELYGKMRDKQTPTGFTLDDVIQTG$	
	ckma	1	S.VLS.Y	
Ζf	ckma	1	MLSV	
	ckma	1	.AKCDY.MKMOEQIL.G.SSV	
	CKM	1	K.LKPEE.YLK.LESV	
Мо	CKM	1	KKPQE.YPDN.LES	
Me	ckma	61	IDNPGHPFIMTVGCVAGDEESYEVFKDLLDPVISDRHGGYKPTDKHKTDLNFENLKGGDD	
	ckma	61	VH	
	ckma	61	VA	
	ckma	60	VA	
	CKM	61	VH	
Мо	CKM	61	VH	
-	ckma		LDPNYVLSSRVRTGRSIKGYALPPHNSRGERRAIEKLSIEALSSLDGEFKGKYYPLKSMT	
Nt	ckma		N	
	ckma		· · · · · · · · · · · · · · · · · · ·	
	ckma	120	A	
	CKM		VNT	
Мо	CKM	121	VNT	
-	ckma	-	${\tt DAEQEQLISDHFLFDKPVSPLLTCAGMARDWPDGRGIWHNDNKTFLVWVNEEDHLRVISM}$	
	ckma	181		
	ckma	181	AALAAEE.	
	ckma	180	A	
	CKM		EKQDLASASS	
Мо	CKM	181	EQ.QDLASAS	
Me	ckma	241	QKGGNMREVFRRFCVGLQKIEEIFKKHNHGFMWNEHLGYILTCPSNLGTGLRGGVHVKLP	
Nt	ckma	241	DD	
Ζf	ckma	241	KK	
$\mathbf{Fu}$	ckma	240	KAA	
Hu	CKM	241	EK	
Мо	CKM	241	EK	
Me	ckma	301	KLSTHPKFEEILTRLRLQKRGTGGVDTASVGGVFDISNADRLGSSEVAQVQLVVDGVKLM	
	ckma		E	
	ckma		AEC	
Fu	ckma		Q	
Hu	CKM	301	HKE	
Mo	CKM	301	NK	
			Identity (%) Similarty (%)	
Me	ckma	361	VEMEKKLEKGEAIDSMIPAQK 100 100	
	ckma		•••••• <b>s</b> •••••• 98 94	
Ζf	ckma	361	•••••• <b>5</b> •••••• 97 93	
Fu	ckma	360	••••••••••••••••••••••••••••••••••••••	
Hu	CKM	361	93 87	
Мо	CKM	361	QSD 92 87	

Table 3. Identity and similarity rate between medaka (Me) and Nile tilapia (Nt), zebrafish (Zf), fugu (Fu), human (Hu) and mouse (Mo)

*Note:* The dots and lines refer to repeating amino acids and undetectable amino acids, respectively.

medaka emerged as a result of teleost genome duplication seen in bony fish. As known, teleost fish may have two copies of genes found as a single copy in other vertebrates as a result of whole genome duplication (Amores et al., 1998; Meyer and Schartl, 1999; Postlethwait et al., 2000; Braasch and Postlethwait, 2012; Çapan, 2019). It was observed that tilapia, puffer fish, stickleback, platyfish, Midas cichlid, Makobe island cichlid, fugu, Amazon molly and medaka have just one copy of

the creatine kinase gene (*ckma*), while zebrafish has two copies of this gene, *ckma* and *ckmb*, when explored Ensembl database. In this case, it is thought that one copy is lost following teleost whole genome duplication in these species except from zebrafish. Yamamoto (1953), firstly created a gender linkage map for medaka and described differences in the frequency of recombination between genders. It was also reported for the first time that there was an autosomal connection between *i* and





ci loci in fish. Following the development of polymerase chain reaction (PCR) technology, several attempts have been made to create a genetic linkage map in medaka, zebrafish, puffer and other fish species, and finger-print markers were used in the early stages of these experiments, as they did not require prior genome information. In subsequent steps, single locus markers were used to amplify specific regions of the genome in the presence of sequence information, and the map generated using activated single locus markers was used to compare linkage relationships between orthologous genes. All genome amplification specific to the teleosts were then applied (third WGD). Finally, in addition to the tetraodon genome project, the medaka genome sequencing project provided a high quality outline genome sequence for both medaka and tetraodon. All these data confirmed the third WGD, which revealed a potential scenario in which reconstruction of protochromosomes prior to duplication and the formation of existing medaka, tetraodon and zebrafish genomes.

Phylogenetic relationship can be seen in the tree (Figure 2) which created using protein sequences of medaka (*O. latipes*), Monterrey platyfish (*X. couchianus*), platyfish (*X. maculatus*), Amazon molly (*P. formosa*), stickleback (*G. aculeatus*), Midas cichlid (*A. citrinellus*), tilapia (*O. niloticus*), lyretail cichlid (*N. brichardi*), Makobe island cichlid (*P. nyererei*), fugu (*T. rubripes*), zebrafish (*D. rerio*), human (*H. sapiens*) and mouse (*M. musculus*) according to maximum likelihood method using MEGA6 (Tamura et. al., 2013) program. It was observed that the medaka showed clustering with other teleost fishes, and that living organisms such as humans, chickens and mice were clustered in a different region (Figure 2).

# **Conflict of Interest**

The authors declare that there is no conflict of interest.

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# Marine Science and Technology Bulletin

# **RESEARCH ARTICLE**

# First molecular record of the alien species Pacific oyster (*Crassostrea gigas*, Thunberg 1793) in the Marmara Sea, Turkey

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# ABSTRACT

The Pacific oyster (Crassostrea gigas) has a very important economic potential for aquaculture, but on the other hand, is among the highly invasive species in the world and within the Mediterranean ecosystem. In the 1960s, C. gigas was brought to Europe for aquaculture in the Mediterranean and Black Sea regions from Japan and Canada. The Turkish waters are the part of the Mediterranean Sea, which is the world's most invaded sea. The invasion of alien species results from marine transportation and aquaculture activities of non-native species. A heavy maritime traffic is also present in the Marmara Sea, which connects the Black Sea and Mediterranean Sea. The identification of the invasive species and their distributions is very prominent in terms of protecting natural habitat and monitoring the effects of invasive species. In this study, 30 individuals, morphologically identified as C. gigas, were collected from Bandırma bay. The genomic DNAs were extracted from each sample's muscle tissue using universal salt extraction method. Partial sequences of COI and 16S Mitochondrial DNA loci of the sample DNAs were obtained for species identification. The sequences were searched against the database and results were retrieved from BLAST. All the sequences obtained in this study showed significant similarity with the C. gigas sequences present in the database (E=0). The sample sequences resulted in 9 different haplotypes for the COI locus (hd: 0.5296 and variance: 0.01256±0.112) and 5 different haplotypes for the 16S rDNA locus (hd: 0.2529, Variance: 0.01076±0.104). The results of this study provided the first molecular evidence for the presence of non-native Pacific oyster individuals in the Marmara Sea.

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# Introduction

Oysters are bivalves widely distributed all around the world estuaries. They are benthic, sessile filter-feeders, and reefbuilders that are playing important roles in estuary ecosystem (Ren et al., 2016). The Pacific oyster (*Crassostrea gigas*) is one of the world's 20 most cultured species with high economic values owing to their useful traits for aquaculture like efficient filter feeding, high growth rates, strong reproductive ability and tolerance to a wide range of environmental conditions (Laugen et al., 2015). However, it is also one of the most invasive species and may exert some negative impacts on native oyster species. The possible effects of the invasive species on the native species are; sharing the same area and food resources, genetic pollution due to hybridization, introgression and decrease of genetic diversity.

As human population continues to grow, the demand on seafood continuous to increase as on any other food sources. Aquaculture is important to ensure a consistent supply of aquatic species as harvesting the wild populations (fish, crustaceans and others) cannot keep up with the increasing human population's demand. For example, *C. gigas* production in 2016 by fishery was 17370 tons meanwhile its production by aquaculture was 639030 tons (FAO, 2020).

The spread of economically important, but invasive species throughout the world has been greatly facilitated by means of aquaculture, maritime transportation and the trade of aquatic organisms (Crocetta et al., 2015). The Mediterranean Sea is the world's most invaded sea. A total of 5% of the whole marine species in the Mediterranean habitat is considered non-local, 13.5% of these species are considered as invasive species and this ratio is increasing due to abovementioned human activities (Galil, 2009, Zenetos et al., 2012; Segvic-Bubic et al., 2016). These activities also lead to the transport of invasive species from the Mediterranean to the Marmara Sea (Çınar et al., 2011).

Mollusks show an important native distribution in the eastern and middle Mediterranean. The European flat oyster (*Ostrea edulis*) is a native oyster species in the Mediterranean region. This species live in muddy, muddy sandy, rocky, muddy pebbly and dense alluvium. They feed on microalgae and they either live freely or by fixing themselves with their right shells in coastal waters (Tebble, 1966). In economic and food quality terms, *O. edulis* is a very valuable species in the markets (Yildiz et al., 2011; Acarli et al., 2015; Smyth et al., 2018). Unlike *O. edulis*, *C. gigas* is not a native species in Mediterranean region. On the contrary, it is a black-listed invasive species in conservation programs prepared for its non-native Mediterranean ecosystem (DAISIE, 2016).

The Pacific oyster is a particularly euryhaline and eurythermal species. Its salinity and temperature tolerances vary widely (Miossec et al., 2009). It attaches to rocks, debris and shells and found from the lower intertidal zone to depths of 40 m. It is naturally found in the northeastern Asia and had been widely distributed in the tropical seas (Zibrowius, 1992; Galil, 2000). It has become a popular species for aquaculture in Europe in the second half of the 20th century (Lallias et al., 2015). The aquaculture trials of C. gigas started in the south of France using the imported breeding populations from Japan and Canada in the late 1960s (Grizel and Heral, 1991). Then, they were found in Adriatic and soon, their distribution expanded from Cyprus to Tunisia (Dridi et al., 2006) including most regions of the Mediterranean. In 1991, an aquaculture study was conducted in Homa lagoon area in Izmir using the juvenile samples obtained from France (Özden et al., 1993). The breeding practices have resulted in the establishment of wild C. gigas populations in the Black Sea, the Mediterranean Sea and along the Atlantic European coasts (Nehring, 2011; Angles d'Auriac et al., 2017).

Oysters are easily affected by environmental changes and show a wide variety of morphological traits such as shell formation and color, and these factors make the accurate identification of the oyster species very difficult (Galvão et al., 2017) and may lead to taxonomic misclassifications and misidentifications (Lam and Morton, 2006; Liu et al., 2011; Pagenkopp Lohan et al., 2015; Ren et al., 2016). Therefore, besides the morphometric measurements, the use of genetic markers (e.g. SNP, RAPD, RFLP, microsatellites, etc.) is inevitable. The use of genetic markers is also very valuable in studies with different aims (Işık, 2019; Işık and Bilgen, 2019; Özdil et al., 2019). In the last few decades, the developments in the molecular science have provided better results for species identification employing suitable molecular tools (Reece et al., 2008; Salvi et al., 2014; Pagenkopp Lohan et al., 2015). DNA barcoding analysis provides high accuracy in identifying species with high morphological plasticity, based on a standard mitochondrial cytochrome c oxidase subunit I (COI) and 16SrDNA fragments (Lapègue et al., 2002; Boudry et al., 2003; Hebert et al., 2003; Varela et al., 2007; Lazoski, 2011; Keskin and Atar, 2013; Crocetta et al., 2015; Segvic-Bubic et al., 2016; Galvão et al., 2017).

The first records about the existence of *C. gigas* in Turkey was reported in (i) Marmara Island, Southern Marmara Sea, by Yüksek (1989); (ii) Tuzla, Levantine Sea by Çevik et al. (2001) (ii) Çeşme, Aegean Sea by Doğan et al. (2007), and (iv) Marmara Sea (Acarlı et al., 2017). These studies were based on morphologic investigations. However, Özcan Gökçek et al. (2017) identified oysters from *Crassostrea* genus among the



samples collected from the north Aegean Sea by using RAPD technique. The present study aimed to genetic identification of the morphologically identified non-native oysters found in the southern Marmara Sea based on two molecular markers; partial COI and 16S rDNA sequences.

# Material and Methods

# Sampling

A total of 30 individuals were collected from Bandırma Bay, the Marmara Sea (40°22'03.43"N, 27°55'29.47"E) (Figure 1). The individuals were selected as they all had Pacific oyster (*C. gigas*) shell characteristics (Figure 2).

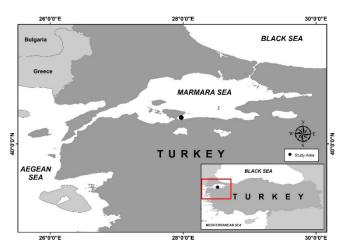


Figure 1. Sampling location of the study



Figure 2. Oyster samples collected from Bandırma Bay, Marmara Sea

# DNA extraction, PCR amplification and sequencing

The adductor muscles were taken from live samples and stored at -20°C until DNA extraction. Genomic DNAs of the samples were extracted using Universal-Salt Method (Aljanabi and Martinez, 1997). The quality and quantity of the extracted DNA were checked by both agarose gel electrophoresis and spectrophotometry techniques. The RedSafe (Intron-Korea) dye was used to stain and visualize the DNA bands under UV light. For the PCR amplification of the COI gene, the universal primers (LCO1490 and HCO2198) designed by Folmer et al. (1994) were employed. In addition, the primers (16S.AR and 16S.BR) designed by Palumbi (1996) were employed for the PCR amplification of the 16S gene. The 30  $\mu$ L PCR volume contained: 50-100 ng genomic DNA, 0.4  $\mu$ M of each primer, 1×PCR Buffer, 200 $\mu$ M dNTP, 2.5mM MgCl<sub>2</sub> and 0.6U of Taq DNA polymerase (i-Star Taq, Intron- Korea). The cycling protocol was 1 min at 94°C, 30 cycles of 94°C for 45 s annealing temperature (50°C for COI and 55°C for 16S gene) for 90 s, 72°C for 60 s with a final extension at 72°C for 10 min (Liu et al., 2011) annealing.

Having checked the PCR amplicons by electrophoresis, all the quality PCR amplicons were sent to Medsantek (Istanbul, Turkey) for sequencing by an automated capillary electrophoresis system (Applied Biosystems, 3500xL Genetic Analyzer, Thermo Fisher Scientific, UK). The electropherograms were carefully checked by Chromas Pro v1.42 (Technelysium Pty. Ltd. Australia) for miscalls and base spacing. Afterward, the contigs were formed for each sample individually by aligning its forward and reverse sequences, and a final data file consisting of consensus sequences for each sample was obtained. These sequences were deposited in the NCBI GenBank database (MN862563, MN862564, MN862565, MN862566, MN862567, MN862568, MN862569, MN862570, MN862571, MN862572, MN862573 MN862574, MN862575, MN862576).

# Data analysis

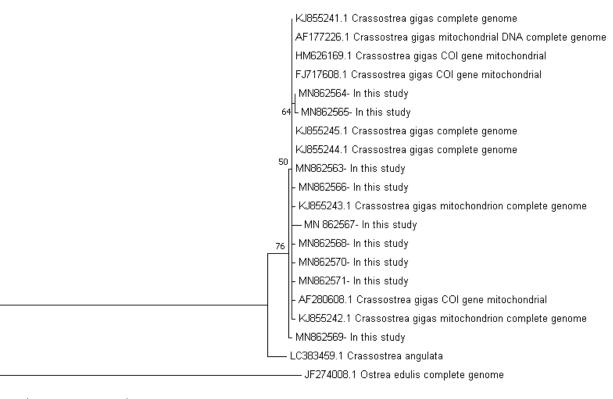
The BIOEDIT software (Thompson et al., 1994) was used for multiple sequence alignment of the consensus sequences and trimming of both ends to prepare the data file for further statistical analysis. Later, the trimmed file consisting of COI and 16S gene nucleotide sequences was analyzed by the software DnaSP v5. (Librado and Rozas, 2009) for estimating the haplotype and nucleotide diversity parameters. Afterwards, the sequence data obtained for the COI and 16S regions and the reference sequences taken from GenBank were used in reconstruction of the phylogenetic tree based on Maximum Likelihood (ML) method applying HKY nucleotide substitution model for COI and T92 nucleotide substitution model for 16S rDNA by MEGA (Molecular Evolutionary Genetics Analysis) software version 7 (Kumar et al., 2016). The nucleotide substitution models were selected based on the results obtained from ModelTest implemented in the software MEGA. In order to test the reliability of the tree topology, bootstrapping (×1000) was performed.





# Results

A total of 60 DNA sequences from 30 individuals and two loci were obtained. The partial mtDNA COI sequence (655 bp long) revealed 11 polymorphic sites leading to 9 different haplotypes (hd: 0.5296 and variance:  $0.01256\pm0.112$ ). One of these 9 haplotypes had a very high frequency (20/30).The 492 bp long partial 16S rDNA sequence revealed 4 mutations leading to 5 different haplotypes (hd: 0.2529, Variance:  $0.01076\pm0.104$ ). One common haplotype was observed in 26 individuals. The nucleotide sequences of the COI and 16S rDNA were found to be 98-99% identical with *C. gigas*'s mt genome when searched against the database using BLAST. For the phylogenetic reconstruction based on the 9 different mtDNA COI sequences (representing the 9 different haplotypes), some reference sequences were retrieved from the database initially. These sequences belonged to *C. gigas* (KJ855241, AF177226, HM626169, FJ717608, KJ855242-KJ855245, AF280608), *Crassostrea angulata* (LC383459) and *O. edulis* (JF274008) species. The Maximum Likelihood tree based on HKY nucleotide change model revealed one clade containing the *C. gigas* sequences from the database as well as all of the nine sequences of the present study (Figure 3). All the samples of the *Crassostrea* genus were separated from the *O. edulis* sample.



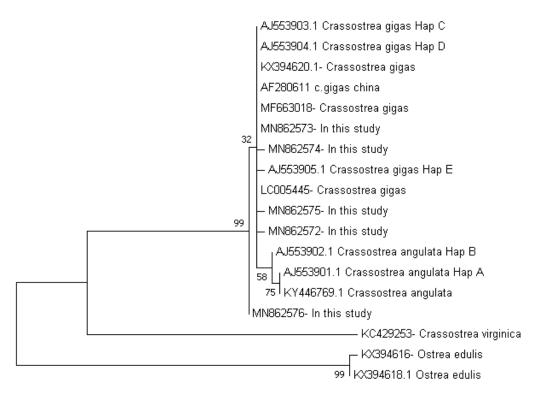
0.050

**Figure 3.** The ML tree was reconstructed based on the partial mtDNA COI sequences representing 9 haplotypes (MN862563-MN862571) of the present study and included the sequences of the species *O .edulis*, *C. gigas and C. angulata* that were retrieved from the GenBank. For this phylogenetic reconstruction MEGA 7 software was employed. Numbers at the nodes represent bootstrap supports.

In order to infer evolutionary relationship of the sequences obtained from the present study with the other Oyster species based on the partial 16S rDNA sequence, some reference sequences were also retrieved from the GenBank. These sequences belonged to *C. gigas* (AJ553903-AJ553905, KX34620, AF280611, MF663018, LC005445), *C. angulata* (AJ553901, AJ553902, KY446769), *Crassostrea virginica* (KC429253) and *O. edulis* (KX394616, KX394618) species. The ML tree based on

16S rDNA sequences and T92 nucleotide substitution model revealed one clade containing all the haplotypes of the present study together with the *C. gigas* sequences and *C. angulata* sequences from the database (Figure 4). Yet, the *C. angulata* sequences grouped together with a 58% node support. All these sequences separated from the *C. virginica* sample with a 99% bootstrap support. Furthermore, all the samples of the *Crassostrea* genus were separated from the *O. edulis* samples.





0.020

**Figure 4.** The ML tree was reconstructed based on the partial 16S rDNA sequences representing 5 haplotypes (MN862572-MN862576) of the present study and included the sequences of the species *O.edulis*, *C. gigas*, *C. angulata* and *C. virginica* that were retrieved from the GenBank. For this phylogenetic reconstruction MEGA 7 software was employed. Numbers at the nodes represent bootstrap supports.

#### Discussion

The sequences obtained in the present study clustered with the *C. gigas* samples obtained from the database. The two DNA markers employed in the study provided different resolutions when discriminating between the two closely related species: *C. gigas* and *C. angulata*. The partial mtDNA COI sequences revealed more haplotypes and separated species from each other statistical support (76%). Nonetheless, the16S rDNA sequences could not differentiate between these two species. Therefore, it can be suggested that the mtDNA COI gene provides better information in barcoding studies. Yet, it should be noted that the length of the sequences was different. The mtDNA COI sequences were 655 bp long, and the 16S rDNA sequences were 492 bp long. Increasing the sequences.

Although there is a study (Albayrak et al., 2004) mentioning the existence of *C. gigas* in the Marmara Sea; this is the first study investigating the presence of this species in this region based on molecular markers. Since oysters have high levels of morphological plasticity, it can be misleading to make identification only based on the morphological characters (Boudry, 2003). For instance; Segvic-Bubic et al. (2016) reported that some of the oyster specimens classified as *Crassostrea* clade according to the morphological investigations were actually *O. edulis* based on the 16s mitochondrial DNA marker. Therefore, it is important to use molecular markers as well as morphological measurements for species identification in oysters.

There has been no record of aquaculture practices for the Pacific oyster in Marmara Sea. It is known that *C. gigas* is capable of long-distance transport in the planktonic phase of 20-30 days (Schmidt et al., 2008). They are found around aquaculture areas and they can attach to the vessels. It is highly likely that human activities may induce their spread to nonnative ecosystems (Pecarevic et al., 2013). Therefore, it can be concluded that the transportation and spread of *C. gigas* to the Marmara Sea have probably occurred via vessels or water currents (Albayrak, 2011); the international maritime traffic being probably the main factor.

Considering the habitat preferences of *C. gigas*, Marmara Sea may provide a very suitable habitat for this invasive species due to its proper environmental conditions. Acarli et al. (2017) reported that the meat yield (AFNOR index-oyster quality) of *C. gigas* has changed from "fine" to "special" in the Bandırma Bay population. In this study, the oysters sampled for sequencing had an average length of  $88.02\pm22.26$  mm. These large individuals observed in the area and the DNA sequencing data obtained in this study provide support for the existence of a self-sustaining population of *C. gigas* in the southern Sea of Marmara. These results suggest that oysters had adapted to environmental conditions in Bandırma Bay such as temperature, salinity, etc., and showed good development performance when evaluated commercially. Furthermore, the large individuals in the study area indicate that the oysters have adapted and reproductive activity was performed. Similarly, Segvic-Bubic et al. (2016) provided the evidence of selfsustaining *C. gigas* populations in Adriatic Sea based on the mt 16S rRNA sequence analysis.

C. gigas is listed in the Delivering Alien Invasive Species Inventories for Europe (DAISIE, 2016). Due to the high physiological capacity and adaptation ability of Pacific oyster, the competition risk with other indigenous species is a very important issue (Laugen et al., 2015). C. gigas prefers similar habitats to the native blue mussel (Mytilus edulis) and Mytilus galloprovincialis found in different areas of Mediterranean and Atlantic coasts as reported by different studies (Diederich et al., 2005; Crocetta, 2011; Lipej et al., 2012; Dolmer et al., 2014; Angles d'Auriac et al., 2017). There are some negative impacts exerted by C. gigas on these native species such as competition for food and space (Nehls et al., 2006; Nehring, 2011). In addition, cross-fertilization may occur and hybridizations may be observed. During the sampling work of this study, it was observed that C. gigas shared the same beds with O. edulis (the native species) at the sampling site (Bandırma Bay, Marmara Sea) possibly causing competition for space and food between the two species.

# Conclusion

The Pacific oyster has been reported to cause a decline in natural populations of native oyster and mussel species, with which it shares the habitat and resources (Markert et al., 2009; Wilkie et al., 2012). As the presence of this species was confirmed for the first time based on molecular markers by this study, it can be a start signal for monitoring studies employing both molecular markers and morphological markers when assessing the status of both invasive and native species. Molecular markers are especially important when the species of interest has high phenotypic plasticity.

The native oyster species are part of their natural habitat and they have an economic value. However, they are under threat by invasive species. The invasive Pacific oyster *C. gigas* species have already established populations in the Mediterranean Sea. Considering the reports from Turkish waters based on morphology and the results of this study, it can be suggested that this species has already established populations in Turkish waters, too. Moreover, this species has a high economic value on its own, too. Immediate programs on monitoring the possible effects of Pacific oyster on *O. edulis* and the other bivalve species sharing the same habitat should be started in Bandırma Bay as well as in the other areas of the Marmara Sea. The results to be obtained from monitoring studies should aid in the development of accurate action plans for the sustainable protection of the ecosystem. In addition, even though currently it is not cultured/harvested for economic purposes, monitoring studies may help in the consideration of this invasive species economically.

# Acknowledgments

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# **Conflict of Interest**

The authors declare that there are no conflicts of interest to disclose.

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## Marine Science and Technology Bulletin

#### **RESEARCH ARTICLE**

Length-weight relationship of the most landed pelagic fish species European pilchard (*Sardina pilchardus* Walbaum, 1792) and European anchovy (*Engraulis encrasicolus* Linnaeus, 1758) in the Izmir Bay (Aegean Sea, Turkey) purse seine fishery

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ARTICLE INFO	ABSTRACT
Article History:	Length-weight relationships (LWR) of the most landed pelagic fish species Sardina
Received: 10.01.2020 Received in revised form: 04.02.2020 Accepted: 04.02.2020 Available online: 05.02.2020	<i>pilchardus</i> Walbaum, 1792 and <i>Engraulis encrasicolus</i> Linnaeus, 1758 in the Izmir Bay purse seine fishery were determined to reveal latest situation. Purse seine is a non-selective fishing gear compare to the other fishing gear such as gillnet or trammel net. For this reason, sampling all size individuals is very important to calculate mean length and other LWR parameters. In this study, seasonal LWR coefficient and minimum-maximum lengths
Keywords: Sardine pilchardus Engraulis encrasicolus LWR Izmir Bay Purse seine fishery	were established as monthly basis. LWR of <i>S. pilchardus</i> and <i>E. encrasicolus</i> were W = $0.0059L^{2.7930}$ (r <sup>2</sup> = 0.94) and W = $0.0019L^{3.4207}$ , (r <sup>2</sup> = 0.87), respectively. Growth type of the <i>S. pilchardus</i> was found negative allometric whereas <i>E. encrasicolus</i> was positive allometric. A decrease of the mean total length of <i>S. pilchardus</i> has been considerable variable from 1994 to 2014 in Izmir Bay but with this study, it is observed that mean length of the sardine found near of 2006 value related to seasonal fishing pressure.

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#### Introduction

The length-weight relationship (LWR) is an important tool in fish biology, physiology, ecology and fisheries assessment (Oscoz et al., 2005) and also, provide invaluable information on stock assessment studies (Moutopoulos and Stergiou, 2002; Gonzalez Acosta et al., 2004) for conversion of length observations into weight estimates to provide some measurements of biomass (Froese, 1998; Gonzalez Acosta et al., 2004).

Purse seine fishery is especially important for the Turkish fishery since it is the most important gear that targets small pelagic species especially anchovy and sardines as well as big pelagic species such as tunas. Once a fish school has been detected and surrounded by the purse seine net, there is no selectivity for individual size, species or catch quantity (Handegard et al., 2017). The catch quantity of a purse seiner is too much to compare with other fishing gears (e.g. trawls, seines). However, scientific studies on this fishing gear and method are quite limited in Turkey (Özbilgin et al., 2015).

Landing coming from purse seine accounts for about 30% of the world's total catch (Watson et al., 2006). Vast majority marine fish landing (approximately 60-70%) achieved by purse seine in 2018 fishing season (TurkStat, 2019). According to the official catch records, anchovy is the most landed fish species in Turkey with 96452 tons (43%). Although sardine landing is only 8.5% in Turkey, this value is substantially higher for the Aegean Sea (67%). Anchovy (12969 tons) and sardine (12654 tons) are the most landed pelagic fish species in the Aegean Sea (TurkStat, 2019). However, anchovy landing was the first time recorded higher than the sardine's in 2018 in the Aegean Sea.

So far, a few studies conducted to determine the LWR of *S. pilchardus* and *E. encrasicolus* with 10-year intervals (Hoşsucu et al., 1994; Özaydin and Taskavak, 2006; Acarli et al., 2014). For this reason, the purpose of the study is to reveal the current LWR parameters and compare it with the previous studies.

#### Material and Methods

During the study, a total of 567 of *S. pilchardus* and 212 of *E. encrasicolus* individual sampled in seven months. All the materials obtained from the monthly purse seine operations between September 28, 2017 and March 21, 2018 from Izmir Bay (Fig. 1) in depths between 26 and 60 m. The purse seine net used by the commercial purse seiner Afala 24 m LOA is overall 750 m in length, 164 m net in height and 14 mm mesh size. Purse seine is a non-selective fishing gear compare to the other fishing gear such as gillnet or trammel net. For this reason, sampling all size individuals is very important to calculate mean length and other LWR parameters.

In this study sampling was made only for seven months (three seasons) due to the 4/1 notification regulates commercial fishery by the Ministry of Agriculture and Forestry of Turkey. According to the regulation, there was a closed season for purse seine fisheries between 15th April and 31st August in Turkish waters. In the analysis of LWR, monthly data was converted to seasons and seasons converted to the total value. Final estimations made on the total values.

Total length (TL) of all individuals were measured to the nearest centimeter (cm), and wet weight (W) was recorded to the nearest gram (g). The functional relationship between the size and weight of the samples were fitted to the equation:  $W = aL^b$ , where W is the wet weight in grams, L the size in centimeters, a and b are the parameters to be estimated, with b being the coefficient of allometry (Ricker, 1975). The basic statistical data of the measured values were calculated and the relationships between them were determined (Sokal and Rohlf, 1973). Additionally, *t*-test was used for carried out to determine if the b coefficient was different from "3" (Sokal and Rohlf, 1969).

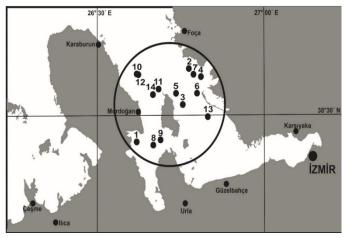


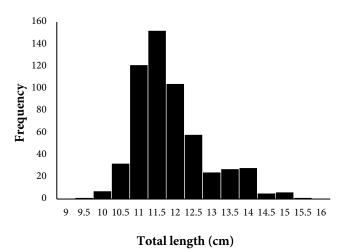
Figure 1. Sampling areas

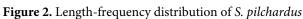
#### Results

The overall mean length of the *S. pilchardus* was found 12.1 cm. However, vast majority of the sardine individuals (91%) accumulated between 11.0 and 14.0 cm (Fig. 2). It was found that there was no significant allometry coefficients of LWR among seasons (Table 1) and also, the LWR curve of the *S. pilchardus* has shown in Fig. 3. The estimated total value of *b* coefficient indicating negative allometric growth (b=2.79; *t*-test,  $t < t_{0.05, n>500} = 1.65$ ) (Table 1). Furthermore, the r<sup>2</sup> values of *S. pilchardus* indicated a strong relationship between length and weight as 0.94.









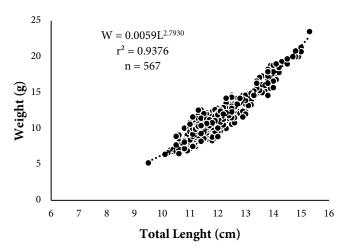


Figure 3. Length – weight relationship of S. pilchardus

Table 1. Overall estimated LWR values of S. pilchardus

		L	ength (cm	)	Weight (g)							
Seasons	Ν	$L_{min}$	L <sub>max</sub>	L <sub>mean</sub>	$\mathbf{W}_{\min}$	W <sub>max</sub>	W <sub>mean</sub>	a	b	SE(b)	$\mathbf{r}^2$	t-test
Spring	56	11.5	15.0	12.7	9.1	21.3	13.4	0.0077	2.7981	0.009745	0.9405	-20.7
Autumn	303	9.5	15.3	11.6	5.2	23.5	9.8	0.0049	2.9256	0.005431	0.9289	-13.6
Winter	209	10.5	14.8	11.9	8.3	20.8	11.7	0.0227	2.5182	0.008901	0.8460	-54.1
Total	567	9.5	15.3	12.1	5.2	20.8	3.0	0.0059	2.7930	0.005862	0.9376	-35.3

*Note:* SE is the standard error.

**Table 2.** Overall estimated LWR values of *E. encrasicoulus*.

		L	ength (cm	)	Weight (g)							
Seasons	Ν	$\mathbf{L}_{\min}$	L <sub>max</sub>	L <sub>mean</sub>	$\mathbf{W}_{\min}$	W <sub>max</sub>	$\mathbf{W}_{mean}$	a	b	SE(b)	$\mathbf{r}^2$	t-test
Spring	33	11.4	13.9	12.7	8.6	16.5	12.0	0.0040	3.1460	0.014423	0.9485	10.1
Autumn	31	9.2	11.4	10.2	3.6	8.0	4.7	0.0024	3.2584	0.055790	0.7196	4.6
Winter	148	9.7	13.8	11.3	4.1	15.5	8.1	0.0019	3.4370	0.008759	0.9142	49.8
Total	212	9.2	13.9	11.4	3.6	16.5	8.3	0.0019	3.4207	0.005935	0.8687	70.8

*Note:* SE is the standard error.

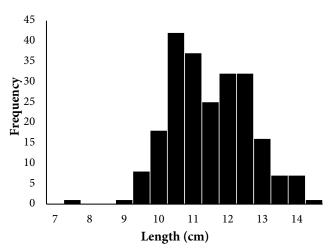
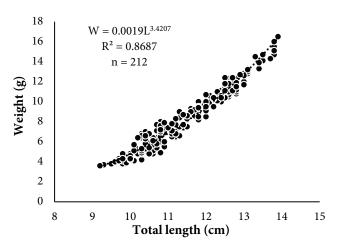
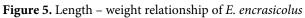


Figure 4. Length-frequency distribution of E. encrasicolus









According to length-frequency distribution, mean length of the *E. encrasicoulus* was found as 11.4 cm and vast majority (87%) accumulated between 10.5 and 14.0 cm (Fig. 4). Allometry coefficient of the seasonal LWR parameters estimated and have been found for every season (Table 2). In detail, *b* value of the *E. encrasicoulus* was found for months as 3.1460, 3.2584, 3.4370 and total as 3.4207, respectively (Table 2) and these values are indicating positive allometric growth (b=3.42; *t*-test,  $t>t_{0.05, n>200}=1.65$ ) (Fig. 5). Also, r<sup>2</sup> values of *E. encrasicoulus* shown a strong relationship between length and weight as 0.87.

#### Discussion

Overall results of *S. pilchardus* and *E. encrasicolus* showed dissimilarities in total length (TL) and mean length based on sampling sites (Table 3 and Table 4). So far, TL of *S. pilchardus* has been shown a great variety in the Aegean Sea. However, maximum total length value of *S. pilchardus* reported from Izmir Bay by Hoşsucu et al. (1994) as 17.0 cm and it is still maintaining validity. In Izmir Bay, prior records indicating that

**Table 3.** Comparative results of LWR parameters of S. pilchardus

the mean length of European pilchard has been reported as 14.2 cm by Hoşsucu et al. (1994), 11.82 cm by Özaydin and Taskavak (2006) and 9.39 cm by Acarli et al. (2014). In the results of this study, total length distribution of *S. pilchardus* between 9.5-15.3 cm. For the estimation of the mean length values of *E. encrasicolus* distribution range has been reported as 9.95 cm by Acarli et al. (2014) and 12.09 cm by Özaydin and Taskavak (2006). In this study, the mean length found as 11.4 cm and it has been shown similarity and also, it has been found as a medium value of these results.

Furthermore, the reported results of the b coefficient, which show different types of growth, such as isometric and allometric growth depending on different sampling areas are notable. As a short note, the b value is useful in explaining the body shape (growth type) according to the conditions in which the fish is present. If this value is equal to "3" it is called isometric but if it is a different value than "3", then it is called allometric growth (Ricker, 1975; Sparre et al., 1989; Sparre and Venema, 1992; Avsar, 2016).

Author	Location	Sex	n	а	b	r <sup>2</sup>	Growth
Present study	Aegean Sea- Izmir Bay	ďŶ	567	0.0059	2.793	0.94	- allometric
Petrakis and Stergiou, 1995	South Euboikos Gulf	ďŶ	82	0.00003	2.754	0.82	-allometric
Sinovčić et al., 2004	Adriatic Sea	ďŶ	4441	0.0038	3.230	0.98	+allometric
Mendes et al., 2004	Portuguese west coast	ďŶ	113	0.0017	2.772	0.77	- allometric
Tarkan et al., 2006	Marmara Region -Turkey	ďŶ	11	0.0021	3.540	0.98	+allometric
Pešić et al., 2006	Boka Kotorska Bay	ďŶ	2489	-0.0047	3.167	0.99	+allometric
Özaydin and Taskavak, 2006	Aegean Sea- Izmir Bay	ďŶ	388	0.0076	3.190	0.89	+allometric
Karachle et al., 2008	North Aegean Sea	ďŶ	752	0.0053	3.144	0.90	+allometric
Veiga et al., 2009	Southern Portugal	ďŶ	676	0.0051	3.140	0.95	+allometric
Mustac et al., 2010	Middle Adriatic Sea	ď	668	0.0425	2.371	0.58	- allometric
		Ŷ	541	0.0342	2.465	0.73	- allometric
Torres et al., 2012	Gulf of Cadiz	ďŶ	1656	0.0082	3.016	0.87	isometric
Acarli et al., 2014	Izmir Bay – Homa Lagoon	ď₽	77	0.0070	3.053	0.99	+allometric

**Table 4.** Comparative results of LWR parameters of *E. encrasicolus*

Author	Location	Sex	n	а	Ь	r <sup>2</sup>	Growth
Present study	Aegean Sea- Izmir Bay	ď₽	212	0.0019	3.421	0.87	+allometric
Sinovčić et al., 2004	Adriatic Sea	ď₽	4234	0.0039	3.160	0.99	+allometric
Özaydin and Taskavak, 2006	Aegean Sea- Izmir Bay	ď₽	513	0.0116	2.840	0.94	-allometric
Ismen et al., 2007	Saros Bay	ď₽	212	0.0050	2.970	0.87	-allometric
Karachle et al., 2008	North Aegean Sea	ď₽	759	0.0008	3.822	0.95	+allometric
Veiga et al., 2009	Southern Portugal	ď₽	278	0.0039	3.190	0.98	+allometric
Torres et al., 2012	Gulf of Cadiz	ď₽	2293	0.0049	3.125	0.97	+allometric
Acarli et al., 2014	Izmir Bay – Homa Lagoon	ďŶ	68	0.0070	2.917	0.99	-allometric





So far, many studies of S. pilchardus indicating allometric growth and only one study reported as isometric growth such as Torres et al. (2012). As it seems in Table 3, there were differences between allometric growth. So that, Mendes et al. (2004), Petrakis and Stergiou (1995) and this present study results has been shown negative allometric growth. Otherwise, the rest of them has been indicated positive allometric growth. Comparison of the reported values of *E. encrasicolus* shown that all researchers have been agreed on the allometric growth of this species. However, growth type of depending on *b* value have a variety among conducted studies. Such that, Sinovčić et al. (2004), Karachle et al. (2008), Veiga et al. (2009), Torres et al. (2012) and this present studies b value indicating positive allometric growth. On the contrary, other studies has been shown negative allometric growth (Table 4). Length-frequency distributions and b value is directly associated to the fishing gear and method. While gillnets/trammel nets are shown higher selectivity for sardine related to mesh size and mesh shape, selectivity of the purse seine bunt is so poor that even very small sizes of juveniles are not selected. For instance, Torres et al. (2012) reported an unusually isometric growth of S. pilchardus with bottom trawl and this shows us the importance of sampling method. Also, except for the method, there are so many contributing variables (feeding, reproduction and temperature of the habitat that fish population live, etc.) to the effect of change of b value. Izmir Bay is also known as an important spawning and nursery ground for several fish species, mainly because of lagoons which serve as sheltered habitats and the input of nutrients from the Gediz River (Özaydın and Taskavak, 2006). So that, sampling sites that fish caught is also an important variable to establish the b value, even in the Izmir Bay.

#### Conclusion

*S. pilchardus* and *E. encrasicolus* are highly demanding and invaluable fish species for human consumption as well as fish meal and oil industry in worldwide and also in Turkey. We believe that this study will contribute to understanding the changing of the populations of *S. pilchardus* and *E. encrasicolus* in Izmir Bay. A decrease of the mean total length of *S. pilchardus* has been considerable variable from 1994 to 2014 in Izmir Bay but with this study, it is observed that mean length of the sardine found near of 2006 value related to seasonal fishing pressure.

#### Acknowledgments

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#### **Conflict of Interest**

The authors declare that there is no conflict of interest.

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# Marine Science and Technology Bulletin

#### SHORT COMMUNICATION

### Additional record of *Hemiramphus far* (Forsskål, 1775) (Hemiramphidae) in Northern Aegean Sea (İzmir Bay, Turkey)

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On December 5, 2018, a specimen of the spotted halfbeak, Hemiramphus far has been caught by a purse-seiner off Gediz River delta, İzmir Bay at a depth of 40 m. This paper presents the first occurrence of H. far in İzmir Bay. At the same time, this short note presents the fourth record of H. far for the Turkish Aegean Sea, including Gökova and Güllük Bays, and Eski Foça.

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#### Introduction

Spotted halfbeak, Hemiramphus far (Forsskål, 1775) is an epipelagic, schooling fish that usually swims close to the sea surface in coastal waters. H. far has wide Indo-Pacific distribution and invaded the Mediterranean from the Red Sea via the Suez Canal and established in its new habitat from Rhodes to Egypt (Collette and Parin, 1986; Golani et al., 2006).

In the Mediterranean Sea, H. far (as H. marginatus) has been first recorded in Palestinian waters (Steinitz, 1927). It has been widespread off Israel and Lebanese waters since 1980s and reached to the coast of Albania (Collette and Parin, 1986), the Libyan coasts (Shakman and Kinzelbach, 2006), Gulf of Tunis



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(Rafrafi-Nouira et al., 2012), eastern Algerian coast (Kara et al., 2012) and Lampedusa Island, Strait of Sicily (Falautano et al., 2014).

In Turkish seas, *H. far* has been recorded first from the Turkish coasts of Mediterranean (Kosswig, 1950). The samples of *H. far* have been documented from Mersin Bay in 1980s (Gücü et al., 1994), then, from Iskenderun and Gökova Bays, Aegean Sea (Torcu and Mater, 2000), from the coasts of Karataş, Iskenderun Bay (Başusta and Erdem, 2000). In the Aegean Sea, Geldiay (1969) mentioned the *H. far* (as *H. marginatus*) in the Aegean Sea only by name. Other successive records were given from the north-western Rhodes Island, Greece (Papaconstantinou, 1990), Gökova Bay (Torcu and Mater, 2000), from Eski Foça (Akça and Bilecenoğlu, 2010) and Güllük Bay (Akyol and Ertosluk, 2019).

This paper presents the first occurrence of *H. far* in İzmir Bay, and at the same time, it is added as a lessepsian fish record going towards to northern latitude of the Aegean Sea.

#### Material and Methods

On December 5, 2018, one specimen of *Hemiramphus far* (Figure 1), was caught by a purse-seiner off Gediz River delta, İzmir Bay (38°34'240 N 26°46'533 E) at a depth of 40 m (Figure 2). The sample, fixed in 6% formaldehyde solution, has been preserved in the fish collection of the Fisheries Faculty, Ege University (ESFM-PIS/2018-10).



**Figure 1.** *Hemiramphus far*, caught from İzmir Bay (Photo: O. Akyol)

**Table 1.** Morphometric measurements, ratios and meristic counts of *Hemiramphus far*, captured from İzmir Bay, northern Aegean Sea and previous records from the Mediterranean [<sup>1</sup>This study; <sup>2</sup>Rafrafi-Nouira et al. (2012); <sup>3</sup>Kara et al. (2012); <sup>4</sup>Falautano et al. (2014); <sup>5</sup>Akyol and Ertosluk (2019)]

Locality	İ	zmir Bay <sup>1</sup>	Tunisia <sup>2</sup>	Algeria <sup>3</sup>	Lampedusa <sup>4</sup>	Güllük Bay⁵
Locality		n=1	n=1	n=2	<b>n</b> =7	n=1
Measurements	Size (mm)	<b>Proportion</b> (TL%)	Size (mm)	Size (mm)	Size (mm)	Size (mm)
Total length (TL)	210		365	259-290	175-252	282
Fork length (FL)	189	90.0	329	222-249	139-205	250
Standard length (SL)	180	85.7	315	213-234	131-197	240
Pectoral fin length	25	11.9	11.4	45-57	23-39.1	32
Pre-dorsal fin length	106	50.5	254	168-181	100-156	145
Pre-anal fin length	113	53.8	268	173-195	104-158	150
Lower jaw length	46	21.9	74.1	70-75	42-60	59
Upper jaw length	5	2.4	12.2	9-10	3.6-6.4	8
Maximum body depth	18	8.6	28.4	-	19-31	20
Body width	12	5.7	-	-	-	20
Head length	31	14.8	118.7	45-56	29.8-43	41
Eye diameter	7	3.3	13.5	12-13	7.8-10.8	11
Interorbital distance	8	3.8	15.2	13-14	7.4-12	-
Meristic counts						
Dorsal fin rays		11	12	_	12-13	11
Anal fin rays		10	11	-	10-12	10
Pectoral fin rays		11	13	-	12	12
Ventral fin rays		6	6	-	6	6
Weight (g)		21	143.2	-	16-84	61.4



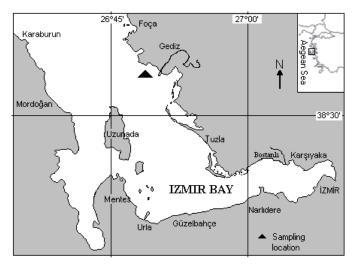


Figure 2. Sampling location of Hemiramphus far

#### **Results and Discussion**

Morphological description, colour, morphometric measurements with percentages of total length (TL%) and meristic counts (Table 1) are in agreement with Collette and Parin (1986), Golani et al. (2006) and Akyol and Ertosluk (2019). In addition, some previous records throughout the Mediterranean were shown in Table 1, and TL and weight of *H*. *far* in the Mediterranean were between 175 and 365 mm, and 16 and 143.2 g, respectively. The largest specimen with 365 mm TL was recorded off Ras Jebel, Tunisia (Rafrafi-Nouira et al., 2012).

#### Conclusion

At northernmost, *H. far* has been recorded off Eski Foça by Akça and Bilecenoğlu (2010), and further individuals were observed in the same area. So, the occurrence of *H. far* in the coasts of İzmir Bay is not unexpected due to the previous records of the Aegean Sea. This record signs that *H. far* has started to settlement in northern Aegean Sea widely.

#### **Conflict of Interest**

The authors declare that there is no conflict of interest.

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# Marine Science and Technology Bulletin

#### **RESEARCH ARTICLE**

# The presence of bristlemouth, *Gonostoma denudatum* (Rafinesque 1810), from the coast of Northern Cyprus (Northeastern Mediterranean)

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#### ABSTRACT

*Gonostoma denudatum* belonging to Gonostomatidae family are small fishes living in deep-sea around Atlantic and Mediterranean. They are vital species for plastic accumulation because they can be available in all depths of the water column during the day. In this study, it was aimed to present the recent record of *G. denudatum*, which captured from North Cyprus in May 2018. The total length of the specimen, which obtained from a depth of between 420 and 640 m, is 12.8 cm. Its photograph was taken and the catalogue number (MEUFC-19-11-108) was given. Morphometric characteristics were measured and calculated. The specimen is stored in the Museum of the Systematic, Faculty of Fisheries, Mersin University.

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#### Introduction

*Gonostoma denudatum* is a species belonging to the Gonostomatidae family of order Stomiiformes. They are bathypelagic species and live at a depth of 100-700 m (Badcock, 1984). They are found in East and West Atlantic waters

(Schaefer et al., 1986). In a study conducted in Iskenderun Bay in 2015, a sample of *G. denudatum* with a standard length of 11.8 cm was reported from a depth of 200 m (Bilecenoğlu et al., 2014). A record of *G. atlanticum* belonging to same genus with *G. denudatum* reported from Cyprus in 2015 (Çoker and Cihangir, 2015). Maximum total length for *G. denudatum* was

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reported as 14 cm (Quéro, 1990). Recently, this species has reported from Mersin Bay as 13.7 cm, TL (Bayhan and Erguden, 2019).

*G. denudatum* migrate vertically in the water column. They are available at depths of 400-700 m during the daytime and 100-200 m during the nighttime (Badcock, 1984). They are the prey of organisms living at different depths because they migrate vertically throughout the day. Some predators of *G. denudatum* are *Beryx splendens* (Dürr and González, 2002), *Chauliodus sloani* (Battaglia et al., 2018), *Etmopterus spinax* (Bengil et al., 2019), *Mesoplodon bidens* (Pereira et al., 2011) and *Todarodes sagittatus* (Rosas-Luis et al., 2014).

All *Gonostoma* species have a pigment spot on the back of the eye. To distinguish those species, natural pigmentations in different parts of their bodies investigated. *G. denudatum* has a deep pigmentation from the caudal fin base to the dorsalcaudal peduncle and the lower caudal-fin base. (Ahlstrom et al., 1984).

*G. denudatum* is mostly feeding on Euphausiids and copepods (Badcock, 1984). In a study on the presence of microplastics in fish species living in the mesopelagic area in the Northwest Atlantic region, *G. denudatum* was found to be the fish having the most microplastic in the body (Wieczorek et al., 2018). With this study, an individual of *G. denudatum* caught off the coast of Northern Cyprus is reported. Besides, some morphometric of the fish is given.

#### Material and Methods

One specimen belonging to *G. denudatum* was caught from North Cyprus offshore waters. Sampling was carried out by a commercial trawl on May 17, 2018. The sampling depth ranged between 420 and 640 m. Sampling gear was prepared according to MEDITS procedures (Bertrand, 2002). The coordinates of the sampling area were 36.07227 N and 34.53326 E (Figure 1).

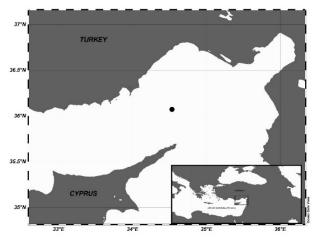


Figure 1. Sampling location of Gonostoma denudatum

Its photograph was taken and catalog number (MEUFC-19-11-108) was given. Species identification fulfilled according to information provided in Ahlstrom et al. (1984). Morphometric measurements were performed according to Bilecenoğlu et al. (2014) and compared with previous studies in the Eastern Mediterranean (Table 1). The specimen was preserved in 4% formaldehyde and deposited in the Museum of the Systematic, Faculty of Fisheries, Mersin University.

#### Results

The total length of *G. denudatum* (Figure 2), an Atlantic deep water species caught off the coast of Cyprus, was measured as 12.8 cm. The maximum total length reported in the literature is 14 cm (Quéro, 1990). In this case, the individual caught in the waters of Cyprus was mature.

Table 1. Comparison of the morphometric measurements of G. denudatum with previous studies

	Preser	nt study	Bayhan and <b>F</b>	Ergüden (2019)	Bilecenoğlu et al. (2014)		
Number of fish		1		1	1		
Measurements	Size (mm)	Values (%)	Size (mm)	Values (%)	Size (mm)	Values (%)	
Total length (TL)	128		130.7		N/A		
Standard length (SL)	122		117.5		118		
Head length (HL)	32	26.2	30.3	25.7	29	24.6	
Eye diameter	4.9	4	4.7	15.5	5	17.2	
Inter orbital distance	4.4	3.6	4.3	14.1	4.5	15.5	
Post orbital length	21	17.2	20.9	68.9	17	58.6	
Snout length	7.2	5.9	6.3	20.8	6.9	24.1	
Pre-dorsal length	77.5	63.5	72	61.2	70.4	59.7	
Pre-anal length	76	62.3	69.9	59.5	69.5	58.9	
Pre-pectoral length	30	24.6	28.9	24.6	N/A	N/A	
Pre-pelvic length	57.3	47.0	55	46.8	N/A	N/A	

Note: N/A means Not Available



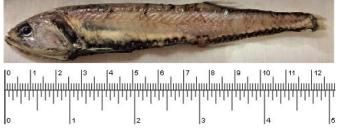


Figure 2. The specimen of the *G. denudatum* 

#### Discussion

The fish caught in this study is a recent report for the Eastern Mediterranean and the first record for Cyprus. It has also contributed to the list of marine fish in Cyprus. In Table 1, morphometric features of G. denudatum compared with previous studies. Total length of the specimen was measured as 12.8 cm. However, caudal fin of the specimen was damaged during the fishing operation (Figure 2). In this way, the actual total length of the sample could probably longer than 12.8 cm. When the standard length of the sample compared with other studies, it is seen that this specimen is the biggest individual caught from the Eastern Mediterranean Sea. The most of percentage values of the G. denudatum in all compared studies are similar except; eye diameter, interorbital distance, postorbital length, and snout length. It is thought that this difference caused by other studies is due to a mistake made during the calculation. When the given lengths are converted to a percentage, it gives different results. Meristic characteristics couldn't be measured and compared because the fin rays were damaged.

*G. denudatum* listed as "Least Concern" in the International Union for Conservation of Nature (IUCN) Red List, and the population trend is unknown (Harold, 2015). Together with the recent record of *G. denudatum* from Mersin Bay (Bayhan and Erguden, 2019), and the present study, it can be said that this fish started to make a population in the Eastern Mediterranean region. Also, this study is the first record for Cyprus.

On the other hand, *G. denudatum* is a fish that can found at different depths during the day, and it hunted by various fish species (Badcock, 1984). Some of these predator species have commercial value for human consumption. According to Wieczorek et al. (2018), *G. denudatum* was found to be the most micro-plastic accumulating species in its body. Together with this information, it can be said that *G. denudatum* plays a crucial role in transporting plastic pollution back to humans via the food chain. A further study on this species about its plastic accumulation and transferring them via food chain is planned.

#### Conclusion

*G. denudatum* lives in the deep sea, and it's rarely seen in the Mediterranean region because of regular fishing activities not aiming at those depths. Now there are two recent records for *G. denudatum* in the adjacent areas means that those fishes started to increase their population. New researches can be done for those fishes about their plastic accumulation properties. They can be unique and powerful indicators for plastic pollution studies in the future.

#### **Conflict of Interest**

The authors declare that there is no conflict of interest.

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## Marine Science and Technology Bulletin

#### **RESEARCH ARTICLE**

### Diet composition of bluefish *Pomatomus saltatrix* (Linnaeus, 1766) in the Sea of Marmara

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#### ABSTRACT

In this study, diet composition of bluefish *Pomatomus saltatrix* (Linnaeus, 1766) was investigated. A total of 512 bluefish samples were monthly collected from commercial fishing boats operating in the Sea of Marmara between January and December 2014. It was determined that the total length distribution of the samples varied between 12.3-47.3 cm. 367 of them (71.67%) were found to be the full of the stomach. The nutritional composition of stomach contents only two main prey groups (teleostei and crustacean) were identified. In evaluation, relative importance indexes (IRI) food groups were calculated. According to the relative importance index (IRI=91.8%) anchovy (*Engraulis encrasicolus*) has been found to be the most preferred food group. The number of individuals whose stomachs were found to be full was low in the winter months; it started to rise with spring and reached the highest level in the autumn months. It was determined that the increase in the total number of stomachs occurred between August and October in relation to the reproductive period. It was found that stomach fullness rates significantly relationship between sex and seasons (p<0.01).

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#### Introduction

Bluefish, *Pomatomus saltatrix* (Linnaeus, 1766), is a pelagic, migratory and cosmopolitan species which inhabits warm and

temperate waters of the Atlantic, Indian, Pacific Oceans, Mediterranean and Black Seas (Slastenenko, 1956; Briggs, 1960; Wilk, 1977; Tortonese, 1975). Bluefish, at the end of spring migrate to the Black Sea for feeding and spawning and stay



along the summer. In early autumn they start to migrate back to the Marmara Sea and Aegean Sea (Ceyhan et al., 2007). Bluefish is a widely exploited and economically important species in coasts of Turkey.

For a good fisheries management, reproduction, nutrition and growth of the species should be well known. Studies on the analysis of fish and stomach contents play a key role in understanding fish biology, ecology, physiology and behavior (Arias, 1980). Stomach content analyzes, describe the nutritional habits of individuals belonging to the population as well as nutritional competition among species (Lawror, 1980). In this way, the role of species in the food chain can be determined, contributing to fisheries modeling and fisheries planning (Hyslop, 1980). In addition, stomach contents gives information about the environment and diet composition of fish species (Wootton, 1990; Buckel et al., 2004).

There are very few studies on the stomach content and feeding regime of bluefish in the seas of Turkey (Türgan, 1959; Artüz, 2003).

In the present study, the effect of season and sex groups on the feeding habits of blue fish *Pomatomus saltatrix* was investigated. The result of the study can be a baseline data for fisheries biologists and also contributes scientifically to the sustainability of regional fisheries.

#### Material and Methods

A total of 512 bluefish samples were monthly collected from commercial fishing boats operating in the Sea of Marmara between January and December 2014 (approximately coordinate of sampling area: 40° 34' 16.6" N-27° 30' 01.3" E; 40° 31' 52.4" N 27° 30' 03.9" E).

Samples were preserved in iceboxes for examination in the laboratory. Specimens were measured to the nearest 0.1 cm for total length (TL). The abdominal region was opened for gonads and stomach contents examination. Stomach contents and gonads were examined by macroscopic observation. In evaluation, relative importance indexes (IRI) were calculated (Frost, 1946; Pinkas, 1971; Windell and Bowen, 1978; Hyslop, 1980). Samples whose stomach contents were completely digested were excluded from the evaluation. To determine the differences in gastric occupancy rate between sexes, seasons and length analysis of variance test was used.

#### Results

The nutritional composition of bluefish only two main prey groups (teleostei and crustacean) was determined. Samples whose stomach contents were completely digested were excluded from the evaluation. While a food organism was found in the stomach of 367 of the examined samples, it was observed that a total of 145 stomachs, 67 female and 78 male samples were completely empty. 73.30% of females (n=184), 70.11% of males (n=183) and 71.67% of all individuals were found to have full stomach. Nutritional concentrations of bluefish have been found to reach their maximum levels in autumn months when they begin at the end of summer (Figure 1).

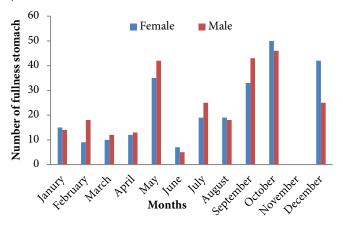


Figure 1. Monthly stomach fullness and distribution of samples

It was determined that the increase in the number of full stomachs occurred between August and October in relation to the reproductive period. As the stomach contents of the samples could not be examined in November, they were not included in the graph. With the Analysis of variance test, it was found that the difference between sex, seasons and length according to stomach fullness rates was important (P<0.01). Stomach fullness status, sex, maximum length, minimum length, mean length values and standard deviation of the samples examined monthly were given in Table 1.

Stomach contents were shown in Figure 2. The majority of the prey groups identified were bony fishes (Osteichthyes), while a small number of them were found to be crustaceans (crab and shrimp). 63.81% of the bony fish anchovy (*Engraulis encrasicolus*), 14.52% horse mackerel (*Trachurus mediterraneus*), 8.83% whiting (*Merlangius merlangus*), 4.84% red mullet (*Mullus barbatus*), prey groups included in the crustacean class consisted of 4.30% of stomach contents. In addition to these results, 3.70% bluefish was detected in the stomach content of bluefish. With this result, cannibalism has been determined in population of study area (Figure 3).

According to the relative importance index (IRI=91.8%) anchovy (*Engraulis encrasicolus*) has been found to be the most preferred food group. The importance indexes of other food groups are as follows, *Trachurus mediterraneus* IRI=5.0%, *Merlangius merlangus* IRI=1.8%, *Mullus barbatus* IRI=0.5%, *Pomatomus saltatrix* IRI=0.3% and crustacean (crab and shrimp) IRI=0.6%.



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			ł	Female ( <b>♀</b> )		Male (♂)				
Months				Total Le				Total Length (cm)		
	Ν	NFS	NES	Min-Max	in-Max Mean±SD		NFS	NES	Min-Max	Mean±SD
January	15	10	5	12.3-31.0	20.7±0.75	14	7	7	22.0-33.0	14.4±1.84
February	9	5	4	20.0-32.0	24.3±0.91	18	9	9	18.5-32.0	23.3±0.73
March	10	1	9	15.4-19.4	17.2±1.32	12	3	9	15.8-19.0	17.1±1.05
April	12	5	7	22.0-33.0	24.5±0.40	13	3	10	22.0-28.0	24.1±1.71
May	35	19	16	15.0-36.7	25.2±0.53	42	23	19	15.0-47.3	21.8±0.56
June	7	7	0	21.6-29.0	23.0±2.59	5	5	0	22.3-23.1	22.6±0.29
July	19	9	10	22.0-23.9	22.6±0.56	25	14	11	19.9-23.4	22,1±0.81
August	19	14	5	24.3-32.0	26.8±2.66	18	15	3	22.3-32.3	26.1±3.09
September	33	23	10	14.6-33.4	20.5±0.38	43	33	10	14.9-34.0	21.9±0.26
October	50	50	0	13.1-28.7	16.4±1.41	46	46	0	12.7-31.7	16.6±0.12
November	-	-	-	-	-	-	-	-	-	-
December	42	41	1	17.4-23.6	19.9±1.52	25	25	0	18.0-22.7	20.1±1.44
Total	251	184	67	12.3-36.7	21.9±1.15	261	183	78	12.7-47.3	20.9±1.08

Table 1. Monthly stomach fullness of sex groups

Note: 'N: sample size; NFS: number of fullness stomach; NES: number of empty stomach; Min: Minimum; Max: Maximum; SD: Standard Deviation



Figure 2. Stomach contents of the *P. saltatrix* in the Sea of Marmara

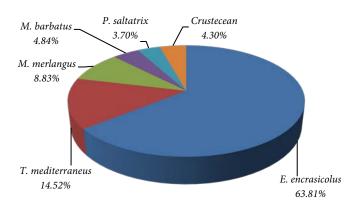


Figure 3. Distribution of prey groups in stomach contents (%)

#### Discussion

Although there are very few studies on determining diet of bluefish in the seas of Turkey, there are many research results related to the subject in different parts of the world (Buckel et al., 1999; Grant, 1962; Lassiter, 1962; Marks, 1993; Creaser and Perkins, 1994).

In a study carried out by Türgan (1959) it was reported that the bluefish migrated between the Black Sea and the Marmara Sea and they feed mainly on fish. In a different study, gastric contents of bluefish caught in the Bosphorus were examined and found to be feed on *Engraulis encrasicolus*, *Trachurus mediterraneus*, *Belone belone*, *Scomber scombrus*, *Scomber japonicus*, *and Sarda sarda* species (Artüz, 2003). In addition, the presence of bluefish, representing 3.70% of the food groups, shows that there is cannibalism (Bade, 1977). These results support the findings of the present study.

As a result of a similar study carried out in the shallow waters of estuaries on the Eastern coast of America, stomach contents of juvenile and adult bluefish were examined and it was found that anchovy was the dominant species (Buckel et al., 1999). Lassiter (1962) reported that nutrient ratios of invertebrates decrease with increasing length of predators.

In a different study, it was reported that the majority of gastric contents of young bluefish (10-20%) were invertebrates, whereas adult individuals were fed on fish and anchovy was preferred (Buckel et al., 1999). In a study conducted in the



48



estuaries in India, 40.5% of the stomach contents were reported to be small sea creatures, 15.8% herring, 13.9% silver fish and 8% anchovy (Grant, 1962).

However, there are also different results in the literature. Creaser and Perkins (1994) investigated the stomach contents of juvenile and adult bluefish in the Marsh River in Maine, USA and reported that the average of 0.7% terrestrial plants and 0.3% insect (Hymenoptera) group was found in the stomachs of the examined individuals. In another study, stomach contents of the juvenile bluefish in two different periods (spring and summer season) were examined by Marks (1993). Author reported that approximately 89% of the stomach contents found to be full and copepods were dominant.

It is determined that bluefish are generally fed on fish but depending on environmental conditions, in some periods they are fed on invertebrates. In addition, another important issue has been identified with cannibalism in the species with this study. It is estimated that the cause of cannibalism in the species is due to lack of nutrients depending on environmental conditions.

#### Conclusion

Analysis of fish diet, play a key role in understanding fish biology, ecology, physiology and behavior. Bluefish economically is one of the important species in coasts of Turkey. In this study, the role of bluefish in the food chain has been determined and results, may contribute to fisheries and fish biologists.

#### **Conflict of Interest**

The authors declare that there is no conflict of interest.

#### Acknowledgements

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# Advice Science and Technology Bulletin

#### **RESEARCH ARTICLE**

# Investigation of active tectonics of Edremit Gulf, Western Anatolia (Turkey), using high-resolution multi-channel marine seismic data

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ARTICLE INFO	ABSTRACT
Article History:	The Edremit Gulf is situated on the upper Miocene transtensional basin in the Western
Received: 21.10.2019 Received in revised form: 17.02.2020 Accepted: 17.02.2020	Anatolia and formed by the interaction between the North Anatolian Fault (NAF) and the N-S extensional tectonic regime of the Aegean domain. Our study is aimed to investigate the structural effects of these tectonic forces in the Gulf. Thus, approximately 300km.
Available online: 21.02.2020	seismic data were collected within the Gulf area using the high-resolution seismic reflection method. The results indicated that the interpretation of the data, an E-W oriented, strike-
Keywords:	slip fault system (Edremit Bay Fault - EBF) was identified in the Gulf as a possible
Marine tectonics	continuation of the Havran - Balıkesir Fault Zone which can be followed on land. Likewise,
Marine seismic	a second strike-slip fault system (Edremit - Lesbos Fault; ELF) was observed which crosses
Western Anatolia	the Gulf towards Lesbos Island in the NE-SW direction. This system was interpreted as the
Edremit Gulf	possible continuation of the Yenice - Gönen Fault Zone which is thought to be the branch
Strike-slip tectonics	of the North Anatolian Fault.

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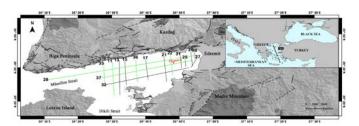
#### Introduction

Edremit Gulf is a basin, located in the eastern Mediterranean, Aegean Sea, between the Biga Peninsula at the north, the Lesbos Island at the west and the Madra Mountains at the south. It is connected to the Aegean Sea by Müsellim Strait at the west and Dikili Strait (or Lesbos Strait) at the South (Figure 1). It has been shaped by both westward progression and N-S oriented extension of the Anatolian Plate (Dewey and Şengör, 1979; Barka and Reilinger, 1997; Yılmaz, 1997; Armijo, Meyer, Hubert and Barka, 1999; Yılmaz et al., 2000; Westaway, 2003) (Figure 2).

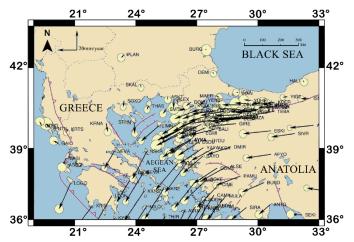


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A counter-clockwise rotation of the Aegean Region has proven by numerous studies, especially by GPS measurements (Le Pichon, Chamot-Rooke, Lallemant, Noomen and Veis, 1995; Oral et al., 1995; Yılmaz et al., 2000; Boztepe Güney et al., 2001) (Figure 2).



**Figure 1.** Location map of the research area and survey lines (green ones are presented in the paper) with Edremit-1 borehole location, compiled from (Boztepe Güney et al., 2001; Kurtuluş, Doğan, Sertçelik, Canbay and Küçük, 2009; Gürer et al., 2016).



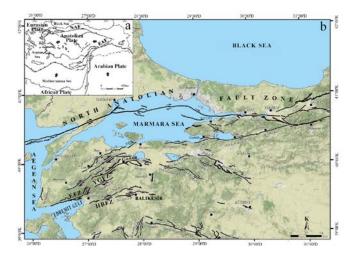
**Figure 2.** Western Anatolian speed vectors (Le Pichon et al., 1995; Oral et al., 1995; McClusky et al., 2000; Yılmaz et al., 2000; Boztepe Güney et al., 2001; Tur et al., 2015)

Besides, Paleomagnetic studies also prove that the Edremit Gulf region was affected by a counterclockwise rotation during the Pliocene-Quaternary times (İşseven et al., 1995; Orbay et al., 1999; Sözbilir, et al., 2016a). The gulf is affected by NE-SW trending fault zones such as Yenice – Gönen Fault Zone (YGFZ), Edremit Fault Zone (EFZ) and Havran – Balıkesir Fault Zone (HBFZ). The seismotectonic analysis shows that most of the faults of Edremit Gulf and surroundings are right lateral and strike-slip faults (Sözbilir et al., 2016b) (Figure 3b).

Paleostress studies done in the study area show that there is a dominant NE-SW opening regime that dominates the region. This model shows the main effects of the North Anatolian Fault System and the Aegean Region Extension System on the region (Gürer et al., 2016) (Figure 3b).

Despite many types of research in the region, most of all are focused on land and marine neotectonics studies are quite small

in number. While Kurtuluş et al. (2009) evaluated 21 deep seismic profiles in the inner and middle parts of the Gulf of Edremit by 2009, Çiftçi, Temel and Terzioğlu (2004) demonstrated the Neogene stratigraphy in and around the gulf. The aim of this article is to contribute to such marine studies and to connect both the land and marine tectonic structures to better understand the regional tectonism.



**Figure 3.** Tectonic map of Anatolian Plate (EAF; East Anatolian Fault, NAF; North Anatolian Fault) (**a**) and North Western Anatolia (**b**) compiled from Kaymakçı, 2006; Özkaymak, 2015; Sözbilir et al., 2016a.

#### **Regional Geology**

Biga Peninsula consists of Paleozoic and Mesozoic metamorphic, ophiolitic and early Cenozoic plutonic rocks as the basement and late Cenozoic sedimentary and volcanic rocks lying on the basement. At the southern margin of the Biga Peninsula, there is a rise of the Kazdag Massif between Edremit Gulf and Yenice – Bayramic Basin with a lithology of marbles, amphibolites and Paleozoic-Triassic gneiss (Gürer et al., 2016) (Figure 4).

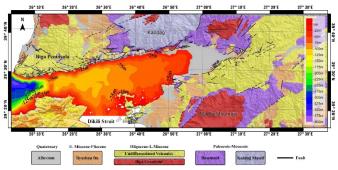
Magmatic rocks are quite common in the Biga Peninsula. They may be identified as Middle Eocene and Oligo-Miocene plutonic and volcanic rocks. The latest magmatic phase in the region is represented by the Late Miocene - Quaternary alkaline rocks (Genç, 1998; Yılmaz and Karacık, 2001; Beccaletto and Steiner, 2005; Gürer et al., 2016).

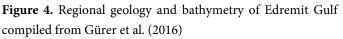
The sedimentary cover in the region is represented by Neogene-Quaternary units. The largest sedimentary rock formations in the southern part of the Biga Peninsula are the Lower-Middle Miocene Küçükkuyu, the Upper Miocene İlyasbaşı, and the Plio-Quaternary Bayramiç formations (Sengun et al., 2011).

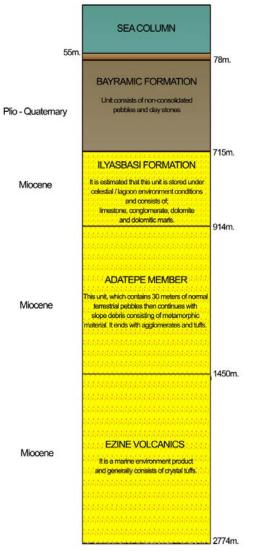
Based on ~2800 m of drilling data shown in Figure 5 made by Turkish Petroleum Corporation (TPAO) in Edremit Gulf in



1998; approx. 23 m of unconsolidated sediment, 637 m of Plio quaternary, Bayramic formation (pebble stone and limestone), 200 m of Miocene Ilyasbasi formation (limestone, pebblestone, dolomite, and marl), 536 m of Miocene Küçükkuyu formation, Adatepe member (agglomerate and tuff) and 1324 m of Miocene Doyuran Volcanites lithology could have been collected.







**Figure 5.** Drilling information chart of the TPAO Edremit-1 drill. Kılıç, O. (2018, October 12) Personal interview.

According to Çiftçi et al. (2004), plutonic and metamorphic rocks form the basement of the region. The Küçükkuyu Formation, which consists of Neogene sedimentary and volcanic units lie on the basement while Upper Miocene-Pliocene sediments of the fifth and sixth volcanism lie above the Küçükkuyu Formation with an angular unconformity, which is named as Mutlu or İlyasbaşı Formation by Siyako, Burkan and Okay (1989). The uppermost unit is considered as unconsolidated sediments.

#### Material and Methods

This study has been carried out in the inner and middle parts of the Edremit Gulf by using high-resolution seismic reflection method. Nearly 300 km of 2D multi-channel seismic data were collected using a 45+45 inch<sup>3</sup> GI gun by K. Piri Reis Research Vessel on 3 seismic lines along the NE-SW direction and 12 seismic lines in transverse N-S direction to define the inner gulf (Figure 1). Data were recorded by using a 192 channels streamer with a receiver group interval and shot interval of 6.25 m and 18.75 m, respectively. These parameters have provided 32-fold common-depth-point (CDP) data. Sampling interval and record length were selected as 1ms and 3000 ms, respectively.

#### Results

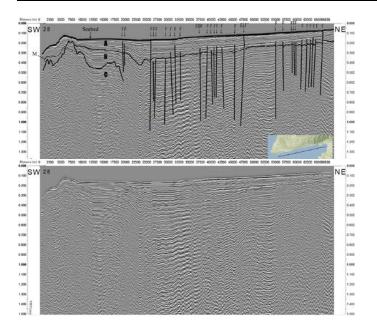
Since the sedimentary structure exhibits uniform stratification of reflectors close to each other, the sedimentary packages couldn't be separated. In this study, the boundaries of the strata, which could be followed, and show a slight impedance difference according to their surroundings have been determined and indicated with the letters A, B, and C in the sections.

A, B and C are seismic stratigraphic units that can be separated from each other by showing different impedance characteristics. Thin stratification in the geological structure of the seismic units A and B creates repetitive multiples which make stratigraphic interpretation difficult by obscures the actual signals.

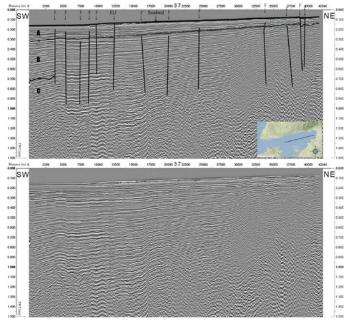
Besides, with the undulations at the SW of the section formed by the E-W compression, some strike-slip faults reaching up to the seabed and the Edremit – Lesvos Fault (ELF) are also being observed. The Edremit Bay Fault (EBF) which is located in the central part of the section ends in Holocene sediments and does not give any surface fracture.

In Section 37, a normal fault at the northeast, and towards the SW, the ELF with some faults which end in sediments close to the seabed, are observed.



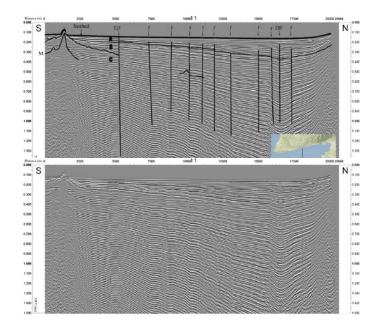


**Figure 6.** SW-NE directed seismic section 28. A, B and C; seismic units, F; fault, M; seabed multiples, ELF; Edremit – Lesvos Fault, EBF; Edremit Bay Fault

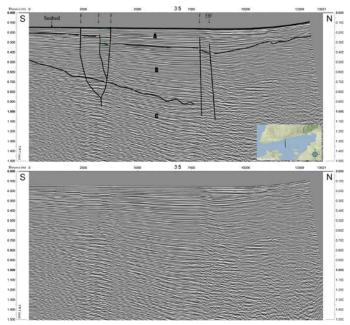


**Figure 7**. SW-NE directed seismic section 37. A, B and C; seismic units, F; fault, M; seabed multiples, ELF; Edremit – Lesvos Fault, EBF; Edremit Bay Fault

In some sections (11, 35, 36) (Figures 8, 9, 10) crossing the Edremit Gulf in the N-S direction, the seismic A, B, and C units are thickened towards the middle of the gulf. The acoustic basement forming the C unit approaches to the seabed in the sections towards the North and South shores of the Gulf. EBF and ELF systems also can be observed in these sections.



**Figure 8.** S-N directed seismic section 11. A, B and C; seismic units, F; fault, M; seabed multiples, ELF; Edremit – Lesvos Fault, EBF; Edremit Bay Fault



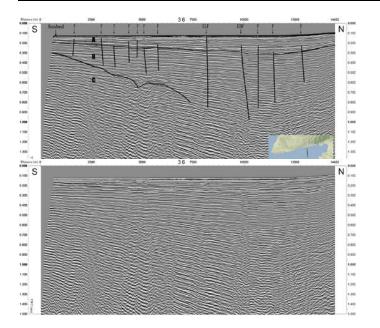
**Figure 9.** S-N directed seismic section 35. A, B and C; seismic units, F; fault, M; seabed multiples, EBF; Edremit Bay Fault

#### Discussion

The Edremit Gulf began to open under the control of lowangle NW–SE trending faults that developed after the compression of western Anatolia in an E–W direction in the early Neogene. Subsequently, regional N–S extensional stress formed the Aegean type basin system from the Neogene to Holocene (Kurtuluş et al., 2009).



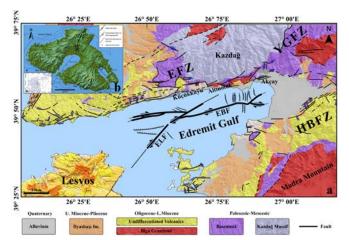




**Figure 10.** S-N directed seismic section 36. A, B and C; seismic units, F; fault, M; seabed multiples, ELF; Edremit – Lesvos Fault, EBF; Edremit Bay Fault

Although there are many opinions about the formation mechanism of the stress regime in Western Anatolia, the most accepted view is the collision of the African and Arabian Plates of different velocities with the Anatolian Microplate and forcing it to escape to the west by using the two important transform faults; the left-lateral East Anatolian Fault (EAF) and the right-lateral North Anatolian Fault (NAF) (Dewey and Şengör, 1979; Mantovani et al., 2000). The North Anatolian Fault System (NAFS) is exposed to the SW-NE rotation and is divided into three main branches as a result of the blockage of the Greek Plate in the east of the Marmara Sea (Jackson and McKenzie, 1988; Barka and Reilinger, 1997; Yaltırak, Alpar and Yüce, 1998; Yaltırak, 2002; Reilinger et al., 2006). The southernmost branch is re-divided into branches on the Biga Peninsula and continues as a zone. One of these branches, the Edremit Fault, formes the northern boundary fault of the Edremit Gulf (Yılmaz et al., 2000; Kurtuluş et al., 2009; Sözbilir et al., 2016a), while the other branch formes the Yenice-Gönen Fault Zone (Barka and Kadinsky-Cade, 1988). The study conducted by Yılmaz and Karacık (2001) propose that the southern strand of the NAFZ deviates toward the SW at the town of Gönen, continues on the same trend of YGFZ and reaches Edremit Gulf near Altınoluk.

Our data reveal that the YFGZ observed on land enters to the sea between Küçükkuyu and Akçay, and extends in Edremit Bay in segments, towards the Lesvos Island, compliance with the geology of Lesvos proposed by Lekkas et al. (2017) and the morphotectonic map of Lesvos Island proposed by Chatzipetros et al. (2013) (Figure 11b). We also infer that the HBFZ, which is described as a Holocene fault zone by Sözbilir et al., 2007 and consists of many strike-slip segments, extends from Balıkesir to the eastern end of the Gulf. The system continues in two segments to the west of the study area and shared by the ELF whilst forming a step over in the middle of the Gulf (Figure 11a).



**Figure 11.** Interpreted ELF and EBF on the base map compiled from Gürer et al. (2016) (**a**), The ELF shows compliance with the morphotectonism of Lesvos Island (**b**) (Chatzipetros et al., 2013). (HBFZ; Havran – Balikesir Fault Zone, YGFZ; Yenice – Gonen Fault Zone, EFZ; Edremit Fault Zone).

#### Conclusion

The interpretations of seismic reflection profiles indicates both the continuation of the southern strand of the NAF, the Yenice-Gönen Fault, within the Gulf, towards Lesvos Island, in the NE-SW direction, and the effect of the E-W oriented Balıkesir-Havran Fault, which formed during the N-S extension system. Due to the fact of the existence of these faults, we also infer a seismic hazard in the Gulf and surroundings.

#### **Conflict of Interest**

The authors declare that there is no conflict of interest.

#### Acknowledgements

We would like to express our gratitude and thanks to Prof. Dr. Derman Dondurur and Oğuz Kılıç for their great contribution and the time they have spent to the realization of this study, and to the Directorate of Dokuz Eylul University Institute of Marine Sciences and Technology, for their valuable support about the allocation of the research vessel. We would like to sincerely thank the reviewers for their insightful comments on our paper that have greatly improved the manuscript.



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# Marine Science and Technology Bulletin

#### **RESEARCH ARTICLE**

A study on maximum length record of saddled seabream (*Oblada melanura* Linnaeus, 1758) caught off Gökçeada Island (Northern Aegean Sea, Turkey)

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ARTICLE INFO	ABSTRACT
Article History:	A single specimen of <i>Oblada melanura</i> with 29.1 cm in total length and 390.00 g in total weigh was obtained off Gökçeada Island (Northern Aegean Sea, Turkey) with gill nets by fisherman or
Received: 19.02.2020	February 2, 2020. Its length and weight were the maximum length record of saddled seabream for
Received in revised form: 09.03.2020	Northern Aegean coasts of Turkey.
Accepted: 14.03.2020	
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Keywords:	-
Oblada melanura	
Saddled seabream	
Maximum length	
Gökçeada	
Turkey	

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#### Introduction

The saddled bream (*Oblada melanura* Linnaeus, 1758) is common throughout the Mediterranean and eastern Atlantic, inhabiting littoral waters above rocky bottoms and posidonia beds, up to 30 m depth (Bauchot and Hureau, 1986). They are omnivorous but feed mainly on small invertebrates (Froese and Pauly, 2019).

Throughout the world, the information on the growth and reproductive of *O. melanura* were given by Zaki et al. (1995) and Mahmoud (2010) from Egypt, by Pallaoro et al. (1998) from Eastern Adriatic. The feeding habits were studied by Pallaoro et al. (2003, 2004), as a summary. There are no studies



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about biological parameters of this species, except of its lengthweight relationships in the Turkish seas.

Accurate estimates of the maximum size of fish in a population are important for biologists and ecologists because biological rates and ecological functions are size-specific (Peters, 1983; Pope et al., 2005). For example, metabolic rate is inversely related to body size, whereas total food intake is positively related to body size. Size at hatch, size at sexual, maturation and longevity are directly related to maximum size of fishes (Freedman and Noakes, 2002; van der Veer et al., 2003). Maximum length or weight is a key component in many fishery models, such as the von Bertlanffy and Gompertz growth models (Quinn and Deriso, 1999). This study presents the maximum length of *O. melanura* for the Northern Aegean coasts of Turkey.

#### **Material and Methods**

Gökçeada Island, the westernmost point and the largest island of Turkey, is located in the Northern Aegean Sea at the entrance of Saros Bay. The waters coming from the Black Sea and Marmara Sea, mixing with the warmer saltier water of the Aegean Sea, forms a rich marine ecosystem. For this reason, the fishing is quite vital for the Island.

A single specimen of *O. melanura* was obtained off Gökçeada Island (Figure 1) with gill nets by a fisherman on February 2, 2020. Total length is defined as the measurement taken from the anterior-most part of the fish to the end of the caudal fin rays when compressed dorso-ventrally (Anderson and Gutreuter, 1983). Therefore, the specimen was subsequently measured to the nearest mm and weighted to the nearest g.

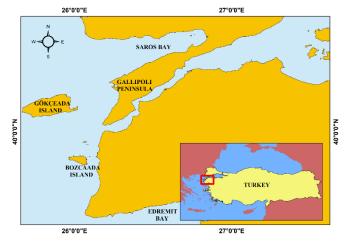


Figure 1. The Northern Aegean coasts of Turkey and Gökçeada Island

#### Results

A single specimen of *O. melanura* with 29.1 cm in total length and 390.00 g in total weight (Figure 2) was obtained off Gökçeada Island.



**Figure 2.** *O. melanura* with 29.1 cm in total length and 390.00 g in total weight

Author(s)	Area	Ν	Fishing Method	L <sub>max</sub> (cm)	W <sub>max</sub> (g)
Karakulak et al. (2006)	Gökçeada Island	25	Gill and trammel nets	28.2	-
Cengiz (2013)	Gallipoli Peninsula	97	Handline, gill and trammel nets	26.1	222.36
Öztekin et al. (2016)	Gallipoli Peninsula	4	Longline	25.8	207.00
This study	Gökçeada Island	1	Gill nets	29.1	390.00

Table 1. The comparison of the lengths and weights for the saddled seabream in the Northern Aegean coasts of Turkey

It has been recorded the maximum length of the species in the Mediterranean to be 35.7 cm in total length (Akyol et al., 2014). The comparison of the lengths and weights for the saddled seabream in the Northern Aegean coasts of Turkey is given in Table 1.

If a fish population in any ecosystem is exposed to overfishing, fish sizes will gradually be smaller over time.

Therefore, individuals who are not subjected to overfishing could reach such a length. However, the factors affecting growth could state as nutrient availability, feeding, light regime, oxygen, salinity, temperature, pollutants, current speed, nutrient concentration, predator density, intra-specific social interactions, and genetics (Helfman et al., 2009; Acarli et al., 2018). Hereby, it follows from these comments that the regional



differences in maximum length and weight depend on the ecological conditions and overfishing pressure (Cengiz, 2019; Cengiz et al., 2019a). The northern Aegean Sea is mainly affected by upwellings. The upwellings occur in the Aegean Sea (Metaxas, 1973) due to summer's (August-September) strong northerly winds. Due to the subsurface cool water upwellings, surface temperature differences create a thermal front between the eastern and western regions of the northern Aegean Sea (Zodiatis and Balopoulos, 1993). Moreover, the less saline and nutrient-rich Black Sea inflow is possibly an important factor in changes in environmental conditions (Altin et al., 2015).

#### Conclusion

Maximum length and weight are important parameters used in life history studies and fishery science. (Borges, 2001; Cengiz et al., 2019b). These measurements are necessary for population dynamics and stock assessment studies. Hence, the recording of such data may be beneficial for scientific databases for life history and fisheries science (Cengiz et al., 2019c). This finding will play an important role in fisheries management.

#### **Conflict of Interest**

The authors declare that there is no conflict of interest.

#### **Ethical Approval**

For this type of study, formal consent is not required.

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# Marine Science and Technology Bulletin

#### **RESEARCH ARTICLE**

# Preliminary results on the growth of larval European lobster (*Homarus gammarus* (Linneaus, 1758)) in Turkey

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#### ABSTRACT

Sea lobsters are among the most valuable seafood traded commodities. Since its production with fishing has decreased every year in the world, many types of cultural studies have been carried out and success has been achieved. The aim of this research is to investigate the survival of European lobster (*Homarus gammarus*) larvae in Turkey and to determine the nature of the direction of growth opportunities. This research is the first study in Turkey investigating the nature of the direction of growth opportunities and the survival of European lobster larvae. Larval release, larvae feeding and survival possibilities were investigated on two egg-bearing lobsters obtained from Çanakkale coasts. Both broodstock larvae were able to survive until the post larval stage. The larvae of the first mature lobster reached 10.857 mm total length and 0.025 g live weight after approximately 30 days. The larvae of the second mature lobster reached 26.9 mm total length and 0.502 g live weight after 33 days. A significant difference was found in the larvae of two mature lobsters at the end of the experiment according to their initial dimensions in both length and weight (p <0.05). In addition, it was determined that the growth was higher due to the higher temperature in the larvae of the second mature lobster.

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#### Introduction

FAO (2020) reported that world human population will have increased by about 34% until 2050, reaching to some 9.1 billion people. To be able to meet this increase with in nutrition, the current food output will have to augment by almost 70% of it. To equilibrate failure of the resultant supply to meet potential demand, protein sources from sea and freshwaters will be of vital importance in terms of feeding humans. Considering the present water sources under pressure of overfishing, likelihood to increase the concerned output is rather poor. Therefore, the most efficient supply for sources is aquaculture production of sea food which has been used through history (Jardas and Pallaoro, 1992). The species European lobster, Homarus gammarus has a region of distribution confined to the continent of Europe. The species has a wide geographic region over Atlantic Ocean in which it inhabits. Moreover, it spreads along the east coast of Europe from Sweden, Norway, Denmark, Lofoten Islands to UK, Ireland and Southern Morocco. Although it spreads less extensively, its presence has been determined along the Mediterranean and the Black Sea as well (Cobb and Castro, 2006: Prodöhl et al., 2007).

Considering global production of H. gammarus species, it was 3000 tons in 1950 increasing to 4800 tons in 1964 and in later years decreasing to 1739 tons in 1979 as the lowest value. However, it ranged from 4000 to 5600 tons following 2006 and reached to 4688 tons in 2018. Of European nations, Britain is the first by 3019 tons in the production from catching processes (FAO, 2020). The early records on trading lobster in Turkey date back to 1925 (21,888 kg). It was reported from Istanbul Fish Market that lobster was sold of 19,431 kg, 23,569 kg and 17,975 kg in 1921, 1922 and 1923, respectively (Deveciyan, 2011). Although production of lobster varied over the years, it increased to 60 tons in 1998 and in later years gradually decreased to 5 tons in 2018. Much of the catch has been obtained from the Aegean Sea and the rest from the Marmara Sea. It can be concluded that in recent years lobster production from catching processes has significantly decreased and import has been gradually increasing to meet the current demand for the product. In 2017, 38 tons of lobster in live and frozen forms (3,610,000 USD) were imported whereas Turkey's lobster export was only 1 ton (260,000 USD) in the same year (FAO, 2020).

It is clear that lobster populations in Turkey has been exposed to pressures caused by over fishing and other processes such as illegal fishing, pollution, degradation of habitat and predator pressure, etc. Therefore, studies and researches have to be conducted aquaculture processes for this species in Turkey. However, investigations are mainly focused on freshwater crayfish species in Turkey (Berber, 2005; Balık et al., 2006; Berber and Balık, 2009; Berber and Mazlum, 2009; Berber et al., 2010, 2011, 2012, 2019; Akhan et al., 2014; Türel et al., 2015; Türel and Berber, 2016; Berber and Kale, 2018). On the other hand, the studies on lobsters conducted are mostly related to those of species-specific artificial reefs (Acarli et al., 2018; Acarlı and Kale, 2020a, 2020b), taxonomy and reproduction biology concerning localities where the species is distributed in Turkey (Balkıs et al., 2002; Kocataş and Katağan, 2003; Bakır et al., 2014; Gönülal and Güreşen, 2014; Erkan and Ayun, 2014).

As for feeding difficulties in larval stages, cannibalism, and effects of environmental factors, rates of survival and growth for the species *H. gammarus* are observed to be low. Therefore, the present study aimed at determining and improving the growth stages of European lobster larvae in eggs and just after hatching processes and studying possibilities of their growth under controlled conditions.

#### Materials and Methods

The present study, which is the first performed study on the determination of larval stages of European lobster in Turkey, was conducted at Marine Life Research and Application Center at Dardanos, Faculty of Marine Sciences and Technology, Çanakkale Onsekiz Mart University in Çanakkale from January 15 to May 11, 2015.

Two individuals of *H. gammarus* with eggs in their gonads were used, which are captured by fishermen off Karabiga, Çanakkale, in this study. They were transferred at the optimum conditions to the research center and separately placed into the two tanks of 500 L. The adults were fed with fresh fish and mussels, and leftovers siphoned from the feeding site. After eggs hatching out, free larvae were picked up using sieves and taken back into the tanks. Measurement of length and weight were made on lobster larvae on a daily with an electronic caliper. Larvae fed with enriched 0.5 L *Artemia* per day one. YSI Pro 2030 and WTW 3110 multimeters were used for temperature, dissolved oxygen, pH, and salinity measurements in the tanks.

Eggs of European lobster were taken to the laboratory in saline water without adding any fixative substance to avoid potential variation in diameters to measure and photograph them with no delay. Every ovum and its ovular diameter were measured and recorded. External capsules of some eggs were opened (exposed) using devices called pin wises to photograph embryo and organs, which was all performed by Olympus SZX7 stereo microscopy attached with v Q-Image Micro Publisher 3.3 RTV imaging program in the laboratory.

After adult lobsters with eggs in their gonads were placed on the study field, larval stages were determined from egg samples every three days based on development of water temperature. A



total of 20 eggs were taken from the different points of gonads attached to the abdomen every three days. Water in 75 cm diameter cylindrical conic tanks was arranged to be changed by 100% every three hours in the first 2 days then by 100% once a day. Tanks were cleaned every 24 hours. Larval density in the tanks were arranged to be 25-30 individuals per liter. For larvae feeding, green water technique of *Nannochloropsis sp.* in 400- $800 \times 10^{-1}$  cell/mL was provided and *Artemia salina* given to meet demand for nutrition as well. When phytoplankton was being entered, *A. salina* started to be given to the medium (5mL twice a day).

#### Statistical Analysis

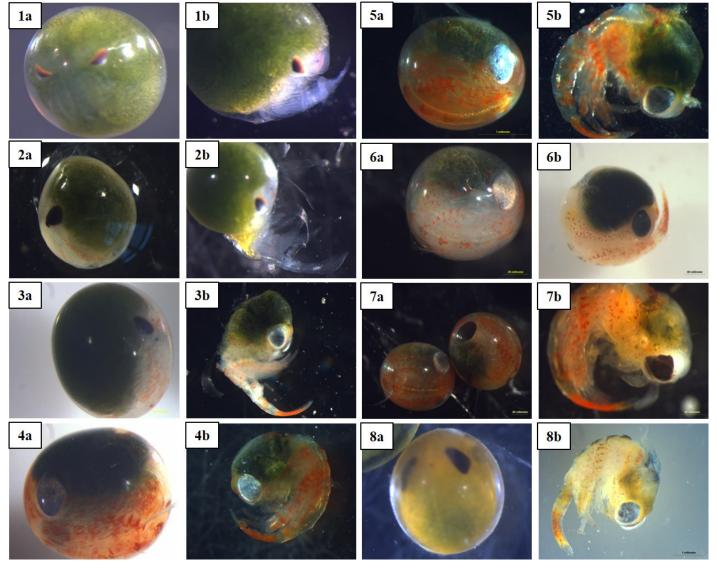
Data were obtained for regression analysis and analyzed using one-way analysis of variance (ANOVA) to examine the effects of each passing day on the growth. Differences were considered significant at 0.05 significance level. All statistics analyses were evaluated using SPSS 19.0 statistical package.

#### Results

One of the physical properties of sea water used during the trial, temperature in particular was found to increase in larval hatching of the second adult with a significant impact on the larval development. Other properties were seen to be stable in the experiment during the study period (Table 1).

#### Developmental Stages of Embryos and Larvae

The egg size (width and length) and the embryo's eye size (length and width) are shown in Table 2. Table 2 also presents the measurements of length, width and values of eye size and width of the embryo.



**Figure 1.** Embryonic development stages of *Homarus gammarus* green yolk 100% (1a-1b), consuming 20% of green yolk (2a-2b), consuming 30% (3a-3b), consuming 50% (4a-4b), consuming 60% (5a -5b), consuming 70% (6a-6b), consuming 80% (7a-7b), and the appearance of the larva that is about to hatch (8a-8b)







Figure 2. Larval development stages of Homarus gammarus; stage 1 (a), stage 2 (b), stage 3 (c), stage 4 (d) and juvenile (e)

Table 1. Measurements of temperature (°C), pH, dissolved oxygen  $(O_2)$  (mg/L) and salinity (S) (‰) in the experiments (SE: standard error)

Experiments (Duration)	Values	Temperature (°C)	Dissolved oxygen (O <sub>2</sub> mg/L)	pН	Salinity (‰)
Experiment 1	<i>x</i> =±SE	11.26±0.243	6.73±0.144	8.19±0.032	28.48±0.309
(25 days)	min-max	8.9-12.6	5.46-7.73	8.01-8.44	25.3-31.1
Experiment 2	<i>x</i> ±SE	12.96±0.131	7.24±0.145	$8.55 \pm 0.056$	28.27±0.223
(33 days)	min-max	12.4-13.4	6.79-7.83	8.37-8.72	27.3-29.1

**Table 2.** Measurements of length, width and values of eye size and width of the embryo (EW: egg width, EL: egg length, EEL: eye length,EEW: eye width; SE: standard error)

Experiments	EW±SE (µm)	EL±SE (µm)	EEL±SE(µm)	EEW±SE (µm)
Experiment 1	2.144±0.0056	$1.974 \pm 0.0059$	$0.683 \pm 0.042$	$0.509 \pm 0.042$
min-max	1.9-2.48	1.63-2.3	0.44-0.76	0.21-0.71
Experiment 2	2.165±0.033	$2.452 \pm 0.035$	$0.709 \pm 0.028$	$0.497 {\pm} 0.017$
min-max	1.99-2.31	2.3-2.648	0.609-0.838	0.408-0.548





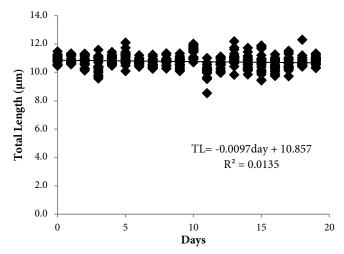
Considering egg development stages, especially consumption of nutrition sac and its related color, a 30% consumption showed the sac with dark green color on it and its gradual consumption indicated a more visible body form with the eye turning from bright and light color to darker in tone (Figure 1, illustrations 3a and 3b). The egg membrane was torn apart to take the embryo out and to study its organelles. The process when hatching was about due showed the nutrition sac above the eye and preopod development was apparent. The abdomen was found to be in a visible extensional form and change to stage 1 in character when the hatching was due (Figure 1, illustrations 8a and 8b). The stages were examined by observing larval activities in the tank as well as microscopic examinations to establish development of larvae during the study. Accordingly, 4 larval and 1 juvenile stages were determined (Figure 2). For stage 1, pigmentation was the first characteristic in larval development in newly hatched individuals. Although the eye aperture did not grow in volume, variation was hardly observed to emerge in body length index until the first molting. Even if rostrum pointedness was not much, it was visible. Development of clamp was not strengthened yet (Figure 2a). In stage 2, coloration was seen to increase. Size of the eye was more obvious than in stage 1 and rostrum pointedness became clearer (Figure 2b). Development of clamps and preopods was found to be satisfactory enough to catch foods in suspension. Because pleopods and telsons did not sufficiently develop, larvae could not swim freely and suspended on water. Moreover, another significant characteristic at this stage is that development of clamp, preopod and telson enabled them to begin to swim on water and strengthening and deepening of clamp scissors emerged. Juvenile stage emerges until the period of time when growth, mating, spawning and incubation each has become part of annual cycle and those which has reached to this stage molt less frequently than previous stages. Individuals at juvenile stage hardly differ than adults. The front body was found to strengthen with visible hairs. Due to pointedness of rostrum, it was observed to elongate towards frontally in a way to effect body length (Figure 2c). At stage 4, mean carapace length, total length and weight were 3.75 mm, 12.6 mm and 0.0245 g, respectively. Individuals at post larval stage resembled adults but variously represented a stage of transition (Figure 2d).

#### Larval Growth

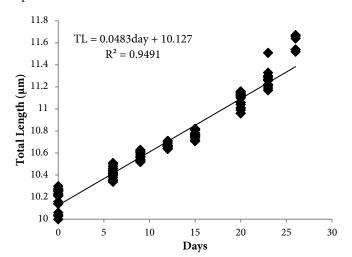
A significant increase was not found (p>0.05) when growth characteristics of larvae hatched on February 13, 2015 from the first adult until April 10, 2015 were examined (Figure 3) whereas those hatched from the second adult on April 15, 2015 showed significant differences in growth until May 18, 2015 (p<0.05; Figure 4).

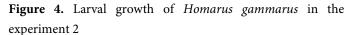
#### Discussion

Temperature is widely known to have an impact on gonad and embryonic development of Crustacea species in the way it has on other living organisms (Acarli and Lök, 2009; Yildiz et al., 2011; Küçükdermenci and Lök, 2012; Acarli et al., 2015, 2018). Agnalt et al. (2013) reported that lobster development exhibits a positive relationship with temperature. Optimal water temperature is generally 20-22°C for *H. gammarus* species (Prodöhl et al., 2007). Moreover, lobster larvae are more tolerant to low temperatures than young or adult individuals. At 20°C larval period ends for about 20 days while it extents to 35 days at 15°C (van Olst et al., 1980). It was found that healthy larval development did not occur below 14°C. Schmalenbach and Franke (2010) reported that survival rate of *H. gammarus* larvae increased from 9% at 14°C to 80% at 22°C and its larval development decreased from 26 to 13 days.



**Figure 3.** Larval growth of *Homarus gammarus* in the experiment 1





During the study, sea water filtered and fed to the system was used and no interference was made to increase temperature of water. Temperature of water was measured by 11.26°C and 12.60°C at the first and the second trials, respectively. The study showed that individuals could reach to juvenile stage for 33 days, expansion of which is believed to be temperature as the most effective factor. Although temperature below 14°C retarded larval development significantly, it still continued to develop. Schmalenbach and Franke (2010) reported that the molting did not occur under 10°C.

Molting in Decapods are affected by salinity, light density, social interaction, volume of habitat and water quality (Mikami and Kuballa, 2007). Considering the parameters below in terms of ideal water properties in studies on growing larvae, changes of salinity, pH and dissolved oxygen have been reported to have to be above 29-35‰, 7.8-8.2 and 8 mg/L, respectively (Burton, 2003). The lowest salinity tolerance in *H. americanus* was found to be 13.8‰ and 8‰ for larvae and young adults, respectively (Cobb, 1976). Under natural conditions, lobsters especially at larval stages do not prefer areas with salinity rate below about 20‰ (Fefer and Schetting, 1980). Low pH increases physiologic stress and affect individuals already under metabolic stress negatively (Agnalt et al., 2013). Salinity and pH of sea water used for larvae in the study are assumed to be at appropriate values for the organisms to growth.

Though food quality is considered an important factor which increases and controls productivity in decapod larvae, what is known is relatively little about food requirement and zooplankton for larval growth. One of the reasons for this is absence of an efficient nutrition which is acceptably digestible (Meyers, 1973, 1979; Eagles et al., 1986). For feeding larvae, European hatcheries uses minced fish, bivalves (Wickins and Beard 1991; Nicosia and Lavalli, 1999; Burton, 2003) and live baits such as Artemia spp. and Acartia tonsa as well as and wet or damp plankton preparations until recently (Fiore and Tlusty, 2005; Scolding et al., 2012). However, larval survival rate and growth rate of Homarus spp. is negatively affected especially when amount of nutrition has been insufficient in high density culture studies. One of the ways to reduce cannibalism to a minimum is to increase food density and thus prevent larvae starving much. In recent years, trials have been conducted to use ready-made feeds and rations prepared to meet content needed by larvae, which could not change importance of Artemia at all (Fiore and Tlusty, 2005; Powell et al., 2017). Their natural diets are composed of copepods and zooplankton as well as phytoplankton in less rate but feeds to be provided under culture conditions are supposed to have ability to produce high level of proteolytic enzyme. Since digestive enzymes of the carnivorous larvae are quite low, they have poor

capacity to benefit from artificial feeds thus can feed on zooplankton such as copepods and Artemia. Recent developments in uses of micro capsules has enabled achievements to emerge in meeting nutritional requirements of penaeid shrimp larvae, which is promising in their uses for lobster larvae as well. The fact that recent developments in uses micro capsules have led to successful results for meeting nutritional requirements of penaeid shrimp larvae is promising in potential uses for lobster larvae as well (Meyers, 1973, 1979; Beal et al., 2002; Jørstad et al., 2005; Scolding et al., 2012; Drengstig and Bergheimb, 2013; Daniels et al., 2015). Evjemo et al. (2009) reported that larvae fed with formulated diets showed very poor development and were able to reach to stage 2 only after 20 days. The authors determined that Artemia-fed individuals entered stage 5 the same period with a survival rate of 91-94%. Lobster larva can ideally be fed with live Artemia but cannibalism occurs when given diets have been tasteless or insufficient (Wickins and Lee, 2002). It is known that H. gammarus generally have poor digestive enzyme activity. In other words, the species has very low stomach enzymes of trypsin and chymotrypsin though high activity of cathepsin L in their stomach fluid different from many other Decapod species, which has developed a strategy of keeping ingested foods long in the stomach to increase their digestion. High energy content and easily digestible food is needed to increase larval survival and growth rates in lobster aquaculture, in which context A. salina is also chosen as an important food (Kurmaly et al., 1990; Kumlu and Jones, 1997). At initial developmental stages of Homarus sp. larvae, Artemia nauplii is widely used. A. salina was employed as food in the present study. Individuals were grown until juvenile stage with length and weight from 10 mm to 25.60 mm and 0.023 g to 0.34 g, respectively.

In comparison with other lobster species, *Homarus* species including European lobster species are accepted as very resistant ones to thanks to their simple and short larval periods. However, production dynamics need to be comprehended well in order to be able to ideally manage present lobster stocks. Special feeding requirements are little understanding in larval survival and growth which are cited among the reasons for commercial inventorial fluctuations in the market.

Annually prepared and declared official statistics on sea foods indicate that they tend to decrease in parallel to current stocks due to output from catching processes. However, output amounts of species grown from aquaculture is observed to continuously increase. Similarly, production of marine lobster from fisheries is known to decrease every year. Decrease in natural stocks and necessity to protect natural sources, their high values of food and economics and employment potentialities if realized are among justifications for



aquaculture related to marine lobster. Although growth results from the present study are low as compared to those of other research, positive and promising signals exist under limited means. Environmental conditions such as temperature and abundance of nutrition tend to effect meroplanktonic larval development as well as distribution and quantity of populations (Kirby et al., 2007; Jackson et al., 2014). The conducted studies showed that regions where insufficient amount of food in the environment specifically have impact on survival and growth of Crustacean and fish larvae (Olson and Olson, 1989). On condition that Crustacean larvae have not sufficiently been fed, their hepatopancreatic cells would be irreversibly affected (Storch and Anger, 1983). Studies to be made further are supposed to focus on determining appropriate conditions for optimum output productivity and solutions to the problems of feeding at larval and juvenile stages.

#### Conclusion

In this study, the larval development stages of *H. gammarus* were investigated under two different temperature values. The results showed that high temperature has an increasing effect on larval development. The larvae of the first mature lobster reached 10.857 mm total length and 0.025 g live weight after approximately 30 days. The larvae of the second mature lobster reached 26.9 mm total length and 0.502 g live weight after 33 days. A significant difference was found in the larvae of two mature lobsters at the end of the experiment according to their initial dimensions in both length and weight (p <0.05). All developmental stages of *H. gammarus* larvae were observed and they were able to survive until juvenile stage by the present study. This study is the first study in Turkey on the growth of *H. gammarus* larvae. The preliminary results of the present paper will encourage the further investigations on the subject.

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This paper is a part of MSc thesis of the first author.

#### **Compliance with Ethical Standards**

#### Authors' Contributions

Author SB designed the study, SA and AB wrote the first draft of the manuscript, performed and managed statistical analyses. EÖ worked in the experimental studies. All authors read and approved the final version of the manuscript.

#### **Conflict of Interest**

The authors declare that there is no conflict of interest.

#### Ethical Approval

For this type of study, formal consent is not required.

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#### SHORT COMMUNICATION

### An observation about maximum size record of blotched picarel (*Spicara maena* Linnaeus, 1758) from Northern Aegean coasts of Turkey

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ARTICLE INFO	ABSTRACT			
Article History: Received: 22.01.2020 Received in revised form: 28.03.2020 Accepted: 30.05.2020 Available online: 30.05.2020	Maximum length and weight are important parameters and they are commonly used in life history studies and fishery science. Therefore, it is important to regularly bring up to date the maximum size of commercially important species. The accurate estimates of the maximum size of fish in a population are important issues. Because the parameters related			
Keywords: Spicara maena Blotched picarel Maximum size Saros Bay	to maximum length, weight and age in fish communities within an ecosystem are constantly used in population dynamics and stock estimation studies, recording of such data is vital for determining the life history of fish. In this connection, a single specimen of blotched picarel ( <i>Spicara maena</i> ) with 20.3 cm in total length and 159.00 g in total weight was caught off Saros Bay (Northern Aegean Sea, Turkey) with handline at 20 m water depth			
Turkey	by a commercial fisherman on October 20, 2019. Its length and weight were the maximum values of <i>Spicara maena</i> for Saros Bay.			

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#### Introduction

Blotched picarel (*Spicara maena* Linnaeus, 1758) is a commercial species inhabiting the Mediterranean Sea, the Black Sea, and the European and African coasts of the Atlantic Ocean, from Morocco to Portugal and the Canary Islands (Jardas, 1996). This species mostly occurs over Posidonia beds and sandy or muddy bottoms, and distributes up to 100 m depth. *S. maena* feeds on mainly zooplankton and is a protogynous hermaphrodite (Froese and Pauly, 2019).

As to Turkish seas, information on the biology of species come from Saros Bay (Cengiz, 2019), Gallipoli Peninsula (Cengiz et al., 2014), Sea of Marmara and Edremit Bay (Saygılı et al., 2016), Izmir Bay (Soykan et al., 2010) and Babadillimani Bight (Çiçek et al., 2007), as a summary.

Maximum length and weight are important parameters used in life history studies and fishery science. These measurements applied directly or indirectly in most stock assessment models (Borges, 2001). Therefore, it is important to regularly update the maximum size of commercially important

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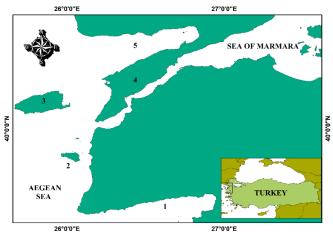
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species (Navarro et al., 2012). Its length and weight were the maximum values of *Spicara maena* for Saros Bay (Northern Aegean Sea, Turkey).

#### **Material and Methods**

Saros Bay, which is situated in the Northeastern Aegean Sea, is connected to the North Aegean Sea with a depth of approximately 600 m to the west. The shelf extends at a water depth of 90-120 m. The length of the bay is about 61 km and the width at the opening to the Aegean Sea is about 36 km (Eronat and Sayın, 2014). As Saros Bay had been closed to bottom trawl fishing since 2000 (Cengiz et al., 2011) and no industrial activity was prevalent in the area (Sarı and Çağatay, 2001), the bay can be considered as a pristine environment (Cengiz et al., 2013; Cengiz et al., 2019).

A single specimen of *Spicara maena* was caught off Saros Bay (Figure 1) with handline by a commercial fisherman from 20 m depth on October 20, 2019. Total length is defined as the measurement taken from the anterior-most part of the fish to the end of the caudal fin rays when compressed dorso-ventrally (Anderson and Gutreuter, 1983). Therefore, the specimen was subsequently measured to the nearest mm and weighted to the nearest g.



**Figure 1.** The Northern Aegean coasts of Turkey (1: Edremit Bay; 2: Bozcaada Island; 3. Gökçeada Island; 4: Gallipoli Peninsula; 5: Saros Bay)

#### **Results and Discussion**

The blotched picarel obtained from Saros Bay was 20.3 cm in total length and 159.00 g in total weight (Figure 2). The comparison of the maximum lengths and weights *Spicara maena* for Northern Aegean coasts of Turkey is given in Table 1.



**Figure 2.** The blotched picarel with 20.3 cm in total length and 159.00 g in total weight

The accurate estimates of the maximum size of fish in a population are important for biologists and ecologists because biological rates and ecological functions are size specific (Peters, 1983; Pope et al., 2005). If a fish population in any ecosystem is exposed to overfishing, fish sizes will gradually be smaller over time. Therefore, individuals who are not subjected to overfishing could reach such a length (Filiz, 2011). However, the factors affecting growth could state as nutrient availability, feeding, light regime, oxygen, salinity, temperature, pollutants, current speed, nutrient concentration, predator density, intraspecific social interactions and genetics (Helfman et al., 2009; Acarli et al., 2018). It could be possible that the sampled specimen had reached to such length on account of the high nutritional concentration and intensive feeding activities.

Table 1. The comparison of the maximum	lengths and	weights Spicara	maena for Northern Aegean coasts of Turke	y

Author(s)	Area	Ν	$L_{max}(cm)$	$W_{max}\left(g ight)$
Karakulak et al. (2006)	Gökçeada Island	830	22.0	-
İşmen et al. (2007)	Saros Bay	353	17.8	67.00
Karakulak and Erk (2008)	Gökçeada Island	897	21.9	-
Altın et al. (2015)	Gökçeada Island	77	16.8	55.31
Saygılı et al. (2016)	Edremit Bay	168	18.8	157.88
Cengiz (2019)	Saros Bay	620	17.8	82.23
This study	Saros Bay	1	20.3	159.00

*Note:* \*;  $L_{max}$  is the maximum total length,  $W_{max}$  is the maximum total weight





#### Conclusion

In order to obtain more reliable results from the studies on fish stocks, if possible, it is important that such proven researches is carried out separately for each fish species along with its weight-height relationships and updated within certain time periods. Thus, the findings to be obtained in the light of this information can reveal the current situation of stock more clearly. This enables the strategies planned in fisheries management to be built on more solid foundations.

#### **Compliance with Ethical Standards**

#### **Conflict of Interest**

The author declares that there is no conflict of interest.

#### **Ethical Approval**

For this type of study, formal consent is not required.

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