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A Study on Zooplankton Fauna and Some Water Quality Parameters of Kozan Dam Lake (Adana, Turkey)

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ABSTRACT

Some water quality parameters (Secchi disk depth, water temperature, dissolved oxygen, pH, conductivity, chlorophyll a, NO_2-N, NO_3-N, NH_4-N, PO_4-P) and zooplankton fauna were determined in the Kozan Dam Lake. While the quality of the reservoir water was first class water in terms of temperature, dissolved oxygen, NH_4-N, PO_4-P, it was second-class water in terms of conductivity, pH, NO_3-N. In total, 50 zooplankton species belonging to 26 families were determined (29 species belonging to 17 families from Rotifera, 15 species belonging to 7 families from Cladocera and 6 species belonging to 2 families from Copepoda). Brachionidae (Rotifera) was the most species rich family with 7 species, followed by Chydoridae (Cladocera) and Cyclopidae (Copepoda) with 6 and 5 species respectively. The most dominant species were *Synchaeta pectinata* (38.33%) from Rotifera, *Bosmina longirostris* (5.71%) from Cladocera and *Cyclops vicinus* (0.67%) from Copepoda. At the same time, the species found in every month were *Asplanchna priodonta*, *Polyarthra dolichoptera*, *Bosmina longirostris*, *Ceriodaphnia pulchella*, *Cyclops vicinus* and *Diacyclops bicuspidatus*. In the study, Rotifera was the most abundant group with 67%, followed by Cladocera with 29% and Copepoda with 4%. On the other hand, total Rotifera was found mostly in December (10099 individual/ m^3), Cladocera in January (4928 ind./m^3) and Copepoda in September (1091 ind./m^3).

Keywords: Rotifera, Cladocera, Copepoda, Kozan Dam Lake

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Kozan Baraj Gölü (Adana, Türkiye) Zooplankton Faunası ve Bazı Su Kalite Parametreleri Üzerine Bir Çalışma

Öz: Kozan Baraj Gölü'nde bazı su kalitesi parametreleri (Secchi derinliği, su sıcaklığı, çözünmüş oksijen, pH, iletkenlik, klorofil *a*, NO_2-N, NO_3-N, NH_4-N, PO_4-P) ve zooplankton faunası belirlenmiştir. Baraj gölü suyu sıcaklık, çözünmüş oksijen, NH_4-N, PO_4-P açısından birinci sınıf su iken, iletkenlik, pH, NO_3-N açısından ikinci sınıf sudur. Rotifera'dan 17 familyaya ait 29 tür, Kladosera'dan 7 familyaya ait 15 tür ve Kopepoda'dan 2 familyaya ait 6 tür olmak üzere toplam 50 tür tespit edilmiştir. Brachionidae (Rotifera) 7 türle en zengin familya olup, bunu 6 ve 5 tür ile Chydoridae (Cladocera) ve Cyclopidae (Copepoda) familyalarının takip ettikleri belirlenmiştir. En baskın türün Rotifera'dan *Synchaeta pectinata* (%38,33), Kladosera'dan *Bosmina longirostris* (%5.71) ve Kopepoda'dan *Cyclops vicinus* (%0.67) olduğu belirlenmiştir. Araştırmada her ay bulunan türler rotiferlerden *Asplanchna priodonta*, *Polyarthra dolichoptera*, kladoserlerden *Bosmina longirostris*, *Ceriodaphnia pulchella*, kopepodlardan *Cyclops vicinus* ve *Diacyclops bicuspidatus*'tur. Çalışmada Rotifera'nın %67 ile en çok bulunan grubu oluşturduğu, bunu %29 ile Kladosera'nın ve %4 ile Kopepoda'nın takip ettiği bulunmuştur. Öte yandan toplam Rotifera'nın en çok Aralık'ta (10.099 birey/m^3), Kladosera'nın Ocak'ta (4.928 birey/m^3) ve Kopepoda'nın Eylül'de (1.091 birey/m^3) bulundukları belirlenmiştir.

Anahtar kelimeler: Rotifera, Kladosera, Kopepoda, Kozan Baraj Gölü

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Introduction

Turkey has very rich potential in terms of lakes and dam lakes. The dams have been built in order to control the regime of the rivers and meet the various needs (eg. drinking water supply, irrigation, flood control, and energy generation) of the people have increased this potential in recent years.

The zooplankton have long been recognized as a secondary producer by occupying almost middle positions of the food chain and indicate environmental status in a given time (Khan 2003).

They have been known as an important energy resource for small sized fish that, in turn, provide energy to piscivorous fish consumers higher up in the food web in fresh water. Zooplankton is known to respond quickly to environmental conditions, and only a few attempts have been made to use the zooplankton community to evaluate the quality of aquatic ecosystems (Lougheed and Chow-Fraser 2002).

Crustacean plankton has been described as preferred fish food items by several authors (Zaret 1972; Dodson 1974; Ayodele and Adeniyi 2006). They are preferred by fishes to their Rotifera counterparts for several reasons. First, they are relatively bigger, and planktivorous fishes which practice size selective predation often prefer them to the rotifers (Ayodele and Adeniyi 2006; Brandl 2002). Crustaceans are more important than rotifers in the transfer of energy from autotrophic phytoplankton to fishes based on their ecological niche in freshwater systems (Williamson 1983). Cyclopoid copepods have been described as effective predators of rotifers, and so are some calanoid species which may include rotifers in their diets too (Williamson and Butler 1986; Schulze and Folt 1990). Aside their importance in fisheries, crustacean plankton (some cyclopoids) are also ecologically important by suppressing mosquito larvae (Alekseev 2002). That's why, studies on zooplanktonic organisms are important for the freshwater ecosystem.

Zooplankton are known to respond quickly to environmental conditions, and only a few attempts have been made to use the zooplankton community to evaluate the quality of aquatic ecosystems (Lougheed and Chow-Fraser 2002). Some zooplankton species are used in various studies as indicators of water quality, pollution and eutrophication status due to their sensitivity to environmental changes (Ruttner-Kolisko 1974; Sharma 1983; Saksena 1987).

A detailed study on the zooplankton fauna in Kozan Dam Lake had not been done before. This study was done to obtain insight into the composition of the zooplankton fauna of the dam lake and to contribute to the knowledge of the biological diversity of inland waters in Turkey.

Materials and Methods

The study was carried out between January 2011 and December 2011 on Kozan Dam Lake, which has 6 km^2 lake area, in the Adana province Kozan district (Figure 1). Zooplankton samples were taken from 4 stations with horizontal and vertical hauls by using 60 μm mesh size plankton nets on a monthly basis for systematic analyses. On the other hand, zooplankton abundance was determined from the samples taken from first two stations (station 1 and station 2). Considered to be enough for analysis, two liters of water samples were collected from every water layer (surface, middle and deep) of first and second stations using Nansen Bottles. Water quality parameters and chlorophyll *a* were analysed from water samples.

One lt and 0.5 lt of the water collected with water sampler was used for chlorophyll *a* analysis and chemical analysis respectively. The remaining part (4.5 lt) was filtered from a collector having a mesh size of 60 μm and zooplankton was fixed in 100 cc glass jars. Dissolved oxygen, water temperature, pH and conductivity were measured directly in the field by means of digital instruments (oxygen and temperature: YSI model 52 oxygen meter; pH: YSI 600 pH meter; conductivity: YSI model 30 salinometer). Merck spectroquant Nova 60 spectrophotometer and its procedure were used to determine NO_2-N, NO_3-N, NH_4-N, PO_4-P; the method in APHA 1995 was used to determine chlorophyll *a* spectrophotometrically. Secchi depth was measured using a Secchi disk with a diameter of 20 cm.

At the stations, the lowest depth was 31 m (1. station), 26 m (2. station), 12 m (3. station) and 10 m (4. Station) in October and the highest depth was 47, 44, 31 and 26 m in May, respectively. Therefore, the depth was approximately 18 m in the year, while the mean depths were 45, 39, 20 and 18 m.

All zooplankton samples were fixed in 4% formaldehyde. Species identifications were made using a binocular microscope according to the works of Edmondson (1959), Scourfield and Harding (1966), Dussart (1967), Kiefer and Fryer (1978), Koste (1978), Negrea (1983), Segers (1995), De Smet (1996, 1997), Nogrady and Segers (2002), Hołynska et al. (2003) and Benzie (2005).

Zooplankton count was performed using an inverted microscope in a petri dish with 2 mm lines at the bottom. The sample cup was made homogenized by shaking and 2 cc sub-sample was taken from the cup and it was placed in a petri dish and the individuals of each species were separately counted. This process has been repeated 4-5 times.

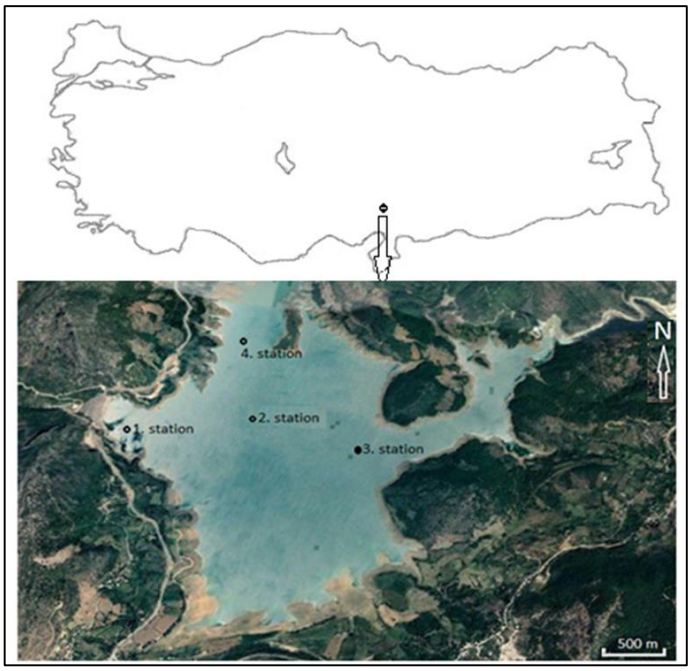

Figure 1. Kozan Dam Lake and Sampling Stations

CTM tolerance of the species (*SPSS* 20.1). Duncan's multiple range test (*DMRT*) was carried out for post hoc mean comparisons. Regression analysis was also carried out to evaluate the relationship between acclimation temperature and *CTMin* and *CTMax* ($p \leq 0.05$).

Results

Maximum, minimum and average values of some water quality criteria were given in Table 1.

Table 1. Maximum, minimum and average values of some water quality parameters.

Parameter	Min.	Max.	Mean±stdev.
Secchi depth (m)	1.40	4.50	2.75± 0.91
W. Temp (°C)	7.97	24.57	16.51± 5.32
Chl. a (mg/m^3)	1.71	7.39	2.96± 1.44
Cond. (µS/cm)	372.00	436.00	406.08± 17.04
DO (mg/l)	5.40	11.53	7.92± 1.85
pH	6.75	9.42	8.22± 0.51
NO_2-N (mg/l)	0.002	0.11	0.024± 0.03
NO_3-N (mg/l	4.31	9.25	6.36± 1.54
NH_4-N (mg/l	0.056	0.188	0.123± 0.04
PO_4-P (mg/l)	0.010	0.053	0.03± 0.012

At the stations, the lowest water depth was 31 m (1. st), 26 m (2. st), 12 m (3. st) and 10 m (4. st) in October and the highest water depth was 47, 44, 31 and 26 m in May, respectively. Therefore, the annual water depth change was approximately 18 m, while the mean depths were 45, 39, 20 and 18 m respectively.

Secchi disk depth reached the maximum depth of 4.50 m in April (station 2) and the minimum depth of 1.4 m on December (station 2), with a mean value of 2.75 ±0. 91 m (Figure 2A). Water temperature varied from 7.97°C (December at second station) to 24.57°C (June at second station) with a mean value of 16.51 ±5.32°C (Figure 2B). Mean chlorophyll *a* concentration was 2.96 ±1.44 mg/m^3 with a range from 1.71 mg/m^3 (at first station) in May to 7.39 mg/m^3 in March (Figure 2C). The conductivity value varied from 372 µS/cm (September at first station) to 436 µS/cm (May at second station) with a mean value of 406.08 ±17.04 µS/cm (Figure 2D). Dissolved oxygen varied from 5.4 mg/l (at first station) in July to a peak of 11.53 mg/l (second station) in January with a mean value of 7.92 ±1.85 mg/l (Figure 2E). pH value did not vary much between the stations. The minimum, maximum and mean pH values were 6.75 (July at first station), 9.42 (March at first station) and 8.22 ±0.51 respectively (Figure 2F). Nitrite nitrogen reached the maximum concentration of 0.11 mg/l (February at first station) and minimum concentration of 0.002 mg/l (October at second station), with a mean value of 0.024±0.03 mg/l (Figure 2G). Nitrate nitrogen (annual average 6.36 ±1.54 mg/l) varied from 4.31 mg/l (October at second station) to 9.25 mg/l (May at second station) (Figure 2H), and ammonium nitrogen (annual average 0.123 ±0.04 mg/l) varied from 0.056 mg/l (February at first station) to 0.188 mg/l (October at second station) (Figure 2I). The maximum, minimum, and mean phosphate values were 0.053 mg/l (November at first station), 0.010 mg/l (January at second station), and 0.03 ±0.012 mg/l, respectively (Figure 2J).

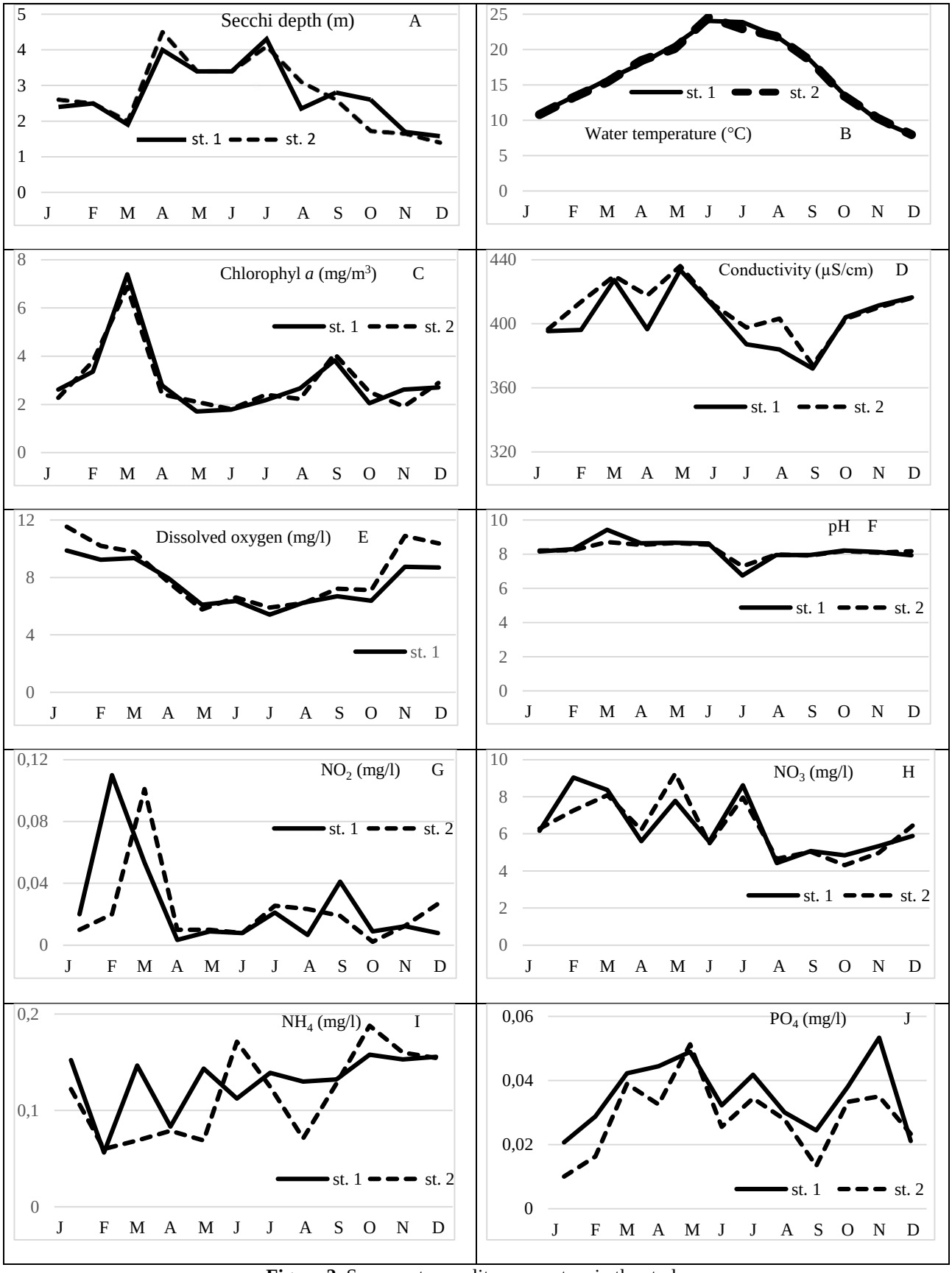

Figure 2. Some water quality parameters in the study

In the study, a total of 50 zooplankton species were identified, including 29 from rotifers, 15 from cladocerans and 6 from copepods.

Seventeen families were identified from Rotifera and Brachionidae was the most species rich family with 7 species, followed by Collothecidae, Hexarthridae, Lecanidae, Lepadellidae, Synchaetidae and Trichocercidae with 2 species each one. The remaining families from Rotifera were found to contain only one species in each one (Table 2). Seven families found from Cladocera, Chydoridae was the most species rich family with 6 species, followed by Daphniidae with 4 species and other families were represented only one species each one (Table 2). Two families were detected from Copepoda, Cyclopidae was the richest family with 5 species, but Ameiridae (Harpacticoida) was represented by only one species.

Table 2. Zooplankton species in the study and their monthly presences

Rotifera	**Months**	**J**	**F**	**M**	**A**	**M**	**J**	**J**	**A**	**S**	**O**	**N**	**D**
Familya: Philodinidae	*Rotaria rotatoria* (Pallas, 1766)	+	-	+	-	-	-	-	-	+	-	-	+
Familya: Collothecidae	*Collotheca pelagica* (Rousselet, 1893)	+	+	+	+	-	-	-	-	+	-	+	+
	Collotheca mutabilis (Hudson, 1885)	-	-	-	+	+	-	-	-	-	-	+	+
Familya: Filinidae	*Filinia terminalis* (Plate, 1886)	-	-	+	-	-	-	-	-	-	-	-	+
Familya: Hexarthridae	*Hexarthra intermedia* (Wiszniewski, 1929)	-	-	+	-	-	-	-	-	-	+	+	-
	Hexarthra oxyuris (Sernov 1903)	-	-	-	-	+	-	-	-	-	-	-	-
Familya: Testudinellidae	*Pompholyx sulcata* (Hudson, 1885)	+	-	+	+	-	+	-	-	+	+	-	-
Familya: Asplanchnidae	*Asplanchna priodonta* (Gosse, 1850)	+	+	+	+	+	+	+	+	+	+	+	+
Familya: Brachionidae	*Anuraeopsis fissa* (Gosse, 1851)	-	-	-	+	-	-	-	-	-	-	-	-
	Brachionus quadridentatus (Hermann, 1783)	-	-	+	-	-	-	-	-	-	-	-	-
	Keratella cochlearis (Gosse, 1851)	-	-	+	+	-	-	-	-	-	-	+	-
	Keratella tecta (Lauterborn, 1900)	-	-	+	-	-	-	-	-	-	-	-	-
	Keratella quadrata (Müller, 1786)	+	-	+	-	+	+	-	+	+	+	+	+
	Keratella tropica (Apstein, 1907)	-	-	-	+	+	-	-	-	-	-	-	-
	Notholca squamula (Müller, 1786)	+	-	+	-	-	-	-	-	-	-	+	-
Familya: Colurellidae	*Colurella adriatica* (Ehrenberg, 1831)	-	-	-	+	-	-	-	-	-	-	-	-
Familya: Euchlanidae	*Euchlanis dilatata* (Ehrenberg, 1832)	-	-	-	-	-	-	-	-	+	-	-	+
Familya: Gastropodidae	*Ascomorpha ovalis* (Bergendahl, 1892)	-	-	-	-	+	+	-	+	+	+	+	-
Familya: Lecanidae	*Lecane bulla* (Gosse, 1886)	-	-	-	-	-	+	-	-	+	-	-	-
	Lecane lunaris (Ehrenberg, 1832)	-	-	+	-	+	-	+	+	+	+	+	-
Familya: Lepadellidae	*Lepadella acuminata* (Ehrenberg, 1834)	-	-	+	-	-	-	-	-	-	-	-	-
	Lepadella ovalis (Müller, 1896)	-	-	+	-	-	+	-	-	-	-	-	-
Familya: Mytilinidae	*Lophocharis salpina* (Ehrenberg, 1834)	-	-	-	-	-	-	-	-	-	-	+	-
Familya: Notommatidae	*Cephalodella gibba* (Ehrenberg, 1832)	-	-	+	-	-	-	-	-	-	+	-	-
Familya: Synchaetidae	*Polyarthra dolichoptera* (Idelson, 1925)	+	+	+	+	+	+	+	+	+	+	+	+
	Synchaeta pectinata (Ehrenberg, 1832)	+	+	+	+	-	+	-	-	-	-	+	+
Familya: Trichocercidae	*Trichocerca similis* (Wierzejski, 1893)	+	+	+	+	+	+	+	+	+	-	+	+
	Trichocerca capucina (Wierzejski and Zacharias, 1893)	-	+	-	-	-	+	+	-	-	-	-	-
Familya: Trichotriidae	*Trichotria tetractis* (Ehrenberg, 1830)	-	+	-	-	-	-	-	-	-	-	-	-

Table 2. Continued

Rotifera Cladocera	Months	J	F	M	A	M	J	J	A	S	O	N	D
Familya: Bosminidae	*Bosmina longirostris* (Müller 1785)	+	+	+	+	+	+	+	+	+	+	+	+
	Alona quadrangularis (Müller, 1776)	-	-	-	-	-	-	-	-	-	+	-	-
	Alona rectangula (Sars, 1861)	-	-	-	-	-	-	-	-	-	+	-	+
	Disparalona rostrata (Koch, 1841)	+	-	-	-	-	-	-	-	-	-	-	+
Familya: Chydoridae	*Chydorus sphaericus* (Müller, 1785)	-	-	-	-	+	-	-	-	-	-	-	-
	Monospilus dispar (Sars, 1861)	-	-	-	-	-	-	-	-	-	+	-	-
	Leydigia leydigi (Leydig, 1860)	-	-	-	-	-	-	-	+	-	-	-	-
	Ceriodaphnia pulchella (Sars, 1862)	+	+	+	+	+	+	+	+	+	+	+	+
	Daphnia galeata (Sars, 1864)	-	-	+	-	-	+	-	-	-	-	-	-
Familya: Daphniidae	*Daphnia longispina* (Müller, 1785)	+	+	+	+	+	+	+	-	+	-	+	+
	Daphnia cucullata Sars, 1862	-	-	+	+	-	+	+	+	+	-	-	-
Familya: Leptodoridae	*Leptodora kindtii* (Focke, 1844)	-	-	-	-	-	+	-	-	-	-	-	-
Familya: Macrothricidae	*Macrothrix laticornis* (Jurine, 1820)	-	-	-	-	-	-	-	-	-	+	-	+
Familya: Moinidae	*Moina micrura* (Kurz, 1874)	-	-	+	-	-	+	-	-	+	+	-	+
Familya: Sididae	*Diaphanosoma birgei* (Korinek, 1981)	+	-	+	-	-	+	+	+	+	+	+	-
Copepoda													
Familya: Cyclopidae	*Cyclops vicinus* (Uljanin, 1875)	+	+	+	+	+	+	+	+	+	+	+	+
	Diacyclops bicuspidatus (Claus, 1857)	+	+	+	+	+	+	+	-	+	-	+	+
	Macrocyclops albidus (Jurine, 1820)	-	+	+	+	-	-	-	-	+	-	-	+
	Mesocyclops leukarti (Claus, 1857)	-	-	-	-	+	-	-	-	-	-	-	-
	Paracyclops fimbriatus (Fischer, 1853)	-	-	-	-	-	-	-	-	-	+		-
Familya: Ameiridae	*Nitocra hibernica* (Brady, 1880)	-	-	-	-	-	-	-	-	-	-	-	+

(+: available, -: absent)

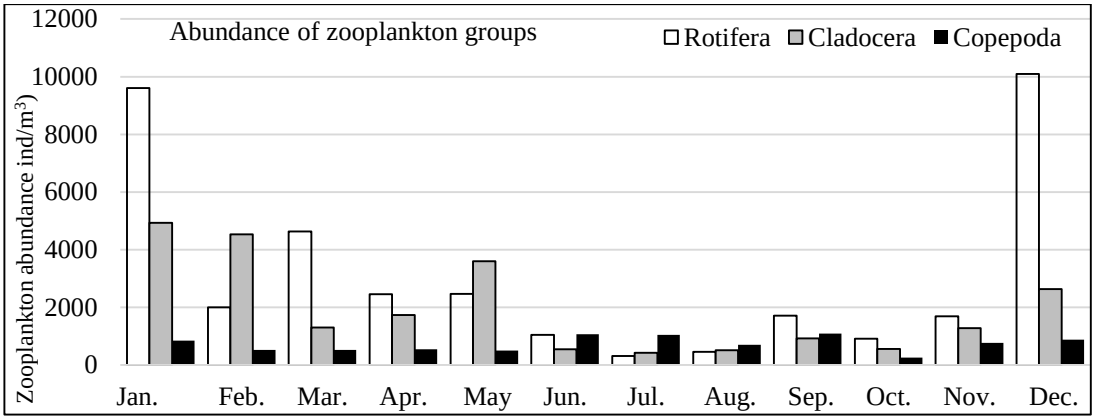

Figure 3. The abundance of zooplankton groups

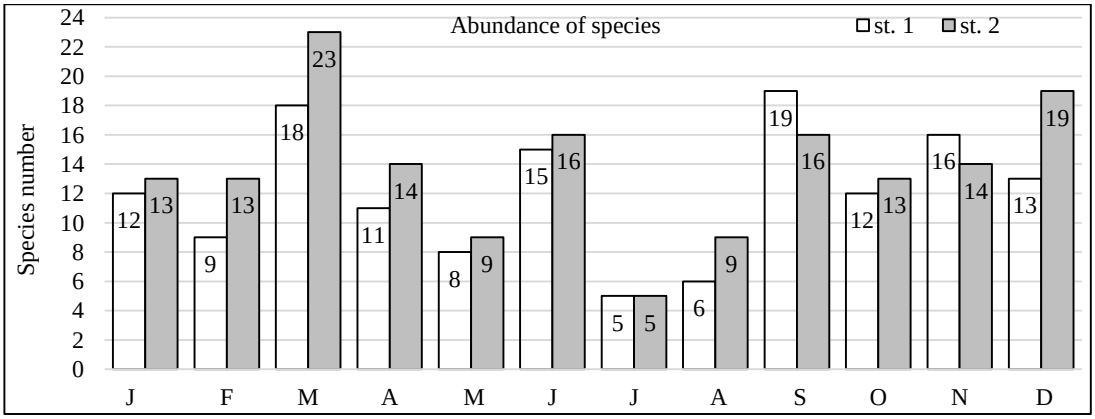

Figure 4. The species number of first and second stations

The species found in the study every month in different abundance were *Asplanchna priodonta*, *Polyarthra dolichoptera*, *Bosmina longirostris*, *Ceriodaphnia pulchella* and *Cyclops vicinus. Trichocerca similis* 11 months, *Daphnia longispina* and *Diacyclops bicuspidatus* 10 months, *Diaphanosoma birgei* 8 months were found (Table 2). On the other hand, the species found only once were *Hexarthra oxyuris*, *Anuraeopsis fissa*, *Brachionus quadridentatus*, *Colurella adriatica*, *Lepadella acuminata*, *Lophocharis salpina*, *Trichotria tetractis*, *Alona quadrangularis*, *Chydorus sphaericus*, *Monospilus dispar*, *Leydigia leydigi*, *Leptodora kindtii*, *Mesocyclops leukarti*, *Paracyclops fimbriatus* and *Nitocra hibernica* (Table 2).

Total Rotifera was the most abundant in December (10.099 ind./m^3), followed by January (9.603 ind./m^3) and March (4.636 ind./m^3). The amount of total Cladocera was the highest in January (4928 ind./m^3), followed by February (4.530 ind./m^3) and May (3.547 ind./m^3). The abundance of copepod was lesser than the other two groups and the most in September (1.091 ind./m^3), then in June (1.070 ind./m^3) and July (1.042 ind./m^3). Rotifera and Cladocera were found the least in July (316 ind./m^3, 421 ind./m^3), but Copepoda was found the least in October (258 ind./m^3) (Figure 3).

The most species were found at station 2 (23 species) in March, followed by 19 species in September (1st station) and December (2nd station). The least species were found in both first two stations in July (5 species) (Figure 4).

Discussion

The physicochemical parameters and zooplankton communities together form a comprehensive ecosystem and as in any ecosystem, there is interaction between the zooplankton and also between the phytoplankton and the water quality parameters. These interactions are directly or indirectly subjected to the complex influences, some of which results in quantitative changes (Welch 1952).

Determined water quality parameters, for animals in water are observed to be within the normal values. According to this, water temperature values (7.97-24.57 °C) detected in the study generally reflect the climatic conditions of the region and they are ideal for zooplankton life and development.

Mean dissolved oxygen concentrations were above 5 mg/l (5.87-8.20 mg/l) which was enough to support aquatic life, especially the zooplankton community (Karpowicz and Ejsmont-Karabin 2017).

The pH range in this study was 6.75-9.42, which was consistent with the reports of Blouin (1989), Beklioglu and Moss (1995). According to these researchers the distribution of plankton species in lakes with pH levels of 3.5–7.6, Beklioglu and Moss (1995) noted that plankton never occurred at low and high pH values (pH <4.6 and 11<).

Since chlorophyll *a* values were found to be quite low (1.71-7.39 mg/m^3), dam lake was in oligo-mesotrophic character, according to Wetzel (1975).

All inorganic forms of nitrogen (NO_3^-, NO_2^- and NH_4^+) can be used by aquatic plants and algae (Tepe and Boyd 2002). If these inorganic forms of nitrogen exceed 0.3 mg/l (as N) in spring, it means there is enough nitrogen to support summer algal blooms. The concentrations of nitrogen forms in Kozan Dam Lake were enough to support algae blooms and indirectly zooplankton biomass.

The quality of reservoir waters generally varied between clean water and much polluted water throughout the year in terms of nitrite values (YSKY 2012). As the nitrate nitrogen values determined in the study were below 10 mg/l, thus the reservoir waters were in the category of clean and less polluted water. The amount of ammonium nitrogen in the water samples was 0.056 - 0.187 mg/l. According to the Regulation on Surface Water Quality (YSKY), these values showed that, dam lake waters are classified as second class polluted waters.

Orthophosphate values changed between 0.01 mg/l and 0.053 mg/l and the reservoir waters generally have the first-class clean water and the second-class polluted water in terms of phosphate according to the YSKY (2012).

As a result, according to the Regulation on Surface Water Quality, reservoir water was first class water in point of temperature, dissolved oxygen, NH_4-N, PO_4-P, second class water in point of conductivity, pH, NO_3-N and third class water in point of NO_2-N (YSKY 2012).

In terms of aquatic organisms, the acceptable electrical conductivity value was reported to be 250-500 µS/cm by Yücel (1990). The lowest conductivity of the study was determined as 372.1 µS/cm, the highest 436.1 µS/cm, and accordingly, the dam lake was among the acceptable values for the aquatic organisms.

A total of 50 zooplankton species were detected, including 29 from rotifers, 15 from cladocerans and 6 from copepods. Twenty-two zooplankton species were previously reported in a study conducted in Kozan Dam Lake (Bozkurt 2004b). Some of the species *Collotheca ornata* (Ehrenberg 1832), *Cyclops abyssorum* Sars, 1863, *Acanthodiaptomus denticornis* (Werzesski 1887) and

Craspedacusta sowerby (Lankester 1880) were not found in the present study. On the other hand, 33 of the 50 zooplankton species in the present study were not reported in the previous study. It is thought that the difference of species in the two studies may depend on the number of sampling and the time difference between studies.

Rotifera was the dominant group followed by Cladocera and Copepoda among zooplankton groups qualitatively and quantitatively in Kozan Dam Lake as in all freshwater ecosystems (Saksena 1987).

It is reported that most of the zooplankton species found in the study are widespread in water bodies of all sizes in different geographic regions, with different types of substrates and vegetation-related species (Hutchinson 1967; Ruttner-Kolisko 1974; Braioni and Gelmini 1983; Ryding and Rast 1989; Ramdani et al. 2001; Eldredge and Evenhuis 2003). They were widespread in Turkey and worldwide because they were found in almost all regions of Turkey (Güher 2000; Alper et al. 2007; Dirican and Musul 2008; Saler and İpek 2009; Yıldız et al. 2010; Günsel and Emir Akbulut 2012; Apaydın Yağcı 2013; Güher 2014; Saler and Alış 2014; Apaydın Yağcı et al. 2015; Güher and Çolak 2015; Ustaoğlu 2015; Gürel and Saler 2015) and they were reported from lots of study inland waters of Turkey (Ustaoğlu et al. 2004; Ustaoğlu 2015).

The species identified in Kozan Dam Lake have been reported in various studies in the region and in the vicinity (Table 3). According to this, *Bosmina longirostris* was reported from 23 different studies in the region. While *Cephalodella gibba* was reported in 21 studies, *Lecane lunaris* was reported in 20 studies. *Keratella cochlearis* in 19 studies, *Euchlanis dilatata* and *Lecane bulla* in 18 studies, *Colurella adriatica* and *K. quadrata* in 17 studies, *K. tecta* and *Alona rectangula* in 16 studies were reported. *Chydorus sphaericus* and *Diaphanosoma birgei* were found in 15 studies, at the same time *Lepadella ovalis*, *Polyarthra dolichoptera*, *Trichotria tetractis* and *Ceriodaphnia pulchella* found in 13 studies. In the region, species found in 12 different working areas *Notholca squamula* and *Cyclops vicinus*, but species found in 11 different areas *Ascomorpha ovalis*, *Asplanchna priodonta*, *Collotheca pelagica*, *Lophocharis salpina*, *Trichocerca similis* and *Moina micrura*. Other species, *Keratella tropica* and *Nitocra hibernica* (10), *Macrothrix laticornis* (9), *Brachionus quadridentatus*, *Daphnia longispina*, *Mesocyclops leukarti* and *Paracyclops fimbriatus* (8), *Pompholyx sulcata*, *Daphnia galeata Disparalona rostrata*, *Diacyclops bicuspidatus* and *Macrocyclops albidus* (7), *Collotheca mutabilis* (6), *Anuraeopsis fissa* (5), *Filinia terminalis*, *Hexarthra oxyuris*, *Lepadella acuminata*, *Trichocerca capucina*, *Leydigia leydigi* (4), *Synchaeta pectinata* (3), *Hexarthra intermedia*, *Rotaria rotatoria*, *Alona quadrangularis*, *Daphnia cucullata*, *Leptodora kindtii*, (2), *Monospilus dispar* (1) have been reported from less aquatic environment. It has also been reported that these species are found all or nearly all of the sampling periods (Bozkurt 1997; Bozkurt 2004a, 2004b; Bozkurt and Dural 2005; Bozkurt 2006; Bozkurt and Sagat 2008; Bozkurt et al. 2009; Bozkurt and Göksu 2010; Bozkurt and Güven 2010; Bozkurt and Tepe 2011; Ülgü and Bozkurt 2015; Bozkurt and Duysak 2016; Bozkurt 2016; Bozkurt and Aktaş 2016; Bozkurt 2017; Bozkurt and Genç 2018a, 2018b; Bozca and Bozkurt 2018; Bozkurt et al. 2018).

The presence of identified species in the study seems to be compatible with their ecological characters and distribution.

There were differences in the number and amount of zooplankton species in the first and second stations. According to field observations, this may be due to the water flow rate, water mix and depth differences. On the other hand, the significant and inverse relationship (R^2 = - 0.65) was found between the dissolved oxygen and the number of species in the 2nd station, while the low level of significance and the inverse relationship (R^2 = -0.33) were determined in the first station. Zooplankton are not directly related to the nutrient, but have an indirect relationship because nutrient affects the presence of phytoplankton or other forms of zooplankton's food (Khan 2003). Thus, zooplankton growth, development, population density and species diversity were affected by the abundance of nutrient. Similarly, in the second station, significant relationship (R^2 = 0.88, R^2 = 0.68) was determined between nitrite and species number, and chlorophyll *a* and species number, while the significance level in the first station was low (R^2 = 0.29, R^2 = 0.4031) relationship was determined. Our results revealed that the level of relationship between other parameters and species numbers was very low in the Kozan dam lake.

Table 3. Distribution of species in our study in the region, according to the studies conducted by various researchers

Species		Study area
A. fissa	5 area	5, 7, 9, 10, 13,
A. ovalis	11 "	3, 4, 5, 6, 9, 10, 14, 15, 16, 21, 24
A. priodonta	11 "	2, 4, 5, 6, 9, 10, 12, 14, 15, 20, 24
B. quadridentatus	8 "	5, 10, 11, 12, 13, 16, 18, 20
C. gibba	21 "	1, 2, 3, 4, 6, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 20, 21, 22, 23, 24
C. mutabilis	6 "	10, 13, 14, 15, 20, 24
C. pelagica	11 "	3, 6, 9, 10, 11, 14, 16, 17, 19, 20, 21
C. adriatica	17 "	1, 2, 3, 4, 6, 8, 9, 12, 13, 14, 15, 16, 17, 20, 21, 22, 23
E. dilatata	18 "	1, 2, 3, 4, 8, 10, 11, 12, 13, 14, 16, 17, 18, 20, 21, 22, 23, 24
F. terminalis	4 "	9, 10, 14, 18
H. intermedia	2 "	14, 18
H. oxyuris	4 "	13, 14, 17, 21
K. cochlearis	19 "	1, 2, 3, 4, 5, 6, 7, 9, 10, 11, 12, 13, 14, 15, 16, 17, 19, 20, 21
K. quadrata	17 "	1, 2, 6, 7, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 20, 21, 24
K. tecta	16 "	3, 4, 7, 9, 10, 11, 12, 14, 15, 17, 19, 20, 21, 22, 23, 24
K. tropica	10 "	7, 10, 11, 12, 15, 16, 17, 19, 20, 24
L. bulla	18 "	1, 3, 6, 7, 8, 10, 12, 13, 14, 16, 17, 18, 19, 20, 21, 22, 23, 24
L. lunaris	20 "	1, 2, 3, 4, 5, 6, 8, 9, 10, 12, 13, 14, 15, 17, 19, 20, 21, 22, 23, 24
L. acuminata	4 "	2, 3, 4, 17
L. ovalis	13 "	1, 2, 4, 5, 8, 10, 12, 13, 14, 20, 21, 22, 23
L. salpina	11 "	5, 6, 9, 10, 12, 13, 16, 17, 20, 21, 23
N. squamula	12 "	1, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 20
P. dolichoptera	13 "	10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 24
P. sulcata	7 "	7, 9, 11, 12, 14, 19, 20
R. rotatoria	2 "	20, 22
S. pectinata	3 "	8, 14, 20
T. similis	11 "	1, 2, 3, 5, 6, 9, 10, 14, 15, 21, 24
T. capucina	4 "	9, 14, 15, 20
T. tetractis	13 "	1, 4, 12, 13, 14, 15, 16, 17, 20, 21, 22, 23, 24
A. quadrangularis	2 "	9, 24
A. rectangula	16 "	3, 5, 6, 7, 9, 11, 12, 13, 14, 15, 16, 17, 18, 20, 21, 24
B. longirostris	23 "	1, 2, 3, 4, 5, 6, 7, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24
C. pulchella	13 "	2, 3, 5, 7, 9, 10, 11, 12, 13, 15, 19, 20, 24
C. sphaericus	15 "	1, 2, 4, 6, 7, 9, 11, 12, 13, 14, 15, 17, 18, 20, 24
D. longispina	8 "	5, 6, 9, 10, 14, 15, 20, 24
D. cucullata	2 "	15, 24
D. galeata	7 "	1, 3, 4, 14, 15, 20, 24
D. birgei	15 "	1, 2, 3, 4, 5, 6, 9, 10, 11, 14, 15, 17, 19, 20, 24
D. rostrata	7 "	4, 5, 9, 10, 14, 20, 24
L. kindtii	2 "	14, 15
L. leydigi	4 "	9, 10, 11, 14
M. laticornis	9 "	1, 6, 9, 10, 11, 12, 14, 15, 16
M. micrura	11 "	7, 10, 11, 12, 13, 14, 16, 17, 18, 19, 24
M. dispar	1 "	5
C. vicinus	12 "	4, 5, 6, 7, 9, 11, 14, 15, 17, 18, 19, 20
D. bicuspidatus	7 "	8, 9, 11, 12, 13, 15, 18
M. albidus	7 "	2, 8, 9, 12, 13, 15, 24
M. leukarti	8 "	8, 10, 11, 13, 16, 18, 19, 20
N. hibernica	10 "	1, 8, 9, 12, 13, 15, 18, 20, 23, 24
P. fimbriatus	8 "	8, 9, 10, 12, 13, 18, 21, 22

(1: Kasımbey Creek, Hatay, 2: Hoplar Creek, Hatay, 3: Yayladağı Dam Lake, Hatay, 4: Hisarcık Dam Lake, Hatay, 5: Guvecci Dam Lake, Hatay, 6: Gorentaş Dam Lake, Hatay, 7: Volcanic pond, Gaziantep, 8: Sarıseki Marshes, Hatay, 9: Kahramanmaraş, 10: Seyhan Dam, Adana, 11: Tahtaköprü Dam, Gaziantep, 12: Gölbaşı Lake, Hatay, 13: Gölkent Lake, Hatay, 14: Aslantaş Dam, Osmaniye, 15: Birecik Dam, Şanlıurfa, 16: Yenişehir Lake, Hatay, 17: Topboğazı Dam, Hatay, 18: Yarseli Dam, Hatay, 19: Yagızlar Dam, Adana, 20: Ceyhan River, Adana, 21: Keşiş River, Osmaniye, 22: Savrun Stream, Osmaniye, 23: Deliçay Stream, Adan, 24. Manavgat River, Antalya).

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Landmark-Based Morphological Differences Among the Exotic *Rhinogobius lindbergi* and Its Two Sympatric Gobies (Actinopterygii: Perciformes: Gobiidae) in Sefid River, in the Southern Caspian Sea Basin

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ABSTRACT

Introduced species may cause harm to native fish populations, which to design any conservative program to control the exotics an identification key is necessary. To find the morphological differences among the exotic *Rhinogobius lindbergi* and its sympatric congeners including the endemic *Ponticla iranicus* and the native *Ponticla gorlap*, a 15-landmark morphometric system was used to examine 90 specimens in Sefid River, in the Southern Caspian Sea basin. Univariate analysis of variance showed significant differences among the means of the three groups for 79 out of 105 standardized morphometric measurements. Principal component analysis (*PCA*) and canonical variates analysis (*CVA*) confirmed the statistically significant difference among these species. The *CVA* scatter plot showed that the 90 studied specimens grouped into three distinct areas with a degree of overlap between *P. iranicus* and *P. gorlap*. Clustering based on Euclidean distances among the groups of centroids using an UPGMA indicated segregation of the three species into two distinct clusters: *P. iranicus* and *P. gorlap* in one group and *R. lindbergi* in the other group. The exotic *Rhinogobius* can be distinguished from the sympatric gobies in Sefid River by short snout (vs. longer), deep body (vs. shallow), deeper head, stout body, and smaller ventral disc.

Keywords: Gobiidae, truss network system, geometric morphometric, landmark, Iran

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Introduction

Invasive species are alien (non-native) organisms that have been introduced into an area outside of their natural range, establishing self-sustaining populations and spreading beyond their initial point of introduction, with deleterious impacts on the environment, economy and human health (Lymbery et al. 2014). Biological invasions are now considered a major environmental issue of public concern (Gozlan et al. 2010).

In Iran, there is an exotic goby, *Rhinogobius lindbergi*, which is reported from the Hari River basin (Coad and Abdoli 2000), Anzali Wetland in the Caspian Sea basin (Abdoli et al. 2000), the Zarrineh River (Zarrinehrud) in Urmia Lake basin (Eagderi and Moradi 2017), the Namak Lake basin (Eagderi et al. 2017), and the Tigris River basin (Sadeghi et al. 2018, Mousavi-Sabet et al. 2019). However, among the above-mentioned basins, the Caspian Sea basin is the only water basin which has native gobies, too.The genus *Ponticola*, with five species is the native genus which its members live in tributaries of the Caspian Sea basin (in Iranian portion). *Ponticola gorlap* (Iljin 1949), *P. syrman* (Nordmann 1840), *P. ratan* (Nordmann 1840), *P. cyrius* (Kessler 1874) and *P. iranicus* Vasil'eva, Mousavi-Sabet and Vasil'ev,

2015 (Vasil'eva et al. 1993; Miller 2003; Medvedev et al. 2013; Vasil'eva et al. 2015) are reported from the basin. Normally, the fishes of the family Gobiidae are usually less important commercially, therefore knowledge on their identification and biodiversity is far from being complete (Miller 2004; Neilson and Stepien 2009; Bogutskaya et al. 2013). Regarding problems in Gobiids identification, separating the exotic and native individuals/populations is not easy. Therefore, to design any conservative program, a practical identification key is strongly necessary. Preparing any key to identify fish species needs powerful analytical manners which the traditional and modern morphological analyzing methods are normally used for fish populations/species. As mentioned above, morphological characters are most important in the identification and taxonomy of fishes, and the only known facts about many fishes. In addition, understanding the function of a morphological structure is a stronghold for practical use in taxonomy and ecology too (Turan 2004; Yamamoto et al. 2006; Pollar et al. 2007). In addition to traditional method, in recent years, truss network system is increasingly used for morphometric measurements with the purpose of species and/or stock differentiation (Turan 2004; AnvariFar et al. 2011; Mousavi-Sabet et al. 2012; Mousavi-Sabet and Anvarifar 2013; Khataminejad et al. 2013; Kohestan-Eskandari et al. 2013, 2014; Heidari et al. 2014). Also, geometric morphometrics (*GM*), a quantitative approach to analysis shape, is widely applied to compare and determine shape variations of biological structures (Adams et al. 2004). Despite traditional approaches, in *GM*, data is obtained from the coordinates of landmark points (Adams et al. 2004; Rohlf 2005), which are morphological points of specimens that are of biological interest (Rohlf 2005).

The present study aimed to clarify the morphometric differences among the exotic *R. lindbergi* and its sympatric endemic *P. iranicus* and native *P. gorlap* in Sefid River, in the southern Caspian Sea basin.

Material and Methods

Fish sampling

The specimens were collected on August 2015 to May 2017 in Sefid River (37º01.153′N, 49º37.985′E) from the southwestern Caspian Sea basin, Iran (Figure 1). Fishes were collected by electrofishing. The sampled fishes were photographed and then fixed in 10% formaldehyde at the sampling site and transported to the Laboratory for further studies. Identification was verified base on Vasil'eva et al. 1993, Miller 2003 and Vasil'eva et al. 2015 in the laboratory.

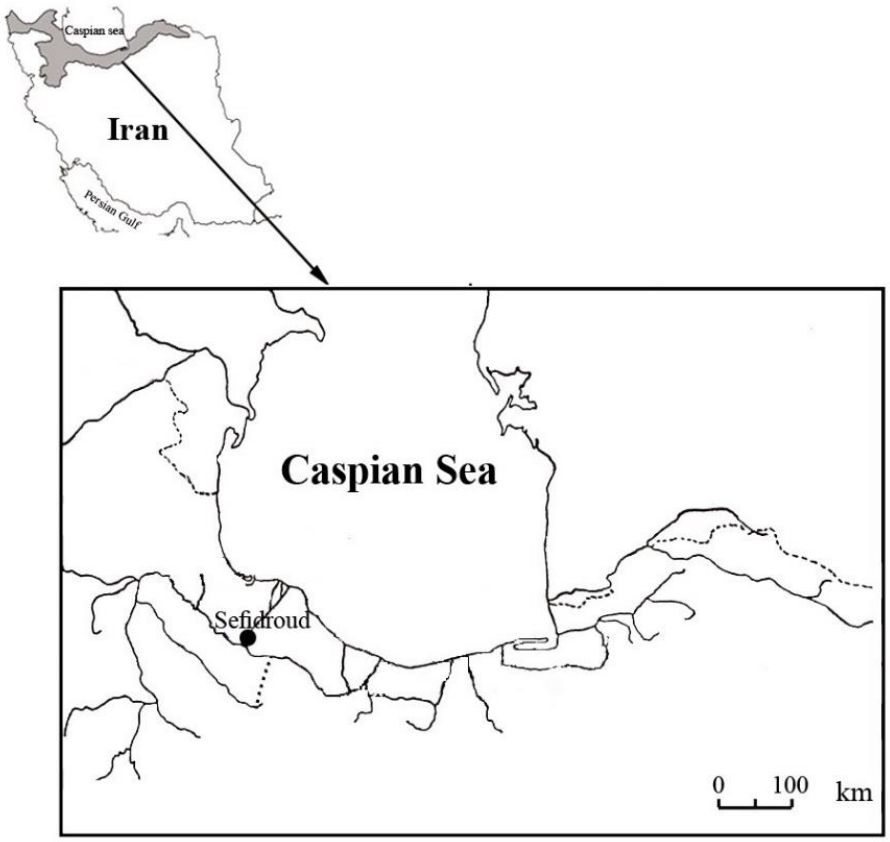

Figure 1. Map of the Iranian part of the southern Caspian Sea basin and location of Sefid River from the southern Caspian Sea basin.

Laboratory work

In the laboratory, Standard Length (±1.0 mm) and body weight (±0.001 g) were recorded for each specimen. For specimens 105 distance measurements between 15 landmarks on the lateral side and 78 distance measurements were surveyed using the truss network system according to Bookstein (1991) with minor modifications for these species (Figure 2). Also, for specimens 44 and 28 distance measurements between 10 and 8 landmarks on ventral and dorsal views, respectively, were surveyed for investigation under and up sides shape variation. The fishes were placed on a white board with dorsal and anal fins held erect by pins. The left body profile of each fish was photographed with a 300-dpi, 32-bit color digital camera (Cybershot DSC-F505; Sony, Japan). Images were saved in jpg format and analyzed with TPSdig to coordinates of landmarks. All measurements transformed into linear distances by computer for subsequent analysis. After image capture, the fish was dissected to identify the sex of the specimen by macroscopic examination of the gonads. Gender was used as the class variable in ANOVA to test for the significant differences in the morphometric characters if any, between male and female specimens.

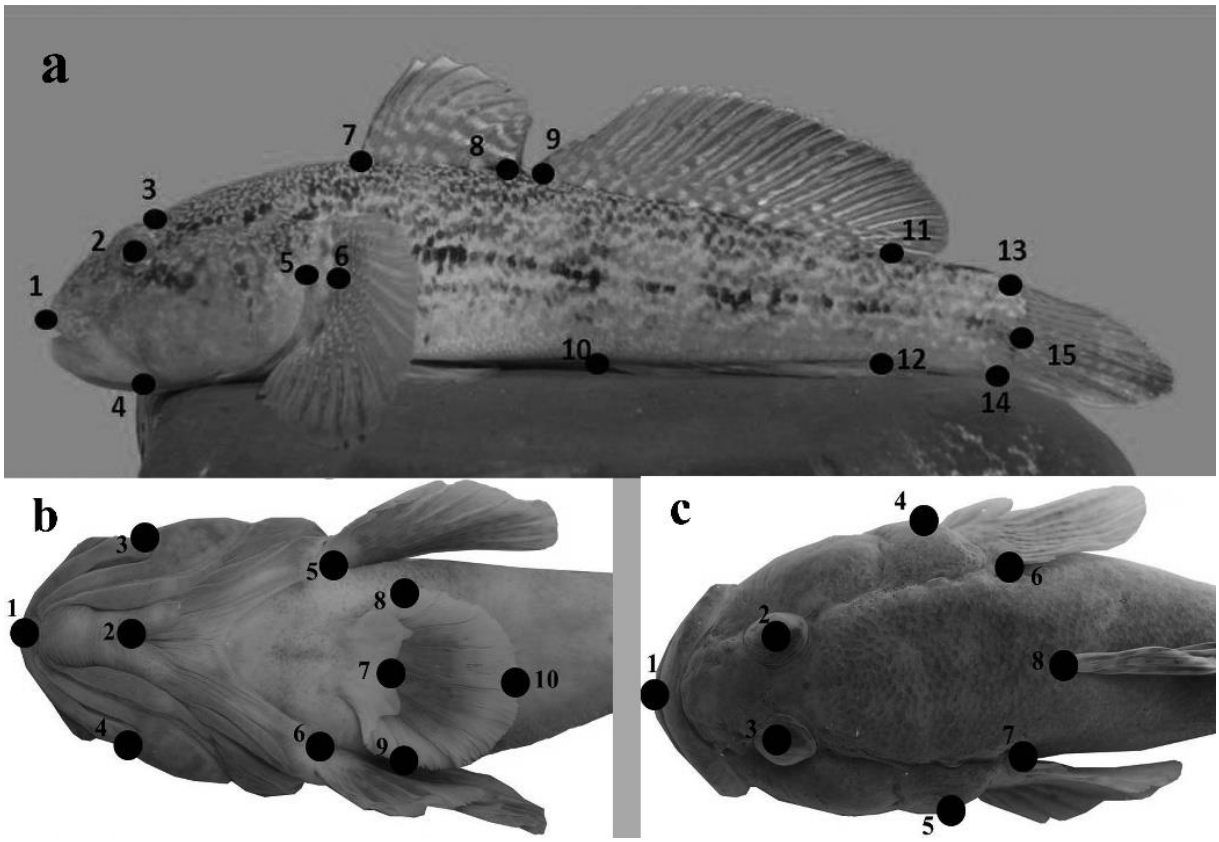

Figure 2. Locations of the landmarks on lateral (a), ventral (b) and dorsal (c) sides of the goby.

Data analysis

As variation should be attributed to body shape differences, and not related to the relative size of the fish, an allometric method (Elliott et al. 1995) was used to remove size-dependent variation in morphometric characters:

$$Madj = M\,(L_s / L_0)^b$$

where M is the original measurement, $Madj$ the size adjusted measurement, L_0 the standard length of the fish, L_s the overall mean of the standard length for all fish from all samples in each analysis, and b was estimated for each character from the observed data as the slope of the regression of log M on log L_0 using all fish in any group. The results derived from the allometric method were confirmed by testing the significance of the correlation between transformed variables and standard length (Mustafic et al. 2008).

Univariate analysis of variance (ANOVA) was performed for each morphometric character to evaluate the significant difference among the fish specimens (Bookstein 1991). Principal component analysis (*PCA*) and canonical variates analysis (*CVA*) were employed to discriminate these fishes from studied rivers. Statistical analyses for morphometric data were performed using the SPSS version 16 software package, past ver. 1.36, MorphoJ and Excel (Microsoft Office 2010).

Results

Descriptive data were examined for the mean and standard deviation (*SD*) of length and weight in case

of sampled specimens (Table 1). Although it is well known that the female and male specimens of the gobies have some morphological differences (Mousavi-Sabet et al. 2012; Vasil′eva et al. 2015), but the interaction between morphometric measurements used in this study by truss network system and sexes uses ANOVA analysis was not significant (P > 0.05), demonstrating a negligible effect of sex on observed variations; hence, the data for both sexes were pooled for all subsequent analyses. There was no significant correlation between any of the transformed measured morphometric variables and standard length (P > 0.05), indicating that size or allometric signature on the basic morphological data was accounted.

Table 1. Descriptive data [mean ± SD standard length (mm) and body weight (g)] of goby specimens.

Species	n	Standard length (mm)		Body weight (g)	
		Min–max	Mean ± SD	Min–Max	Mean ± SD
R. lindbergi	30	35.71-112.87	70.55±21.85	1.15-34.90	10.25±9.30
P. iranicus	30	53.33-139.92	74.23±23.64	3.77-51.61	12.86±11.85
P. gorlap	30	20.80-111.95	55.30±20.80	1.00-33.05	5.81±8.11

Table 2. The results of ANOVA for morphometric measurements of goby specimens in Sefid River from the southern Caspian Sea basin.

Characters	F	P	Characters	F	P	Characters	F	P	Characters	F	P
1-2	11.75	0.00	3-4	2.22	0.09	5-10	1.41	0.24	8-13	9.10	0.00
1-3	15.13	0.00	3-5	0.28	0.84	5-11	2.49	0.06	8-14	8.92	0.00
1-4	6.19	0.00	3-6	2.61	0.05	5-12	4.14	0.00	8-15	12.88	0.00
1-5	11.20	0.00	3-7	4.22	0.00	5-13	9.28	0.00	9-10	23.10	0.00
1-6	9.66	0.00	3-8	3.15	0.03	5-14	4.11	0.00	9-11	4.51	0.00
1-7	17.67	0.00	3-9	3.33	0.02	5-15	8.44	0.00	9-12	4.09	0.00
1-8	18.66	0.00	3-10	5.08	0.00	6-7	1.79	0.15	9-13	7.72	0.00
1-9	5.79	0.00	3-11	5.54	0.00	6-8	2.58	0.07	9-14	6.35	0.00
1-10	9.46	0.00	3-12	13.98	0.00	6-9	4.60	0.00	9-15	9.77	0.00
1-11	11.26	0.00	3-13	8.46	0.00	6-10	0.59	0.62	10-11	0.22	0.88
1-12	23.86	0.00	3-14	11.94	0.00	6-11	1.80	0.15	10-12	8.29	0.00
1-13	7.02	0.00	3-15	8.72	0.00	6-12	0.99	0.40	10-13	7.65	0.00
1-14	8.40	0.00	4-5	10.95	0.00	6-13	14.05	0.00	10-14	2.80	0.04
1-15	5.30	0.00	4-6	9.92	0.00	6-14	7.77	0.00	10-15	9.37	0.00
2-3	8.76	0.00	4-7	4.92	0.00	6-15	12.78	0.00	11-12	16.89	0.00
2-4	3.56	0.02	4-8	4.27	0.00	7-8	0.89	0.45	11-13	10.85	0.00
2-5	4.85	0.00	4-9	1.03	0.38	7-9	7.28	0.00	11-14	4.80	0.00
2-6	6.17	0.00	4-10	4.71	0.00	7-10	13.71	0.00	11-15	19.42	0.00
2-7	9.57	0.00	4-11	6.94	0.00	7-11	2.91	0.04	12-13	16.96	0.00
2-8	4.99	0.00	4-12	13.23	0.00	7-12	1.13	0.34	12-14	9.06	0.00
2-9	2.21	0.09	4-13	0.67	0.57	7-13	10.02	0.00	12-15	18.95	0.00
2-10	4.44	0.00	4-14	0.76	0.52	7-14	11.32	0.00	13-14	1.81	0.15
2-11	3.33	0.02	4-15	0.38	0.77	7-15	20.63	0.00	13-15	1.42	0.24
2-12	20.26	0.00	5-6	14.46	0.00	8-9	2.30	0.08	14-15	7.78	0.00
2-13	7.70	0.00	5-7	1.34	0.26	8-10	19.13	0.00			
2-14	2.12	0.10	5-8	1.48	0.22	8-11	7.15	0.00			
2-15	1.97	0.12	5-9	2.46	0.07	8-12	3.74	0.02			

Statistically significant differences in goby specimens were observed in 79 morphometric characters out of 105 studied. Of these 79 characters, 72 characters were found to be highly significant (P≤0.01) and were used further for multivariate analysis (Table 2). The value of Kaiser-Meier-Olkin coefficient (*KMO*) for the overall matrix is 0.721 for goby specimens and Bartlett's Test of sphericity is significant (P≤0.01). The results of *KMO* and Bartlett's suggest that the sampled data is appropriate to proceed with a factor analysis procedure.

In order to determine which morphometric measurement affected populations differentiates mostly, the contributions of variables to principal components (*PC*) were examined. The *PCA* of 72 morphometric measurements for goby specimens extracted 12 factors with eigenvalues >1, explaining 93.91% of the variance (Table 3). Of these, the first explained 28.37% and the second 16.35% of the variance. The most significant weightings on *PC* I were from 1-5, 1-6, 5-15, 2-6, 2-5, 6-15, 6-9, 4-6, 4-5, 6-13, 6-14, 5-13, 1-14, 5-14, 1-4, 1-13, 1-11, 1-12,

1-2, 5-12, 1-10, 1-8, 1-3, 1-7, 1-9 and on *PC* II were from 1-12, 12-15, 9-12, 12-13, 12-14, 3-12, 2-12, 10-12, 5-12, 4-12 and 3-14.

Canonical variates analysis confirmed the significant difference among the goby specimens. The scores of the two canonical variables for each river revealed that goby specimens grouped into three species while there was a relativity high degree of overlap between *P. iranicus* and *P. gorlap* (Figure 3).

Table 3. Eigen values, percentage of variance and percentage of the cumulative variance of morphometric measurements for goby specimens in Sefid River from the southern Caspian Sea basin.

Factor	Eigenvalues	Percentage of variance	Percentage of cumulative variance
1	20.141	28.367	28.367
2	11.606	16.347	44.714
3	8.061	11.354	56.068
4	6.906	9.727	65.795
5	4.550	6.408	72.203
6	4.242	5.974	78.178
7	2.754	3.879	82.056
8	2.447	3.447	85.503
9	2.097	2.953	88.456
10	1.478	2.082	90.538
11	1.350	1.901	92.439
12	1.041	1.466	93.905

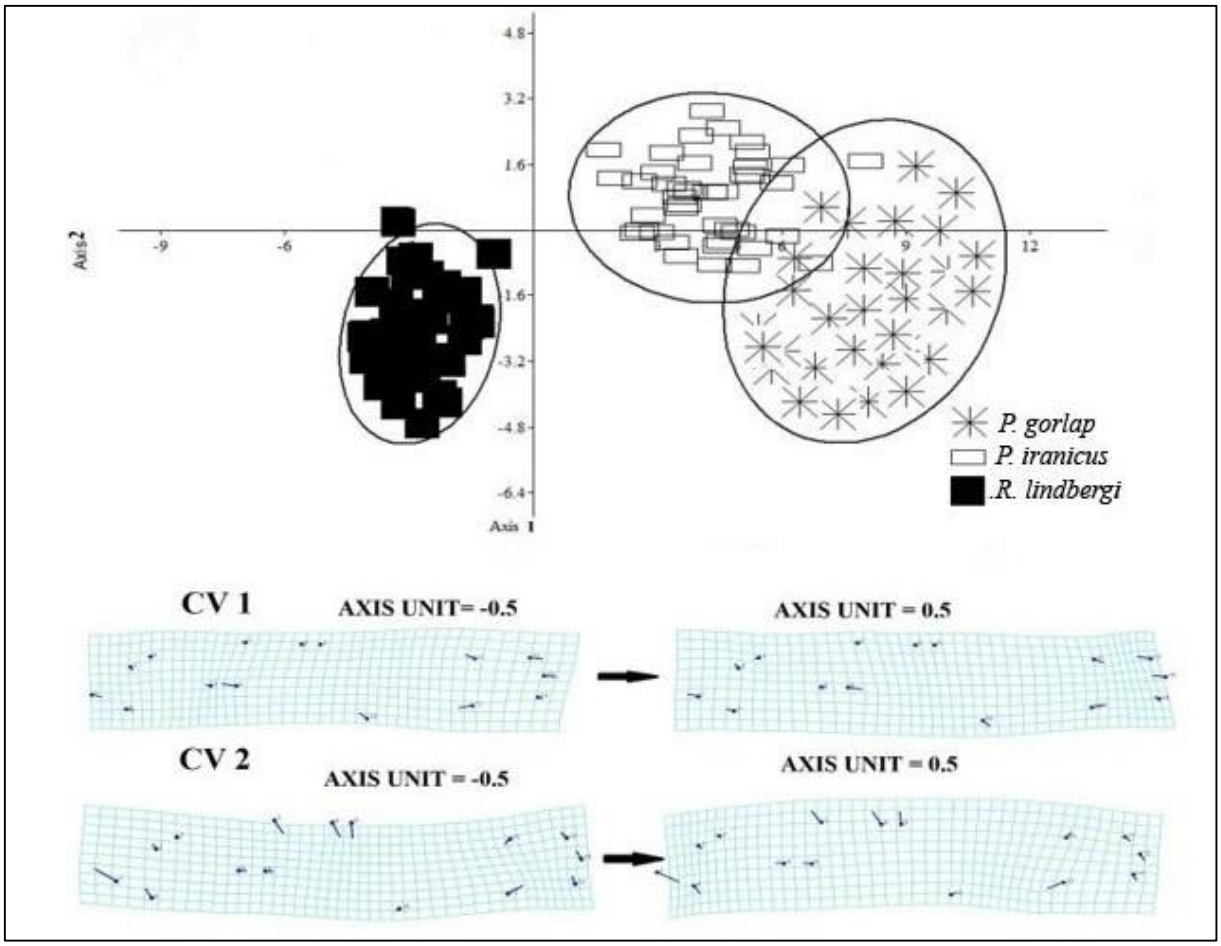

Figure 3. Scatterplot of standardized canonical variates functions 1 (CV1) and 2 (CV2) for morphometric characteristics of goby specimens

The Wilks' lambda tests indicated differences between fishes when their morphometric measurements were compared by means of discriminant analysis. In this test all functions were highly significant ($P \leq 0.01$). The histogram of discriminant functions for pairwise groups in goby specimens is shown in Figure 4.

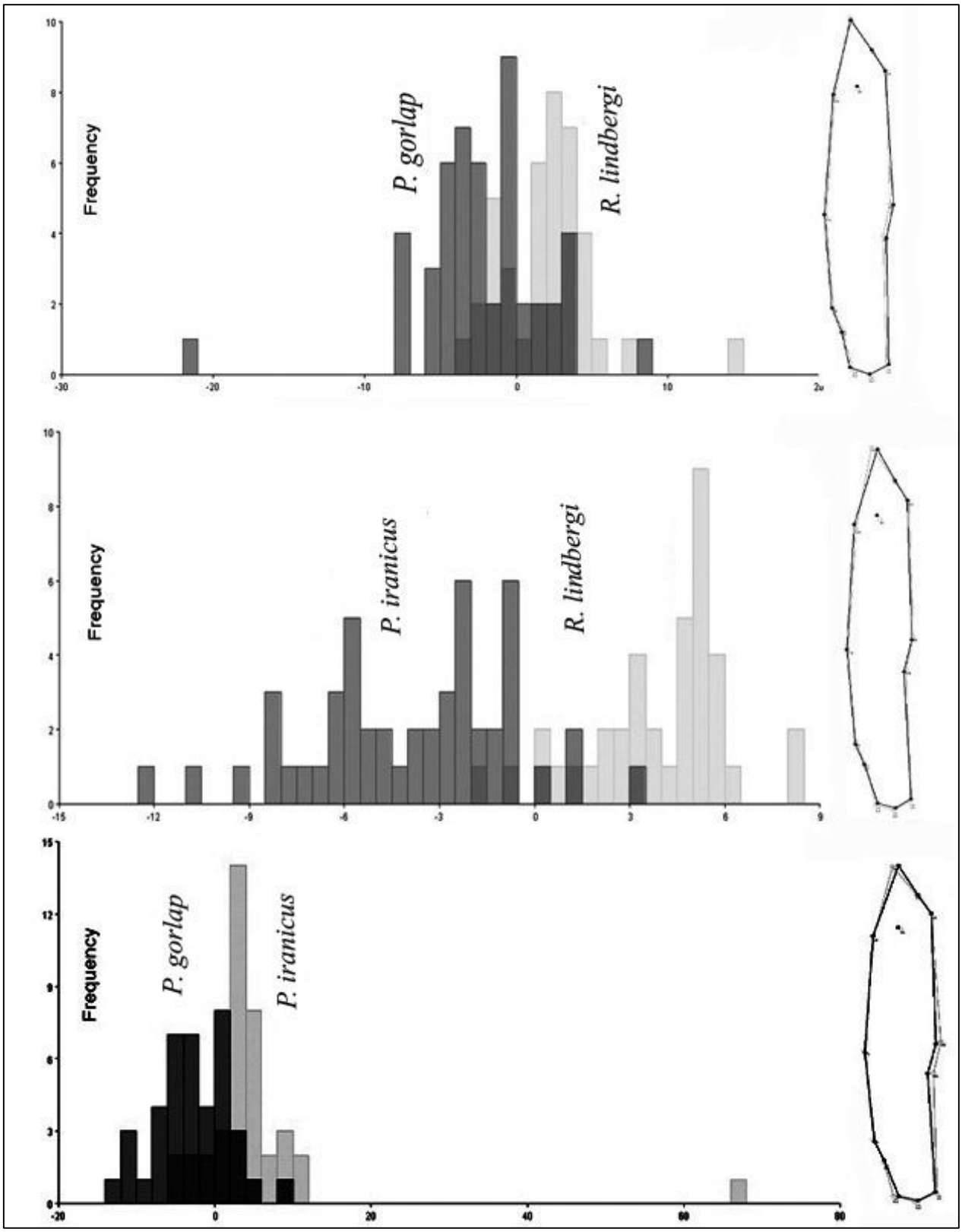

Figure 4. Histogram of discriminate analysis (DA) functions for pair wise competitions' of gobies (left). Shape differences on the extremities of each river (right).

Body shape differences show a longer snout, shallow body and head depths, and elongated body for *P. gorlap* and *P. iranicus*, vs. relatively short snout, high body and head depths and stout body for *R. lindbergi***.** Also, underside and upside shape variation analysis show a longer snout and disc for *P. gorlap* and *P. iranicus*, vs. relatively short snout and disc and high body for *R. lindbergi* (Figure 5).

The dendrogram derived from the cluster analysis of Euclidean distances among groups

of centroids showed that the three populations of gobies segregated from each other into two distinct clusters, *P. gorlap* and *P. iranicus* appeared in one cluster while *R. lindbergi* belonged to the other clusters (Figure 6).

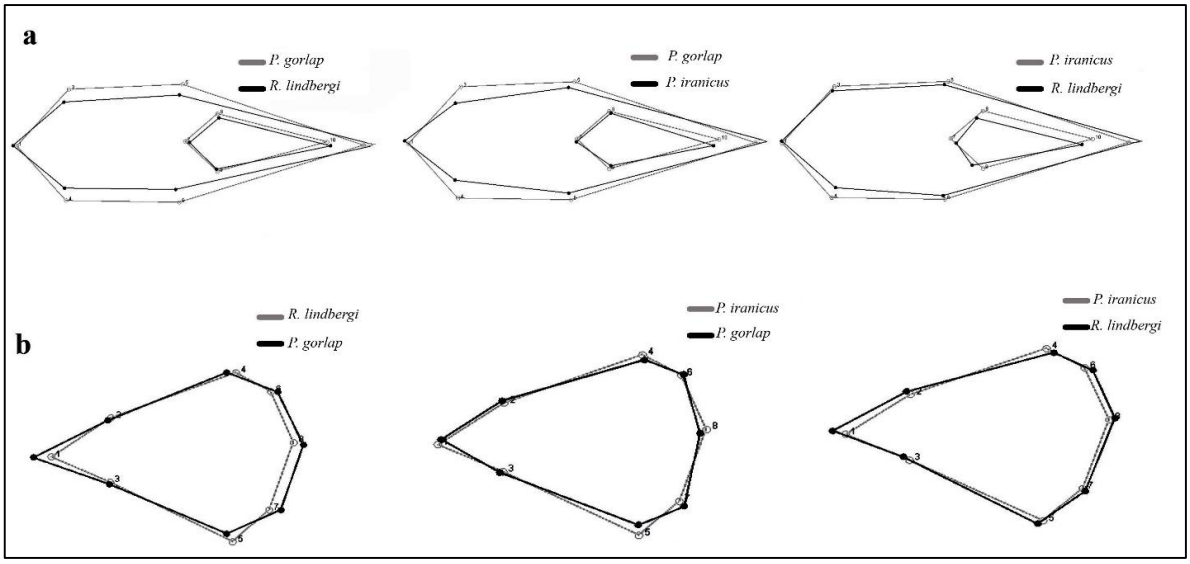

Figure 5. Underside and upside shape variation analysis of gobies in Sefid River from the southern Caspian Sea basin. (a) Ventral view (b) dorsal view.

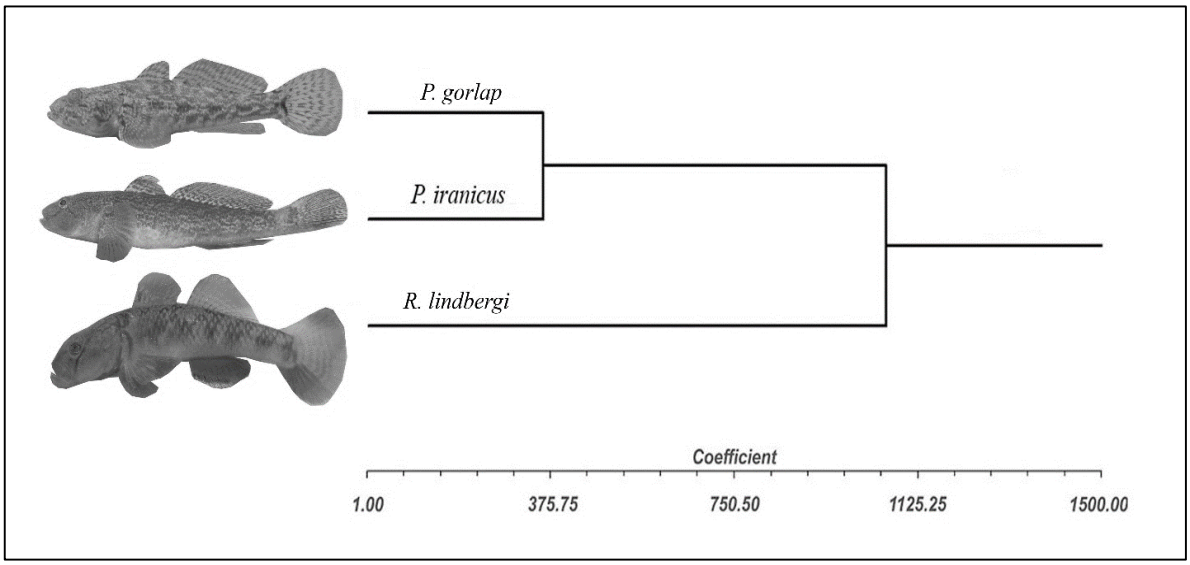

Figure 6. Dendrogram derived from cluster analyses of morphometric measurements on the basis of Euclidean distance for gobies in Sefid River from the Southern Caspian Sea. Mean shape of species in the relation of consensus shape of the species are also represented.

Discussion

The results of the multivariate analysis demonstrated that the three examined species are correctly separated from each other, which the two *Ponticola* (*P. iranicus* and *P. gorlap*) are classified as one clade and the *Rhinogobius* in the other clade. It is well known that both studied genera, *Rhinogobius* and *Ponticola*, are clearly distinct groups morphologically, and as the *Ponticola* are larger species than the *Rhinogobius*, they can be easily distinguished if the *Ponticola* is in its maximum range size. Despite these differences, separating small *Ponticola* specimens in the same size as *Rhinogobius* is not so easy or sometimes is unrecognizable by morphological characters. Therefore, the obtained results of the present study provide helpful identification key to separate them from each other. The results for the ANOVA analysis showed that 79 out of 105 transformed morphometric measurements were significantly different in these groups of Gobies living in the southern Caspian Sea basin, which demonstrates a high phenotypic variation among them. The *PCA* showed morphological segregation of the studied fishes based

on the characters head shape, pre-dorsal, pre-anal distances, pre-orbital and post-orbital distances, body length, body depth, caudal peduncle depth, caudal peduncle length, dorsal and anal fins origin and caudal fin origin for gobies. Body shape differences for these fishes shows a longer snout, shallow body and head, and elongated body for *P. iranicus* and *P. gorlap*, vs. relatively short snout, deeper body and head and stout body for *R. lindbergi*, for general appearance see Figure 7.

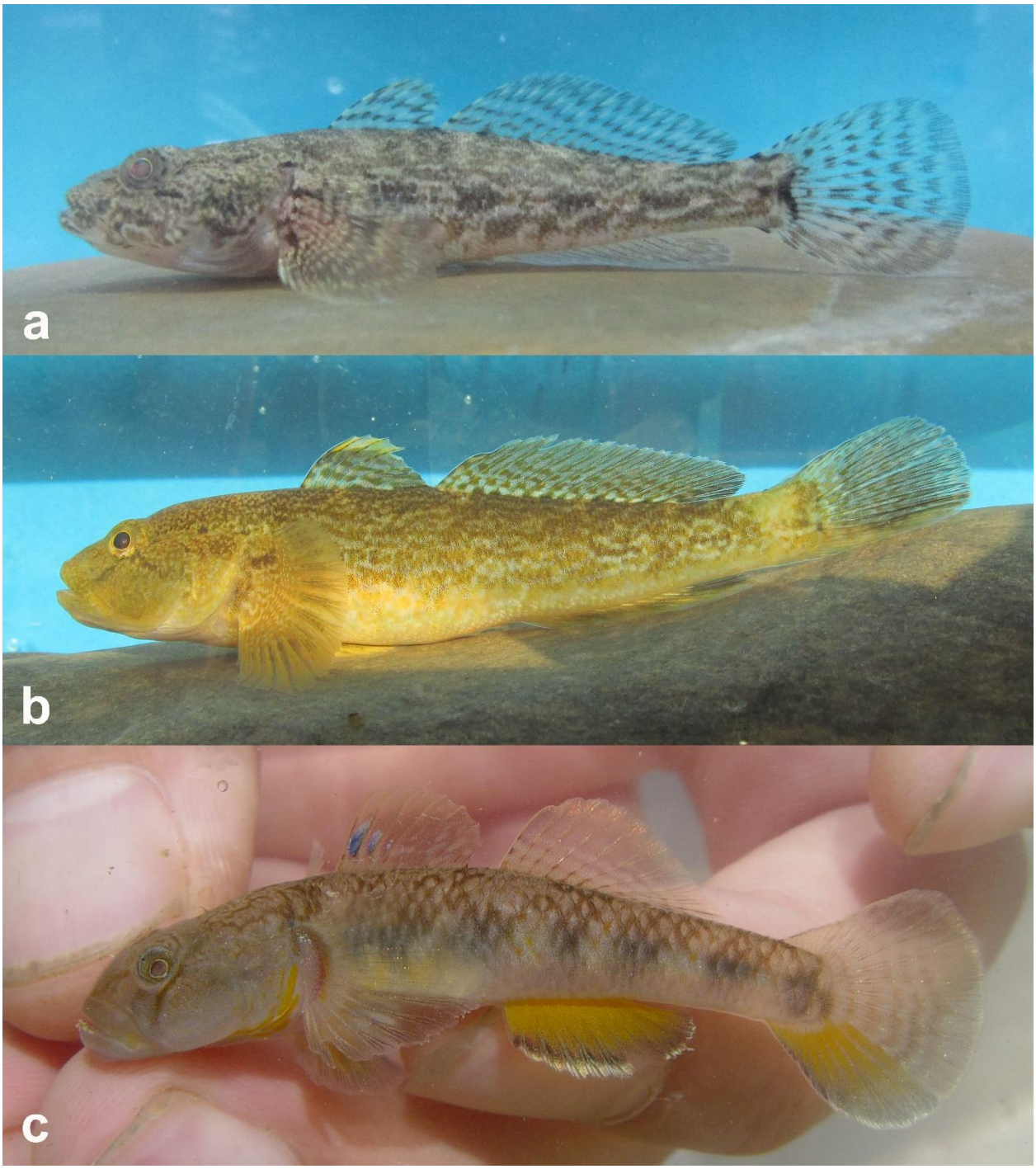

Figure 7. *Ponticola gorlap* (a), *Ponticola iranicus* (b) and *Rhinogobius lindbergi* (c).

CVA could be a useful method to distinguish different species of the same genus or different stocks of the same species, with respect to stock management programs (Bookstein 1991; Buj et al. 2008; Torres et al. 2010; AnvariFar et al. 2011; Heidari et al. 2013; Heidari et al. 2014). The results of *CVA* obtained in the present study indicated the relative segregation among three species of gobies in the studied areas. This was confirmed by plotted *CV*1 and *CV*2 scores for each sample that showed 90 specimens grouped into three distinct areas with a degree of overlap between *P. iranicus* and *P. gorlap*. The morphological differences may be solely related to body shape variation and not to size effects which

were successfully accounted for by allometric transformation. Literature shows that factor of size account more than 80 % of variation among a set of variables in morphometric studies. On the other hand, factor of size plays a predominant role in morphometric analysis and makes result in erroneous status if it cannot be removed in statistical analyses of data (Bookstein 1991). In the present study, size effect was removed successfully by the allometric transformation, so any significant differences indicated by the ANOVA and multivariate analysis, are caused by the body shape variation.

The causes of morphological differences between species are often quite difficult to explain (McLaughlin and Grant 1994; Cadrin and Friedland 1999; Pakkasmaa and Piironen 2000; Cadrin 2000; Vatandoust et al. 2014a; Vatandoust et al. 2014b; Vatandoust et al. 2015; Paknejad et al. 2014). It has been suggested that the morphological characteristics of fish are determined by an interaction between genetic and environmental factors (Pinheiro et al. 2005). The phenotypic variability may not necessarily reflect population differentiation at the molecular level (Turan 2004; Yamamoto et al. 2006; Pollar et al. 2007; Eschmeyer and Fong 2011).

The present findings revealed the potential power of the landmark-based methods for the identification of goby specimens. The present study provides basic information about the differences of *Ponticola* species and the exotic *R. lindbergi* in Sefid River from the southern Caspian Sea basin and it suggests that observed morphological variation should be considered in stock management programs and commercial exploitation of these species as an ornamental fish in the aquarium trade.

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Gambusia holbrooki (Sivrisinek balığı)'nin Türkiye'deki Dağılımına Katkılar

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ÖZ

Bu çalışmada, dünyanın en istilacı 100 canlı türünden birisi olduğu ifade edilen ve özellikle endemik balık türlerinin varlığı açısından risk oluşturan sivrisinek balığı, *Gambusia holbrooki* türünün Türkiye'deki dağılımına katkılar sağlanması amaçlanmıştır. Çalışma süresince Türkiye'nin 6 farklı coğrafik bölgesinde bulunan su kaynakları arasından toplam 130 lokalitede çalışılmıştır. Çalışma sonucunda 39'u lotik, 28'i lentik su kaynağı olmak üzere toplam 67 lokalitede *G. holbrooki* türünün varlığı tespit edilmiştir.

Anahtar kelimeler: Sivrisinek balıkları, yayılım, istilacı tür, endemik tür, Anadolu

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Contribution on the Distribution of *Gambusia holbrooki* (Mosquitofish) in Turkey

Abstract: In this study, it was aimed to contribute the knowledge on the distribution of mosquitofish *Gambusia holbrooki*, which is known one of the 100 invasive species of the earth and pose a risk to the existence of endemic species, in Turkey. During the study period, total of 130 localities from six different geographic regions in Turkey were investigated. The presence of *G. holbrooki* were determined in 67 localities (39 lotic and 28 lentic).

Keywords: Mosquitofishes, spread, invasive species, endemic species, Anatolia.

Alıntılama
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Giriş

Kuzey Amerika kökenli olup, tüm dünyada sivrisinek balıkları olarak bilinen *Gambusia affinis* ve *G. holbrooki* türleri, sıtma hastalığına karşı yürütülen mücadelede biyolojik ajan olarak kullanılmak üzere, dünyanın pek çok ülkesine 1900'lü yıllarda aşılanmaya başlamıştır (Courtenay ve Meffe 1989; Walters ve Freeman 2000). Amerika kıtasında Teksas'tan Hawaii'ye yapılan aşılama çalışması, dünyada bilinen ilk resmî sivrisinek balığı aşılama çalışması olup, Avrupa kıtasındaki ilk aşılama çalışmaları 1920 yılında önce İspanya'da, ikinci olarak da İtalya'da gerçekleştirilmiştir (Walton vd. 2012).

Bu balıkların Türkiye sularına ilk aşılamalarının yine tüm dünyada olduğu gibi sıtma hastalığının vektörü sivrisineklere karşı biyolojik mücadele yürütme amaçlı olarak 1920-1929 yılları arasındaki bir tarihte yapıldığı ifade edilmektedir (Walton vd. 2012). Hatay ilinin Türkiye topraklarına henüz katılmamış olduğu bir dönemde, Fransızlar tarafından Avrupa ülkelerinden getirilerek Amik Gölü'ne aşılanan *G. affinis* (Geldiay ve Balık 1996), ilerleyen yıllarda Türkiye sularında hızla yayılmıştır. Sivrisinek balıkları, Türkiye sularına aşılanan ilk "*yabancı (egzotik)*" balık türüdür (İnnal ve Erk'akan 2006), ancak bu balıkların Türkiye'ye getiriliş tarih ve şekilleri ile ilgili herhangi bir resmî kayıt bulunamamıştır.

G. affinis ve *G. holbrooki* türlerinin 4-42°C gibi oldukça geniş bir sıcaklık aralığında yaşayabildiği ifade edilmekle birlikte, bu türlerin optimum su sıcaklığının 31-35°C arasında olduğu belirtilmiştir (Pyke 2005). Bu türler yaşam alanı olarak genellikle tatlı suları tercih ederler, ancak *G. holbrooki* türünün ‰23 tuzluluğa sahip sularda da popülasyon

oluşturduğu gözlenmiştir. Üremeleri tropikal bölgelerde yıl boyunca devam ederken, kışın sert olarak geçtiği bölgelerde sadece yaz aylarında gerçekleşmektedir. Özellikle yaşlı ve büyük dişiler bir üreme sezonu boyunca birden fazla kez doğum yapabilir (Pyke 2005). Canlı doğuran bu türlerin dişileri ortam şartları uygun olduğunda 3-4 haftada bir doğum yapar. Bu türlerin üremeleri erkek bireylerinin sahip olduğu kopulasyon organı sayesinde olur ve dişi bireyler erkekten gelen sperm hücrelerini döllenme zamanına dek canlı bir şekilde saklayabilir (Krumholz 1948).

Güncel çalışmalarda türün, birbirinden oldukça farklı karakterdeki sucul habitatlara uyum sağlama ve girdikleri ortamlarda çok kısa zamanda büyük popülasyonlar oluşturabilme yeteneğine sahip olduğu kanıtlanmıştır. Bununla birlikte bu türlerin girdikleri sucul ortamlarda meydana getirdikleri pek çok olumsuz etki net olarak ortaya koyulmuş ve bu türler yakın dönemde "*dünyanın en istilacı 100 canlı türü*" arasındaki yerini almıştır (ISSG 2013).

Türkiye sularında günümüzden 15 yıl öncesine kadar olan çalışmaların neredeyse tamamında *G. affinis* türü bildirilmiş olmakla beraber (Geldiay ve Balık 1996; Bahadıroğlu ve Büyükçapar 1997; Öztürk ve İkiz 2003), son dönem çalışmalarının önemli bir kısmında *G. holbrooki* türü rapor edilmiştir (Kuru 2004; Tarkan vd. 2006; Ergüden 2013; Özuluğ vd. 2013). Yaşanan bu durumun temel sebebi, *G. affinis* ve *G. holbrooki* türlerinin morfolojik ve anatomik açıdan birbirine çok benzer olmasıdır (Pyke 2005). Bunun yanı sıra, bu iki türün geçtiğimiz yüzyıla kadar *Gambusia affinis affinis* ve *Gambusia affinis holbrooki* olarak alt tür düzeyinde tanımlanmış olmaları da tür kayıtlarında yaşanan karışıklığın bir başka nedeni olarak karşımıza çıkmaktadır.

Sivrisinek balıklarının Türkiye'deki dağılımını ortaya koyan farklı amaçlarla gerçekleştirilmiş birçok çalışma bulunmaktadır. Bu çalışmaların bir kısmına kronolojik sıra çerçevesinde Tablo 1'de değinilmiştir.

Tablo 1. Önceki çalışmalarda Türkiye'den verilmiş sivrisinek balığı kayıtları.

Kayıt Yeri	Referans	Kayıt Yeri	Referans
Balık Gölü (Bafra)	Kuru (1975)	Taflan Deresi (Samsun)	Uğurlu ve Polat (2007)
Gelemen DÜÇK (Samsun)	Kuru (1975)	Yurtluk Çayı (Samsun)	Uğurlu ve Polat (2007)
Aras Nehri DÜÇK (Iğdır)	Kuru (1975)	Taflan Deresi (Samsun)	Uğurlu ve Polat (2007)
Çiğli Deresi (İzmir)	Balık (1979)	Dipsiz-Çine Deresi (Muğla)	Özcan (2008)
Büyük Menderes Nehri	Balık (1979)	Topçam Baraj Gölü (Aydın)	Özcan (2008)
Büyükçekmece G.(İstanbul)	Balık (1985)	Gölbaşı Göleti (Bursa)	İlhan ve Balık (2008)
Meriç Nehri (İpsala-Edirne)	Balık (1985)	Kumaşır (Kahramanmaraş)	Kara vd. (2010)
Pamuklu Gölü (Edirne)	Balık (1985)	Gavur Gölü (Kahramanmaraş)	Kara vd. (2010)
Köyceğiz Gölü (Muğla)	Balık (1988)	Korkun Deresi (Adana)	Erk'akan ve Özdemir (2011)
Aras Nehri (Erzurum)	Geldiay ve Balık (1996)	Karasu Deresi (Gaziantep)	Birecikligil ve Çiçek (2011)
Akgöl (İçel)	Anonim (1997)	Nizip Çayı (Gaziantep)	Birecikligil ve Çiçek (2011)
Büyükçekmece Gölü (İstanbul)	Özuluğ (1999)	Merzimen Deresi (Gaziantep)	Birecikligil ve Çiçek (2011)
Acısu (Antalya)	Küçük ve İkiz (2004)	Bafra Balık Gölleri (Samsun)	Öztürk vd. (2011)
Kargı Çayı (Antalya)	Küçük ve İkiz (2004)	Davutlar Mevkii (Osmaniye)	Erk'akan ve Özdemir (2011)
Kovada Kanalı (Antalya)	Küçük ve İkiz (2004)	Camizagili Deresi (Adana)	Erk'akan ve Özdemir (2011)
Dalaman Çayı (Muğla)	Öztürk ve İkiz (2004)	Ula Göleti (Muğla)	Önsoy vd. (2011)
Ortaca Mevkii (Muğla)	Öztürk ve İkiz (2004)	Köprüçay Deresi (Antalya)	İnnal (2012)
Ömerli Baraj Gölü (İstanbul)	Özuluğ vd. (2005b)	Büyük Menderes Nehri (Aydın)	Keskin vd. (2013)
Köyceğiz Gölü (Muğla)	Yılmaz vd. (2006)	Sarıçay Rezervuarı (Muğla)	Keskin vd. (2013)
Tersakan Deresi (Muğla)	Yılmaz vd. (2006)	Yuvacık Rezervuarı (Kocaeli)	Keskin vd. (2013)
Afyonkarahisar kaynakları	Yeğen vd. (2007)	Acıgöl K. (Afyonkarahisar)	Yoğurtçuoğlu (2016)

DÜÇ: Devlet Üretme Çiftliği Kanalları. K: Kaynakları.

Ege Üniversitesi Su Ürünleri Fakültesi Temel Bilimler Bölümü İçsular Biyolojisi Anabilim Dalı tarafından Türkiye'de yürütülmüş çeşitli projelerde *Gambusia* genusuna ait kayıtlar bulunmaktadır. Birimin türe ait ilk örneklemesi Prof. Dr. Remzi GELDİAY tarafından 1963 yılında Pınarbaşı'ndan (Bornova, İzmir) yapılmıştır (*Müze kodu*: ESFM-PISI/1963-012, *Tarihi*: 20.12.1963). Çeşitli proje çalışmalarında örneklenmiş olan türe ait bireyler Ege Üniversitesi Su Ürünleri Fakültesi Müzesi'nde saklanmaktadır (Sarı ve Balık 1995; Balık vd. 1996, 2002; Sarı vd. 1999a; Ustaoğlu vd. 2000; Balık vd. 2003; Sarı vd. 2004).

Materyal ve Metot

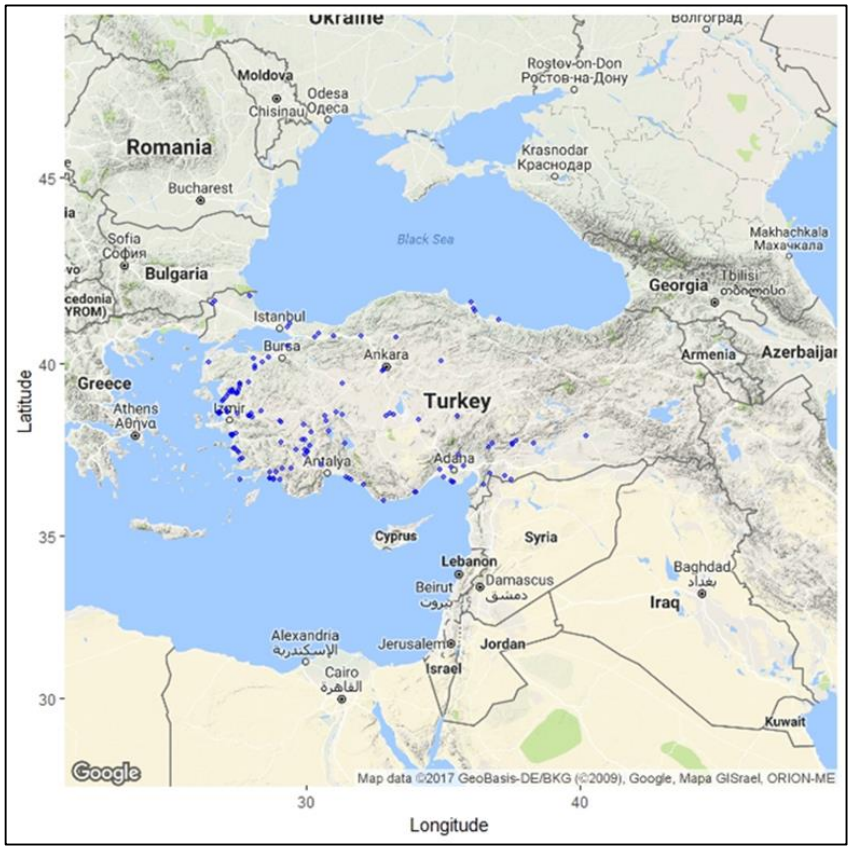
Türün yaşam toleransı çok yüksek olduğundan, arazi çalışmaları süresince çok çeşitli karakteristik özelliklere sahip su kaynakları incelenmiş, seçilen bu su kaynaklarının mümkün olduğunca farklı özelliklere sahip tipolojilerinden tesadüfi olarak örneklemeler yapılmıştır. Tüm örnekleme çalışmaları Nisan 2016 ile Kasım 2017 arasındaki tarihlerde tamamlanmıştır. Çalışma bir tarama çalışması

olduğundan hedeflenen lokalitelere sadece 1 kez gidilmiştir.

Örnekleme için arazi bölgesinin uygunluğuna göre kanatları 10 metre, torbası 5 metre, toplamda 25 metre uzunluğunda, 3 mm tor göz açıklığında tül ığrıp ya da 500 mikron göz açıklığı olan el kepçesi kullanılmıştır. Çalışmalar farklı mevsimlerde, farklı arazi şartlarında gerçekleştiğinden, çalışma süreleri lokalitelere göre değişkenlik göstermiş olup; hava durumuna ve lokalitenin elverişliliğine göre 30 dakika ile 4 saat arasında değişmiştir. Iğrıp ile yürütülen avcılıkta operasyon sayısı 3-5 arasında değişkenlik göstermiş iken; el kepçesi ile yürütülen operasyonlarda bu sayı 16-20 arasında değişmiştir. *Gambusia* türleri özel mikrohabitat tercihleri olan balıklar olduğundan türün bir su kaynağında varlık gösterip göstermediğinden emin olmak için tüm su kaynakları pek çok farklı noktası taranmıştır. Arazideki taramalar sırasında lentik lokalitelerde suyun mansap kısımlarından başlayarak kaynak kısımlarına doğru çıkılmış, lotik lokalitelerde ise kıyı boyunca mümkün olan mesafeler boyunca yürünmüştür. Türün habitat tercihleri açısından suyun durgunluğunun ve vejetasyonun önemi bilindiğinden, taranan su kaynağında su akışının oldukça yavaş olduğu ya da akışın hiç olmadığı noktalar seçilmiş; bununla birlikte vejetasyonun yoğun olarak bulunduğu noktalar özellikle kontrol edilmiştir. Hem lentik hem lotik lokalitelerde türün tespit edilemediğini ifade etmeden önce ilgili kaynağın tesadüfi olarak seçilen en az beş farklı noktası türün varlığı/yokluğu açısından kontrol edilmiştir.

Örneklenen bireyler yüksek dozda fenoksietanole (1 ml/L) tabi tutularak ötenazi edilmiştir. Ötenazi işleminden sonra *Gambusia* bireyleri %4'lük formaldehit solüsyonuna alınarak fikse edilmiştir. Vücut bütünlüğü bozulmayan bireyler Ege Üniversitesi Su Ürünleri Fakültesi Müzesi'nde kodlandırılıp muhafaza altına alınmıştır.

Arazi çalışmaları sırasında örneklerin toplandığı noktaların koordinatları ve rakımları GPS (Global Positioning System, Coğrafi Koordinat Sistemi) ile ondalık derece olarak ölçülmüştür. Bireysel olarak yapılan örnekleme çalışmaları dışında 2'si lotik 2'si lentik olmak üzere toplam 4 lokalite için diğer araştırıcılardan örnek temini yoluna gidilmiştir. Bu lokalitelerin 3'ü Güneydoğu Anadolu Bölgesi'nde, 1'i Ege Bölgesi'nde bulunmaktadır. Bu lokalitelerle birlikte incelenen lokalite sayısı lotik sular için 39, lentik sular için 28'e ulaşmıştır. Arazi çalışması gerçekleştirilen lokalitelerin tamamı Şekil 1'de gösterilmiştir (Harita R istatistik paket programı yardımı ile oluşturulmuştur; R-Development Core Team 2015).



Şekil 1. Türkiye'de *Gambusia* sp.'nin varlığının araştırıldığı lokaliteler (Longitude:Enlem, Latitude:Boylam).

Arazi çalışmalarından elde edilen bireylerin tür teşhis işlemleri Ege Üniversitesi, Su Ürünleri Fakültesi, Su Ürünleri Temel Bilimleri Bölümü bünyesinde bulunan Limnoloji Laboratuvarı'nda gerçekleştirilmiştir. Türlerin teşhis edilmesi bireylerin genel morfolojilerine ve vücut kısımlarının morfolojik yapılarına göre yapılmış; erkeklerin gonopodium yapıları tür teşhislerinde özel olarak dikkate alınmıştır (Berg 1965; Özuluğ vd. 2007).

Bulgular

Gambusia cinsinin Türkiye'deki dağılımını araştırmak amacı ile Türkiye'nin 6 farklı coğrafik bölgesinde (Ege, Akdeniz, Güneydoğu Anadolu, İç Anadolu, Karadeniz ve Marmara) bulunan toplamda 130 lentik ve lotik su kaynağında gerçekleştirilen arazi çalışmaları neticesinde, toplam 67 lokalitede türün varlığı tespit edilmiştir. Geri kalan 63 lokalitede türün varlığına rastlanmamıştır. Tür teşhisleri neticesinde örneklenen tüm bireylerin sadece *G. holbrooki* türüne ait olduğu, bu tür dışında örneklemelerde *Gambusia* genusuna ait başka türde bireyin elde edilmediği tespit edilmiştir.

Arazi çalışmaları sırasında balığın yakalanmış olduğu lentik ve lotik lokalitelerin bulundukları bölgeler, ait oldukları akarsu havzaları, rakımları ve koordinatları Tablo 2 ve 3'te verilmektedir. Türün daha önceden verilmiş her hangi bir kaydına ulaşılamayan lokaliteler tablo üzerinde yeni kayıt olarak belirtilmiştir.

Çeşitli araştırıcılardan temin edilen *G. holbrooki* örnekleri Tablo 4'te verilmiştir.

Yürütülen arazi çalışmaları kapsamında örnekleme çalışması yapılan bazı su kaynaklarının fotoğrafları Şekil 1'de verilmiştir. Türkiye'de yürütülen arazi çalışmaları kapsamında türün tespit edilemediği su kaynakları Tablo 5'te verilmiştir.

Tablo 2. Lentik lokalitelerin adları, bölgeleri, havzaları, rakım, konum ve önceki kayıtları.

No	Lokalite	B	Havza	Rakım(m)	Enlem	Boylam	Önceki Kayıtlar
G1	Belevi Gölü	E	Küçük Menderes	2	38,01˚	27,45˚	*Yeni Kayıt*
G2	Kazan Gölü	E	Küçük Menderes	3	37,98˚	27,26˚	*Yeni Kayıt*
G3	Sazlıgöl	E	Gediz	2	38,56˚	26,86˚	*Yeni Kayıt*
G4	Sülüklügöl	E	Gediz	609	38,57˚	27,50˚	*Yeni Kayıt*
G5	Marmara Gölü	E	Gediz	74	38,58˚	28,00˚	İlhan ve Sarı (2013)
G6	Kırkgöz Kaynakları	A	Antalya	303	37,10˚	30,58˚	Güçlü ve Küçük (2011)
G7	Titreyengöl	A	Antalya	4	36,75˚	31,45˚	Küçük ve İkiz (2004)
G8	Seyhan Baraj Gölü	A	Seyhan	57	37,03˚	35,31˚	Erk'akan ve Özdemir (2011)
G9	Mercimek Gölleri	A	Ceyhan	25	37,05˚	35,75˚	*Yeni Kayıt*
G10	Eber Gölü	İA	Akarçay	967	38,61˚	31,12˚	*Yeni Kayıt*
G11	Karamık Bataklığı	E	Akarçay	1007	38,37˚	30,74˚	*Yeni Kayıt*
G12	Işıklı Gölü	E	Büyük Menderes	816	38,25˚	29,93˚	Balık (1979)
G13	Kocagöl	E	Batı Akdeniz	1	37,28˚	27,65˚	*Yeni Kayıt*
G14	Akgöl	A	Batı Akdeniz	1	36,69˚	29,03˚	Öztürk ve İkiz (2003)
G15	Güllük Lagünü	E	Batı Akdeniz	0	37,25˚	27,62˚	*Yeni Kayıt*
G16	Bafa Gölü	E	Büyük Menderes	2	37,48˚	27,53˚	Balık (1979), Sarı vd. (1999b)
G17	Azap Gölü	E	Büyük Menderes	3	37,58˚	27,43˚	*Yeni Kayıt*
G18	Barutçu Gölü	E	Küçük Menderes	1	37,98˚	27,31˚	Balık ve Ustaoğlu (1988)
G19	Kocagöl	E	Küçük Menderes	1	37,94˚	27,33˚	*Yeni Kayıt*
G20	Gebekirse Gölü	E	Küçük Menderes	2	37,98˚	27,30˚	Balık ve Ustaoğlu (1988)
G21	Eğirdir Gölü	A	Antalya	917	38,07˚	30,85˚	Küçük vd. (2009)
G22	Beyşehir Gölü	A	Konya Kapalı	1123	37,71˚	31,46˚	Yeğen vd. (2006)
G23	Karakuyu Gölü	E	Büyük Menderes	1006	38,05˚	30,21˚	*Yeni Kayıt*
G24	Acıgöl-Başmakçı su kaynakları	A	Burdur Gölleri	838	37,83˚	29,96˚	Wildekamp vd. (1997)
G25	VR.Yazıcıoğlu B.	E	Büyük Menderes	328	37,76˚	29,12˚	*Yeni Kayıt*
G26	Güllübağ Göleti	E	Büyük Menderes	716	38,37˚	29,04˚	*Yeni Kayıt*
G27	Uluabat Gölü	M	Susurluk	2	40,16˚	28,67˚	Berber vd. (2011)
G28	İznik Gölü	M	Marmara	83	40,46˚	29,33˚	Özuluğ vd. (2005a)
G29	Gölbaşı Gölü (Hatay)	A	Asi	84	36,50˚	36,48˚	Öztürk ve İkiz (2004)
G30	Gölbaşı Gölü (Adıyaman)	GDA	Ceyhan	883	37,79˚	37,65˚	*Yeni Kayıt*
G31	Güllapoğlu Göleti	M	Meriç	41	41,63˚	26,61˚	Güner (2010)

Tablo 2. Devamı.

No	Lokalite	B	Havza	Rakım(m)	Enlem	Boylam	Önceki Kayıtlar
G32	Mert Gölü	M	Marmara	0	41,86˚	27,96˚	*Yeni Kayıt*
G33	Poyrazlar Gölü	K	Sakarya	25	40,83˚	30,46˚	*Yeni Kayıt*
G34	Sapanca Gölü	M	Sakarya	30	40,70˚	30,31˚	Özuluğ vd. (2007)
G35	Efteni Gölü	K	Batı Karadeniz	118	40,75˚	31,05˚	*Yeni Kayıt*
G36	Mogan Gölü	İA	Kızılırmak	975	39,77˚	32,78˚	*Yeni Kayıt*
G37	Bahçecik Göleti	İA	Sakarya	894	39,41˚	31,33˚	*Yeni Kayıt*

B: Bölge, E: Ege Bölgesi, A: Akdeniz Bölgesi, İA: İç Anadolu Bölgesi, GDA: Güneydoğu Anadolu, M: Marmara Bölgesi, K: Karadeniz Bölgesi

Tablo 3. Lotik lokalitelerin adları, bölgeleri, havzaları, rakım, konum ve önceki kayıtları.

No	Adı	B	Havza	Rakım(m)	Enlem	Boylam	Önceki Kayıtlar
A1	Karpuz Çayı	A	Batı Akdeniz	1	36,71˚	31,55˚	*Yeni Kayıt*
A2	Alara Çayı	A	Antalya	5	36,66˚	31,65˚	Küçük ve İkiz (2004)
A3	Sultansuyu	A	Doğu Akdeniz	3	36,03˚	32,81˚	Geldiay ve Balık (1996), Balık (1988)
A4	Akgöl kanal	A	Doğu Akdeniz	2	36,33˚	33,99˚	*Yeni Kayıt*
A5	Berdan Çayı	A	Doğu Akdeniz	27	36,95˚	34,88˚	*Yeni Kayıt*
A6	Seyhan Nehri	A	Seyhan	1	36,75˚	35,01˚	Balık (1988)
A7	Karataş kanal (Seyhan Nehri)	A	Seyhan	2	36,60˚	35,38˚	*Yeni Kayıt*
A8	Akyatan Lagünü kanal 1	A	Seyhan	1	36,63˚	35,31˚	*Yeni Kayıt*
A9	Akyatan Lagünü kanal 2	A	Seyhan	1	36,60˚	35,35˚	*Yeni Kayıt*
A10	Sumbaş Çayı Bulduklu k.	A	Ceyhan	38	37,38˚	35,90˚	*Yeni Kayıt*
A11	Gölyazı Köyü su kaynağı	İA	Konya Kapalı	918	38,55˚	33,20˚	*Yeni Kayıt*
A12	Sarısu sazlıkları	A	Batı Akdeniz	1	36,72˚	28,71˚	*Yeni Kayıt*
A13	Sarısu Çayı 1	A	Batı Akdeniz	2	36,71˚	28,71˚	*Yeni Kayıt*
A14	Sarısu Çayı 2	A	Batı Akdeniz	1	36,70˚	28,71˚	*Yeni Kayıt*
A15	Kocagöl kanal	E	Batı Akdeniz	1	36,70˚	28,81˚	*Yeni Kayıt*
A16	Yuvarlakçay	A	Batı Akdeniz	3	36,90˚	28,68˚	Balık vd. (2005), Yılmaz vd. (2006)
A17	Sarıçay	E	Batı Akdeniz	12	37,30˚	27,70˚	*Yeni Kayıt*
A18	Söke Milas yolu Drenaj K.	E	B. Menderes	4	37,58˚	27,35˚	*Yeni Kayıt*
A19	Pınarbaşı Kaynakları	A	Burdur Gölleri	998	37,45˚	30,05˚	*Yeni Kayıt*
A20	Narlı Deresi	E	B. Menderes	550	38,32˚	29,10˚	*Yeni Kayıt*
A21	Gediz Nehri eski kanalı	E	Gediz Havzası	1	38,61˚	26,81˚	Balık (1979)
A22	Miliç Irmağı	K	Yeşilırmak	4	41,18˚	37,01˚	*Yeni Kayıt*
A23	Kızılırmak Deltası	K	Kızılırmak	1	41,66˚	36,03˚	Uğurlu ve Polat (2007)
A24	Bakırçay	E	Kuzey Ege	3	38,95˚	27,01˚	*Yeni Kayıt*
A25	Kocaçay	M	Susurluk	17	40,13˚	28,04˚	*Yeni Kayıt*
A26	Tatarhüyük mevkii	M	Meriç	105	41,69˚	26,69˚	*Yeni Kayıt*

Kısaltmalar için Tablo 2'ye bakınız.

Tablo 4. Çeşitli araştırıcılardan temin edilen *G. holbrooki* bireylerinin lokaliteleri.

No	Tarih	B	Havza	İstasyon Adı
G38	08.04.2014	GDA	Fırat	Tahta Köprü Barajı (Gaziantep)
G39	13.11.2014	GDA	Fırat	Dicle Üniversitesi Göleti (Diyarbakır)
A27	08.06.2014	GDA	Gediz	Kuerk Nehri/Sinneş Deresi (Kilis)
A28	13.10.2016	E	Gediz	Gülbahçe Deresi (İzmir)

Kısaltmalar için Tablo 2'ye bakınız.

Eğirdir Gölü (Isparta)

Gölbaşı Gölü (Antakya)

Berdan Çayı (Mersin)

Poyrazlar Gölü (Adapazarı)

Şekil 1. Arazi çalışması kapsamında örnekleme çalışması yapılan bazı lokaliteler.

Tablo 5. Tarama çalışmaları sırasında türün tespit edilemediği su kaynakları.

No	Tarih	Lokalite Adı	Bulunduğu Şehir
1	06/04/2016	Değirmendere	Menemen-İzmir
2	06/04/2016	Emiralem Regülatörü	Menemen-İzmir
3	06/04/2016	Çukurköy Deresi	Menemen-İzmir
4	06/04/2016	Gediz Nehri eski kanalı	Buruncuk-İzmir
5	06/04/2016	Gediz Nehri eski kanalı	Haykıran-İzmir
6	06/04/2016	Gediz Nehri eski kanalı	Menemen (Seyrek köy drenaj kanalları)-İzmir
7	14/05/2016	Dim Çayı	Alanya-Antalya
8	16/05/2016	Akgöl	Silifke-İçel
9	19/05/2016	Tersakan Gölü	Cihanbeyli-Konya
10	19/05/2016	Bolluk Gölü	Cihanbeyli-Konya
11	19/05/2016	Mamasın Barajı	Merkez-Aksaray
12	20/05/2016	Akşehir Gölü	Akşehir-Konya
13	23/05/2016	Dalaman Çayı	Anakol-Narlıköyü mevkii-Dalaman-Muğla
14	23/05/2016	Dalaman Çayı	Anakol-Ortaca mevkii-Muğla
15	24/05/2016	Bafa Gölü	Serçin Gölü kısmı-Serçin-Söke-Aydın
16	05/06/2016	Eren Çayı	Hacılar-Burdur
17	05/06/2016	Büğdüz çayı	Suludere-Burdur
18	05/06/2016	Karataş Gölü	Bahçeözü-Karamanlı-Burdur
19	05/06/2016	Salda Gölü	Dere ağızları-Salda-Yeşilova-Burdur
20	05/06/2016	Yarışlı Gölü	Kocapınar-Sazak köyleri-Yeşilova-Burdur
21	07/06/2016	Acıgöl[2]	Başmakçı-Afyonkarahisar
22	07/06/2016	Selevir Barajı	Şuhut-Afyonkarahisar
23	07/06/2016	Burdur Gölü	Merkez-Burdur
24	08/06/2016	Yayla Gölü	Süleymaniye Gölü-Buldan-Denizli
25	10/10/2016	Sevişler Barajı	Soma-Balıkesir
26	11/10/2016	Kelebek Çayı	Turgutlu-Manisa
27	11/10/2016	Matdere	Salihli-Manisa

Tablo 5. Devamı

No	Tarih	Lokalite Adı	Bulunduğu Şehir
28	11/10/2016	Mersindere	Salihli-Manisa
29	11/10/2016	Sardes Deresi	Salihli-Manisa
30	11/10/2016	Demirköprü Barajı	Köprübaşı-Manisa
31	11/10/2016	İkizcetepeler Barajı	İnkaya-Balıkesir
32	11/10/2016	Sarıbeyler Barajı	Savaştepe-Balıkesir
33	11/10/2016	Kocaçayı	Manyas-Balıkesir
34	11/10/2016	Sultançayır Nehri	Susurluk-Balıkesir
35	11/10/2016	Mustafakemalpaşa Çayı	Demireli-Bursa
36	03/08/2017	Tekir Göleti	Erciyes-Kayseri
37	03/08/2017	Alaca Barajı	Alaca-Çorum
38	02/08/2017	İnekli Gölü	Gölbaşı-Adıyaman
39	02/08/2017	Azaplı Gölü	Gölbaşı-Adıyaman
40	02/08/2017	Gavur Gölü	Türkoğlu-Kahramanmaraş
41	02/08/2017	Sır Barajı	Ceyhan Nehri üzeri-Kahramanmaraş
42	02/08/2017	Aksu Çayı	Kahramanmaraş
43	02/08/2017	Atatürk Barajı	Fırat Nehri üzeri-Adıyaman
44	04/08/2017	Taflan Deresi	Atakum-Samsun
45	04/08/2017	Horhor Çayı	Samsun
46	05/08/2017	Sarıkum	Merkez-Sinop
47	02/11/2017	Ömerli Baraj Gölü	Çekmeköy-İstanbul
48	02/11/2017	Kuzuludere	Çatalca-İstanbul
49	02/11/2017	Yeniçağa Gölü	Merkez-Bolu
50	02/11/2017	Kömürlük Deresi	Şile-İstanbul
51	02/11/2017	Büyükçekmece Gölü (Güney kısmı)	Çatalca-İstanbul
52	03/11/2017	Yapraklı Barajı	Gölhisar-Burdur
53	03/11/2017	Eymir Gölü1	Çankaya-Ankara
54	04/11/2017	Yenice Barajı	Eskişehir
55	16/10/2017	Büyükdere	Manisa
56	16/10/2017	Yağcılı Deresi	Manisa
58	18/10/2017	Karadere	İzmir
60	18/10/2017	Çaltıkoru Barajı	Bergama-İzmir
61	18/10/2017	Çaltıkoru Deresi	Bergama-İzmir
62	18/10/2017	İlyasdere	Merkez-Aydın
59	19/10/2017	Çandarlı Dalyan	Dikili-İzmir
57	19/10/2017	Kestel Barajı	Bergama-İzmir
63	19/10/2017	Bakırçay anakol	Bergama-İzmir

1:Gölde araştırma yapmak için Orta Doğu Teknik Üniversitesi'nin özel izni gerektiğinden göl çok sınırlı bir şekilde kontrol edilebilmiştir.
2:Acıgöl'ün kendi suları türün yaşaması için uygun fizikokimyasal özellikte değildir. Ancak tür Acıgöl kaynaklarında varlığını sürdürebilmektedir.

Tartışma

Sivrisinek balıklarının Türkiye'deki dağılımının ağırlıklı olarak Ege, Akdeniz ve Marmara Bölgeleri'nde olduğu görülmüş; arazi çalışmaları kapsamında incelenen lokaliteler içinde, türün daha önceden kaydının verilmiş olduğu lokalitelerin neredeyse tamamında varlığını korumaya devam ettiği tespit edilmiştir. Türün daha önceden kaydının verilmiş olup, bu çalışma sırasında varlığının tespit edilemediği 3 lokalite (Büyükçekmece Gölü, Gediz Nehri, Ömerli Baraj Gölü) bulunmaktadır. Çalışma sırasında arazi şartları sebebi ile Büyükçekmece Gölü'nün sadece güney cephesi kontrol edilebilmiş ve gölün bu cephesinde türe rastlanmamıştır (Ancak gölün kuzey cephesinde türün mevcut bulunduğu ifade edilmektedir (Doç.Dr. Özcan GAYGUSUZ 2017, sözlü görüşme)). Gediz Nehri'nin kanal genişletme çalışması yapılan kısımlarında türün varlığı tespit edilememiştir. Ömerli Baraj Gölü ise çok büyük ve içerisinde çok farklı mikrohabitatlar barındıran bir baraj gölüdür, tarama çalışmasında baraj gölü her ne kadar bu farklı kısımlarından

kontrol edilmiş ve tür tespit edilememiş olsa da; türün baraj gölündeki varlığını devam ettiriyor olması olasıdır.

Türkiye'de yürütülen arazi çalışmaları kapsamında türün tespit edilemediği su kaynakları Tablo 5'te verilmiştir. Bu lokalitelerden bazılarının fizikokimyasal özellikleri, bazılarının ise habitat özellikleri türün ihtiyaçlarını karşılamamakta olduğundan türün bu lokalitelerde dağılım gösteremediği tahmin edilmektedir. Örneğin Tersakan ve Bolluk Gölleri yüksek miktarlarda sodyum sülfat barındıran göllerdir ve bu durum türün yaşamına uygun olmayan bir özellik yaratmaktadır. Dim Çayı, Dim Barajı'ndan Akdeniz'e döküldüğü noktaya dek oldukça yüksek bir debi ile akan bir akarsu niteliğindedir ve bu özelliğiyle türün durgun suya olan ihtiyacını karşılamamaktadır. Dalaman Çayı (anakol), Bakırçay (anakol) ve Emirelam Regülatörü de tıpkı Dim Çayı gibi hızlı akan akarsu kaynaklarıdır. Eren ve Büğdüz çaylarının örnekleme yapılan dönemde oldukça az miktarda su bulundurduğu tespit edilmiştir, tür ilgili su kaynaklarında bu nedenle tespit edilememiş olabilir. Mustafakemalpaşa Çayı oldukça kirlidir ve türün bu nedenle burada yaşam şansı bulamadığı tahmin edilmektedir, zira örnekleme çalışmaları sırasında suda başka balık türüne de rastlanmamıştır. Demirköprü, İkizcetepeler ve Sarıbeyler barajlarında vejetasyon oranı oldukça düşüktür ve bu barajlar türün vejetasyon ihtiyacını karşılaşamamaktadır. İnekli Göl, Gavur Gölü ve Azaplı Göl ise örnekleme yapılan dönemde tarımcılık faaliyetleri amaçlı olarak büyük ölçüde kurutulmuştur. Bu göllerden arta kalan su birikintilerinde ise türe rastlanmamıştır. Akşehir Gölü'nün suları örnekleme döneminde çekilmiş durumdadır, göl çevresi balçıkla kaplıdır ve bu nedenle göl çok sınırlı bir şekilde kontrol edilebilmiştir. Gediz Nehri eski kanalında örnekleme yapılan dönemde kanal genişletme çalışması olduğundan habitatlar tahrip edilmiş durumdadır, bu nedenle türün bu alanda bulunamadığı tahmin edilmektedir (Nehrin çalışma yapılmayan kısımlarında türün varlığı tespit edilmiştir). Burdur, Salda, Yarışlı ve Karataş göllerinin sahip oldukları özgün fizikokimyasal özellikler sebebi ile türün yaşamına elverişli olmadığı düşünülmektedir.

Yapılan çalışma neticesinde *G. holbrooki*'nin Ege, Akdeniz, Karadeniz ve Marmara bölgelerindeki su kaynaklarının büyük bir çoğunluğunda dağılım gösterdiği; İç Anadolu Bölgesi'nde ise dağılımının oldukça sınırlı olduğunu ortaya koyulmuştur. Çalışmadan elde edilen türün dağılım haritasının, Köppen-Geiger (Peel vd. 2007) Türkiye iklim modelinde özellikle C sınıfı iklim modelinin (B Sınıfı: Kurak, *Temel Özelliği:* Buharlaşma yağıştan fazladır ve sürekli su eksiği vardır. C Sınıfı: Kışları ılıman nemli, orta enlem, *Temel Özelliği*: En soğuk ayın ortalama sıcaklığının 18°C'nin altında ve 0°C'nin üstünde, en sıcak ayın ortalama sıcaklığı 10°C'nin üstündedir. D Sınıfı: Kışları soğuk ve nemli, orta enlem, *Temel Özelliği*: En soğuk ayın sıcaklığı 0°C'nin altındadır ve en sıcak ayın ortalama sıcaklığı 10°C'nin üstündedir) görüldüğü alanlar ile fazlasıyla örtüşmesi oldukça dikkat çekicidir (Öztürk vd. 2017). Bu durum, türün dağılımında, sıcaklığın oldukça etkin bir faktör olduğunu düşündürmektedir. Ancak bu noktada sıcaklık dışında türün dağılımını sınırlayan bazı fizikokimyasal parametrelerin (tuzluluk, soda, pH vb.) de varlığı açıktır. Bununla birlikte *G. holbrooki* türünün daha çok akarsuların mansap bölgeleri ile göllerin az akıntılı ve vejetasyonlu kısımlarında başarılı popülasyonlar oluşturabildiği; hızlı akan suları yaşam alanı olarak tercih etmediği gözlemlenmiştir.

G. holbrooki'nin giriş yaptığı su ortamlarına hızlı ve kolay adapte olabilmesi, değişen su parametrelerine toleransının çok yüksek oluşu ve sahip olduğu üstün üreme stratejileri, türün 1920'li yıllarda yapılan ilk aşılamasından sonraki 100 yıl gibi kısa bir süre içinde Türkiye'de sahip olduğu hızlı ve güçlü dağılımını açıklamaktadır.

Akvaryum ticareti yolu ile balıkların kıtalararası da dâhil olmak üzere oldukça rahat pazarlandığı günümüz dünyasında, her ne kadar bu çalışma kapsamında tek bir *Gambusia* türünün varlığı tespit edilmiş de olsa, *G. holbrooki* türü dışında *Gambusia* genusuna ait olan başka bir türün sularımıza girip girmediği hala şüphelidir. Bununla birlikte Türkiye'de yapılan kontrolsüz balıklandırma çalışmaları ve bilinçsizce hareket eden kişiler nedeni ile aşılanan türlerin hangi su kaynaklarından getirildiği bilinemediğinden, sivrisinek balık türlerinin Türkiye'deki durumları henüz net bir açıklığa kavuşturulamamıştır.

Dağılım gösterdiği habitatlarda meydana getirdiği olumsuz etkiler ile yerli ve endemik türlerin yok olmasına sebep olabilen *G. holbrooki* türü (Pyke 2008) ile etkin ve doğru bir şekilde mücadele edebilmenin yolu, yapılacak bilimsel çalışmaların niteliğinin ve niceliğinin arttırılmasından geçmektedir. Türün sahip olduğu başarılı üreme stratejilerinin güçlü popülasyonlar oluşturabilmesindeki etkisi bilinmekte olduğundan, türün üremesi özellikle izlenmelidir.

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In Vitro Storage of Unfertilized Eggs of Scaly Carp (*Cyprinus carpio*) : Effect of Different Artificial Mediums and Storage Periods

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ABSTRACT

The present study was carried out in order to explore the effect of different artificial mediums and *in vitro* storage periods on fertilization, eyeing and hatching success of scaly carp (*Cyprinus carpio*) eggs. The batches of about 200 pooled eggs treated with 20-ml three different extenders (Ringer, Dettlaff and Cortland) and Ovarian fluid in 15-cm petri dishes, were stored at 22.5° C for 30, 60, or 90 min. The *in vitro* stored eggs were fertilized by adding of 50 µl sperm which showing motility higher than 80%, in each petri dishes end of the storage period. The highest fertilization rates were determined as 86% and 72% with the egg samples stored for 60 min in Cortland solution and for 90 min in Ovarian fluid respectively ($p<0.05$). The highest eyeing rate (80%) was determined in egg samples kept in Cortland solution for 60 min storage ($p<0.05$). Despite the best hatching rate (60%) of the egg samples determined with Ovarian fluid at 30 min storage, the Cortland solution was (48%) the best for 90 min ($p<0.05$). Results indicate that Cortland solution is the most suitable extender and can be substituted instead of Ovarian fluid for *in vitro* storage of scaly carp eggs.

Keywords: Extender, egg, short-term preservation, fertilization

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Döllenmemiş Pullu Sazan (*Cyprinus carpio*) Yumurtalarının *İn Vitro* Muhafaza Edilmesi: Sulandırıcı ve Muhafaza Sürelerinin Etkisi

Öz: Bu çalışma, farklı sulandırıcı ve *in vitro* muhafaza sürelerinin pullu sazan (*Cyprinus carpio*) yumurtalarının döllenme, gözlenme ve açılma başarısı üzerine olan etkilerinin araştırılması amacıyla yürütülmüştür. 20-ml hacmindeki üç farklı sulandırıcı (Ringer, Dettlaff ve Cortland) ve ovaryum sıvısı ile 15-cm çaplı petri kutularında muamele edilen yaklaşık 200 yumurta, 22.5° C'de 30, 60 ve 90 dk süre ile muhafaza edildi. *İn vitro* muhafaza edilen yumurtalar, muhafaza süresinin sonunda %80'den fazla motil özelliğe sahip olan 50 µl sperma ile her petri kutusunda döllenmiştir. En yüksek fertilizasyon oranları, Cortland solüsyonunda 60 dk ve ovaryum sıvısında 90 dk muhafaza edilen yumurta örneklerinde sırasıyla %86 ve %72 olarak belirlenmiştir ($p<0,05$). En yüksek gözlenme oranı Cortland solüsyonunda 60 dk muhafaza edilen yumurta örneklerinde %80 olarak belirlenmiştir ($p<0,05$). En iyi açılma oranı ovaryum sıvısında 30 dk muhafaza edilen yumurta örneklerinde %60 olarak belirlenmesine rağmen, 90 dk muhafaza süresinde ise en iyi sonuç Cortland solüsyonunda %48 olarak elde edildi ($p<0,05$). Elde edilen sonuçlar, Cortland solüsyonunun en iyi sulandırıcı olduğunu ve pullu sazan yumurtalarının *in vitro* muhafazasında ovaryum sıvısı yerine ikame edilebileceğini göstermektedir.

Anahtar kelimeler: Sulandırıcı, yumurta, kısa süreli muhafaza, döllenme

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Introduction

Conservation of gametes in the cold or frozen state is an important biotechnological tool and has great concern for aquaculture. Growing attention to this biotechnology has guided an increasing number of researches in this field (Carolsfeld et al. 2003; Tiersch 2011). The long-term conservation of fish sperm has been widely explored and optimized with cryopreservation techniques. Nowadays, it is possible using cryopreserved sperm in artificial propagation applications in aquaculture. On the other hand, similar methods regarding the conservation of eggs have not resulted in satisfactory results due to the

difficulty of removing intercellular water during the cooling process in the cells (Stoss 1983).

Mature eggs may be retained for weeks until ovulation. Ovulated eggs retained in the ovarian cavity are exposed to overripening because of morphological and biochemical modifications affecting gamete and fertility quality negatively (Formacion et al. 1993). Nevertheless, time for storage is still available for effective using of ovulated eggs. Thus, prolonging of egg viability is an important issue in term of aquaculture practices (Rothbard et al. 1996).

The ovulated eggs should be fertilized within a certain period of time following the ovulation process in order to obtain viable embryos (Hobby and Pankhurst 1997). The important issue to be known that fertilization of ovulated eggs progressively decreases and then disappears when they are kept in the body cavity of the female for a long time. On the other hand, it is possible maintaining of fertility much longer for the eggs preserved in saline or coelomic solutions (Dettlaff et al. 1993). Conservation of fish eggs diluted with a suitable extender has many applications such as transporting of the gametes to other hatcheries in order to synchronize artificial propagation operations and chromosome manipulations (Suquet et al. 1999). Therefore, the establishment of an effective protocol is an important issue in terms of commercial aquaculture.

From this point of view, the current study was designed to explore the effect of different extenders and storage periods on fertility, eyeing and hatching success of scaly carp eggs following *in vitro* storage under culture conditions.

Materials and Methods

Broodstock management and collection of gametes

Mature male and female scaly carp broodfish which are suitable for the artificial propagation (+3 years of age), were selected in their natural spawning season from large soil ponds and were kept separately according to their sex in the hatchery. The broodfish which kept in 500-l holding tanks supported with a water flow of 0.2 l/s at 24°C containing 6-7 mg/l dissolved O_2, were fed with commercial feed (22% crude protein) at 1.5% ratio of body weight per day. Experiments were performed with 15 females (42.6±4.5 cm total length, 1.2±0.4 kg body weight) and three males (46.8±2.4 cm total length, 1.1±0.5 kg body weight) during the peak (May) of the reproductive period.

Females with a large, soft abdomen, red urogenital papilla, and males appearing sperm with slight abdominal pressure were assumed to be ready for the hormone treatment. Commercially available carp pituitary extract (CPE) was used as spawning agent. In order to induce spawning, CPE in Ringer solution was applied intramuscularly to the females in two portions as the priming and releasing doses (0.5 and 2.0 mg/kg body weight respectively) with a 14-h interval between doses. Males received a single dose of CPE (1.0 mg/kg body weight) intramuscularly at the time of the second dose was applied to the females (Bozkurt and Yavas 2012).

100 mg/L quinaldine sulfate was applied to the broodfish for anesthetization and stripped via gentle abdominal massage. Eggs, Ovarian fluid, and sperm were collected separately from each female and male broodfish. Contamination of gametes with urine, mucus, blood or feces was avoided during the collection of gametes. The Ovarian fluid separated by pouring the eggs onto a screen suspended over a plate was used for storage purposes.

Sperm samples collected from three males into 50-ml glass beakers were checked in term of motility at the end of each storage period. Samples showing more than 80% motility, were pooled in a 100-ml glass beaker. Fresh sperm was placed in a refrigerator (+4^{o} C) to prevent deformation for a few minutes until used fertilization of the pooled eggs.

Evaluation of spermatoa motility and density

10 µl drops of sperm were placed on a microscope slide and 20 µl activation solution (0.3% NaCl) was added. Sperm suspension was thoroughly mixed and spermatozoa motility was evaluated in terms of progressive forward movement using light microscope (Olympus, Japan) with an x400 magnification. The motility percentages were defined as the percentage of spermatozoa moving in a forward motion every 20% motile increment (i.e., 0, 20%, 40%, 60%, 80%, and 100%) (Vuthiphandchai and Zohar 1999).

For the purpose of evaluation of spermatozoa density, sperm was diluted at a ratio of 1:1000 with Hayem solution (5g Na_2SO_4, 1g NaCl, 0.5g $HgCl_2$, 200 ml bicine) and density was determined using a 100µm deep Thoma haemocytometer (TH-100, Hecht-Assistent, Sondheim, Germany) at 400x magnification with phase contrast microscope (Olympus, Japan) expressing as spz. x10^9/ ml (three replicates) (Yıldız et al. 2013).

Storage of ovulated eggs

A composition of three artificial mediums used for the *in vitro* storage of unfertilized scaly carp eggs is shown in Table 1. The final pH of each medium was regulated according to the natural pH of the coelomic fluid of scaly carp using 1.0 N HCl and 1.0 N NaOH. In order to store eggs, a batch of 200 pooled eggs was gently placed in

15-cm diameter petri dishes. The batches of eggs were washed with their storage mediums to eliminate coelomic fluid from the eggs. Petri dishes were filled with 20-ml of artificial mediums and Ovarian fluid and then *in vitro* stored for durations of 30, 60 and 90 min prior to fertilization inside of sterile Laminar air flow cabinet at a constant temperature of 22.5° C.

Table 1. Composition of the artificial mediums

Artificial Mediums	Composition	Reference
Ringer Solution	103 mM NaCl, 1 mM KCl, 1 mM $CaCl_2$, 1.1 mM $NaHCO_3$	(Linhart et al. 1991)
Dettlaff Solution	111.3 mM NaCl, 3.3 mM KCl, 2.1 mM $CaCl_2$, 23.8 mM $NaHCO_3$	(Dettlaff et al. 1993)
Cortland Solution	124 mM NaCl, 5.1 mM KCl, 1.0 mM $MgSO_4$.7H2O, 1.6 mM $CaCl_2$.2H_2O, 5.6 mM glucose	(Wolf and Quimby 1969)
Ovarian Fluid	741 mM Na^+, 0.45 mM K^+, 2.58 mM Mg^{2+}, 6.38 mM Ca^{2+}, 0.35 mM Zn^{2+}	(Linhart et al. 1995)

Fertilization, eyeing and hatching of eggs

Following 30, 60 and 90 min storage periods of unfertilized eggs, extenders and Ovarian fluids were removed from the petri dishes and 50 µl sperm showing motility higher than 80% and containing roughly $2x10^5$ spz. was added on to the eggs for the fertilization.

Fertilization process was performed in 100 ml plastic container using activation solution containing 3 g urea and 4 g NaCl in 1 l distilled water at 24° C for five minutes through slow homogenization. Then, eggs were washed with water in order to eliminate adhesiveness and gently transferred to the labeled Zuger glass incubators with running water (24°C) where kept until eyeing(14-16 h) and hatching(3-4 d).

Batches of eggs fertilized at the time of 0 (immediately after ovulation) served as a control group. Fertilization yield was determined at the gastrulation stage. The eyeing and hatching rate were defined as the number of eyed / hatched eggs divided by the initial number of eggs used for fertilization. Newly hatched larvaes were removed by siphoning and counted. Each treatment was performed in triplicate for each extender and storage period.

Statistical analysis

Differences between treatment groups were analyzed by repeated analysis of variance (one-way ANOVA). Significant means were subjected to a multiple comparison test (Duncan) for post-hoc comparisons at the level of α=0.05. Results are presented as mean±SE and all analyses were carried out using SPSS 17 for Windows Statistical Software Package.

Results

Regarding the effect of storage periods on fertilization of eggs, the highest results were obtained as 71.33±9.45%, 82.00±4.00% and 68.00±4.00% in Dettlaff and Cortland solutions and Ovarian fluid respectively for 30, 60 and 90 min storage periods (Figure 1). The overall meaning of the fertilizations were 64.83±7.74%, 65.5±11.69%, and 60.67±8.52% respectively for 30, 60 and 90 min storage periods that were significantly different ($p<0.05$) from 0 min control. Differences between mean fertilization rates within the same medium group were significant ($p<0.05$) except for the Ovarian fluid when duration was considered.

The highest eyeing rate (72.67±5.03%) was in eggs stored in Dettlaff solution for 30 min storage but Cortland solution was the best medium (74.67 ±5.03% and 63.33±7.02%) for 60 and 90 min storage respectively (Figure 2). The overall meaning of the eyeings were 68.83±6.52%, 66.00±7.67%, and 58.5±7.29% respectively for 30, 60 and 90 min storage periods that were significantly different ($p<0.05$) from 0 min control. Differences between mean eyeing rates within the same medium group were significant ($p<0.05$) when duration was considered.

In spite of obtaining the highest hatching rates (56.00±4.00% and 47.33±5.03%) with the Ovarian fluid for 30 min and 60 min storage, the Cortland solution was (43.33±4.16%) the best medium for 90 min storage (Figure 3). The overall meaning of the hatchings were 47.00±6.74%, 43.33±4.84%, and 38.67±4.84% respectively for 30, 60 and 90 min storage periods that were significantly different ($p<0.05$) from 0 min control. Differences between mean hatching rates only for the Ovarian fluid group were significant ($p<0.05$) when duration was taken into consideration.

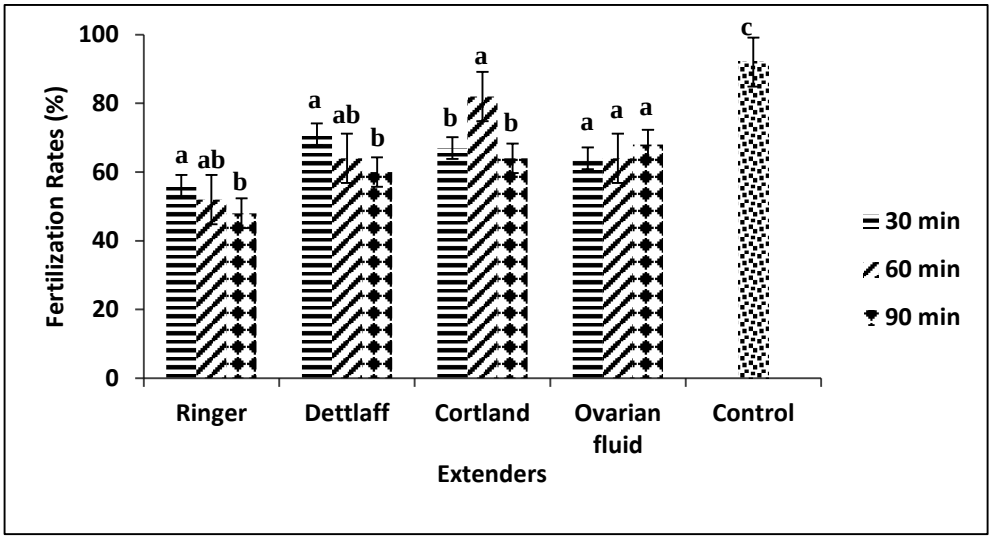

Figure 1. Fertilization rates of scaly carp eggs *in vitro* incubated in four different mediums for 0 (control), 30, 60 and 90 min storage prior to fertilization.

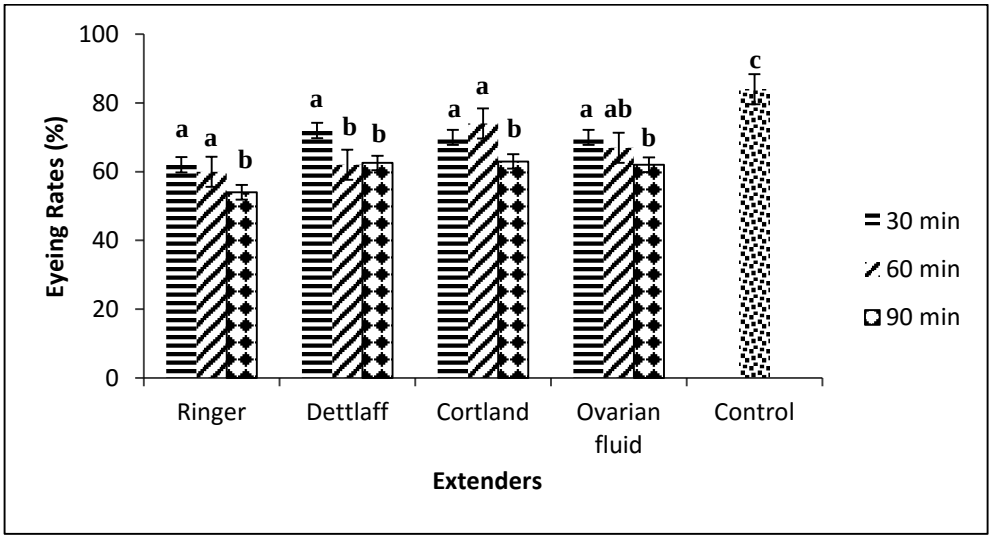

Figure 2. Eyeing rates of scaly carp eggs *in vitro* incubated in four different mediums for 0 (control), 30, 60 and 90 min storage prior to fertilization.

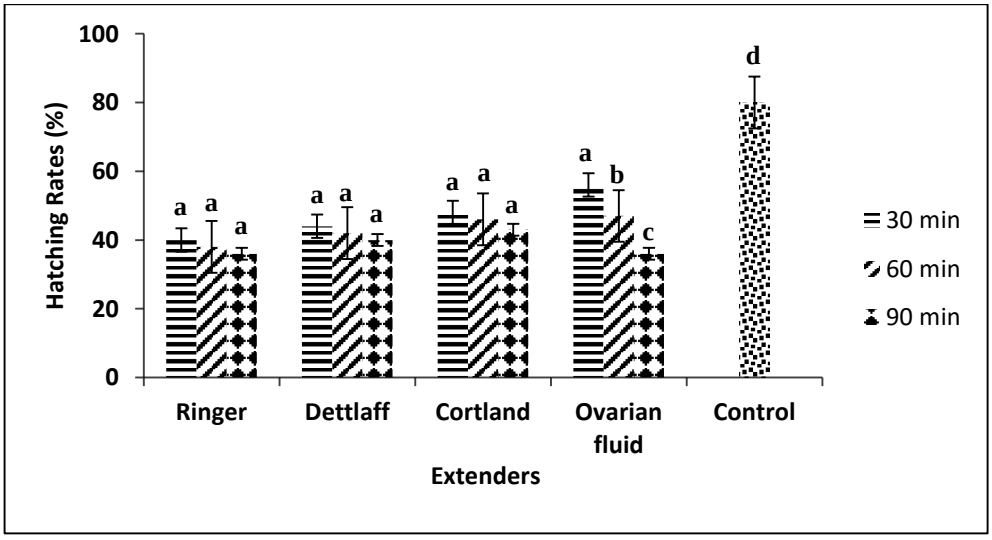

Figure 3. Hatching rates of scaly carp eggs *in vitro* incubated in four different mediums for 0 (control), 30, 60 and 90 min storage prior to fertilization.

Discussion

In spite of cryopreservation of fish sperm developed with a satisfactory level, it has been less successful for the fish eggs due to the difficulty of removing intercellular water during the cooling process. Until now, only a few studies explored *in vitro* storage of fish eggs.

It is known that mature eggs can be kept for several days in Ovarian fluid and the storage duration is related to the holding temperature (Samarin et al. 2017; Ginatullina et al. 2018; Bozkurt and Yavaş 2012). For instance, Rothbard et al. (1996) reported that common carp (*Cyprinus carpio*) eggs can be stored for 6 h at a temperature range of 20°C-24.5°C. Zlabek and Linhart (1987) compared *in vitro* storage of common carp (*C. carpio*), grass carp (*Ctenopharyngodon idella*) and silver carp, (*Hypophthalmichthys molitrix*) eggs at two temperature regimes (3°C-5C° and 14.5°C-18.0°C) and reported that common carp eggs exhibited better fertility and survivality of embryos than those of the Chinese carps.

On the other hand, Takano et al. (1973) reported that storing of eggs as *in vitro* in artificial mediums less effective for similar periods. According to Takano et al. (1973) chum salmon eggs could be stored for only one day in physiological saline. Similarly, Erdahl et al. (1987) indicated the possibility of short-term (5-10 min) storage of unfertilized rainbow trout eggs in physiological saline solution without loss of fertility.

Most researchers have explored the development of eggs from fertility to eyeing stage (Erdahl et al. 1987; Lahnsteiner and Weismann 1999; Goetz and Coffman 2000). It is well known that less attention has been given to the survival of embryos from eyeing to the hatching stage following storage in different mediums for longer periods prior to fertilization. Researches have proven that carp eggs can be *in vitro* stored for several hours in Ovarian fluid outside of the body maintaining its viability and fertility (Rothbard et al. 1996). On the other hand, very few studies have explored *in vitro* preservation of carp eggs in different artificial mediums.

It is well known that eggs belong to fish species must be fertilized quickly following ovulation process otherwise lose their fertility quickly (Formacion et al. 1995). On the other hand, the present study demonstrated that scaly carp eggs can maintain their fertility when stored as *in vitro* in artificial mediums such as Ringer, Dettlaff, and Cortland solutions.

From this point of view, the present study demonstrated that ionic based solutions and Ovarian fluid can be used as artificial mediums for the preservation of scaly carp eggs up to 90 min at relatively high temperature (22.5°C) under *in vitro* conditions. On the other hand, it is obvious that there is a decrease in terms of fertility, eyeing and hatchability of the eggs depending on period especially following 60 min storage in all cases except for Cortland solution.

The main reason for this increase in fertility at the end of the 60 min storage of the eggs in the Cortland solution and Ovarian fluid may be high sperm motility of fresh sperm cells. Results of the current study showed that the best fertilization and eyeing rates at each tested storage period were achieved when the eggs were treated with the Cortland solution. Similarly, Goetz and Coffman (2000) reported that unfertilized trout eggs can be maintained for at least two days without loss of fertility in modified Cortland solution buffered with Hepes or Tris. The present study also showed that while the eggs stored in Ovarian fluid showing the highest efficiency in term of hatching, the lowest one was the Ringer solution in all cases.

It should be known that the overall success of the conservation is highly depending on the extender composition, the origin of the broodfish and egg batch. Additionally, preservation of metabolic activity and maintaining of the viability of the eggs may be related to the health of the brood female, time of ovulation, incubation temperature and also the quality of hatchery water (Linhart and Billard 1995). From this point of view, the results of the current study showed that the survival of eggs in various treatments strongly depends on handling and management of spawners, temperature and egg quality prior to fertilization.

Furthermore, studies have indicated that bacterial infection might be one of the limiting factors in the preservation of fish eggs in artificial mediums and Ovarian fluid (Goetz and Coffman 2000; Niksirat et al. 2007). Hence, future studies should explore using of antioxidants and antibiotics to improve *in vitro* storage of eggs in scaly carp due to the positive effect of these substances in other fishes.

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The Biodiverse Rotifers (Rotifera: Eurotatoria) of Small Wetlands of the Brahmaputra River Floodplains of Lower and Upper Assam, Northeast India

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ABSTRACT

Rotifera assemblages of small floodplain wetlands (*dobas* or *dubies*) of lower and upper regions of the Brahmaputra river basin of Assam state of northeast India (NEI) reveal 157 species, belonging to 34 genera and 18 families, and highlight notable speciose constellation of up to 50 species/sample. One species, each is new to the Oriental region and Assam, and species of global and regional biogeographic interest form notable fractions. The biodiverse rotifer fauna and various interesting species are hypothesized to habitat diversity of the sampled *dobas* or *dubies*, impact of `the Assam-gateway'- an important biogeographic corridor of India, and location of the study area in the Indo-Burmese biodiversity hot-spot. Lecanidae and Lepadellidae are species-rich families, both of lower and upper Assam wetlands; upper Assam wetlands, in particular, are characterized by a distinct paucity of the Brachionidae and *Brachionus* spp., the relative paucity of *Trichocerca, Keratella* and *Mytilina* species, rare occurrence of *Asplanchna* and *Filinia* species, and lack of species of Conochilidae and Hexarthridae. Our results indicate the littoral-periphytic nature and tropical character of the rotifers. Overall, this study is an important contribution to Rotifera biodiversity of small lentic habitats of India, Asia and that of the tropics and subtropics.

Keywords: Biodiversity, *dobas*, *dubies*, interesting species, `Rotifera paradox'

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Introduction

Small lentic ecosystems form over 90% of the standing water resources of our biosphere and ~ 30% of the global lentic biotopes by surface area and are thus suggested to be more explicitly considered in analyses of biodiversity, limnology and global processes (Céréghino et al. 2014). They are recognized as hotspots both in terms of species composition and biological traits (EPCN 2008) and are often considered as keystone systems for the conservation of biodiversity (Oertli et al. 2010; Céréghino et al. 2014; Vad et al. 2017; Oertli 2018). The management of these biotopes and that of their metazoan diversity is usually neglected (Céréghino et al. 2008; Oertli 2018) despite more vulnerability to severe threats of extinction and habitat degradation (Moss et al. 2011). The small lentic biotopes have attracted relatively more attention in hydro-biological works from India, but the relevant literature largely depicts paucity of studies on zooplankton diversity. A critical analysis attributes this lacuna to the limited sampling, ad-hoc reports by amateurs loaded with incomplete species lists, and lack of taxonomic expertise. Nevertheless, our studies from NEI (Sharma and Sharma 2015a; Sharma and Kensibo 2017; Sharma et al. 2017) hypothesized these water bodies to be one of the biodiverse habitats of the Indian sub-region vis-à-vis Rotifera - an important group of freshwater zooplankton, an integral link of aquatic food-webs, and food for larvae and adult fish (Tuna and Ustaoğlu 2016; Apaydın Yağcı et al. 2017).

The state of Assam of NEI, a part of the Indo-Burmese biodiversity hotspot, is notably known for the fluvial floodplains of the Brahmaputra river with characteristic small lentic wetlands (commonly known as *dobas* or *dubies*) forming an integral part of the rural landscape of the Brahmaputra valley. Sharma and Sharma (2014) first indicated ecological diversity importance of small wetlands in context of

Rotifera of NEI and suggested their detailed assessment. The present study is thus an endeavor to analyze and compare species composition and richness of the rotifer assemblages of small wetlands of lower and upper reaches of the Brahmaputra river floodplains of Assam state. We provide an inventory of the documented taxa, illustrate interesting species, and comment on nature and composition of the rotifer fauna with reference to species richness, new records, important taxa, extraordinary high species consortium and elements of global and regional distribution interest. This study merits biodiversity and biogeographic interest for Rotifera of the floodplains of India, Asia as well as that of the rotifer heterogeneity of the small lentic ecosystems of the Indian sub-region.

Material and Methods

The present study is based on analysis of plankton and semi-plankton samples collected, on several occasions during 2005-2015, from small floodplain wetlands (*dobas* or *dubies*) from scattered localities (Figure 1.A-C) of Dhubri, Goalpara, Kokrajhar, Bongaigaon, Barpeta, Kamrup, Nalbari and Dispur districts of lower Assam (24°8′-26°8′N; 89°8′-92°2′E), and from Biswanath, Golaghat, Jorhat, Majuli, Lakhimpur,

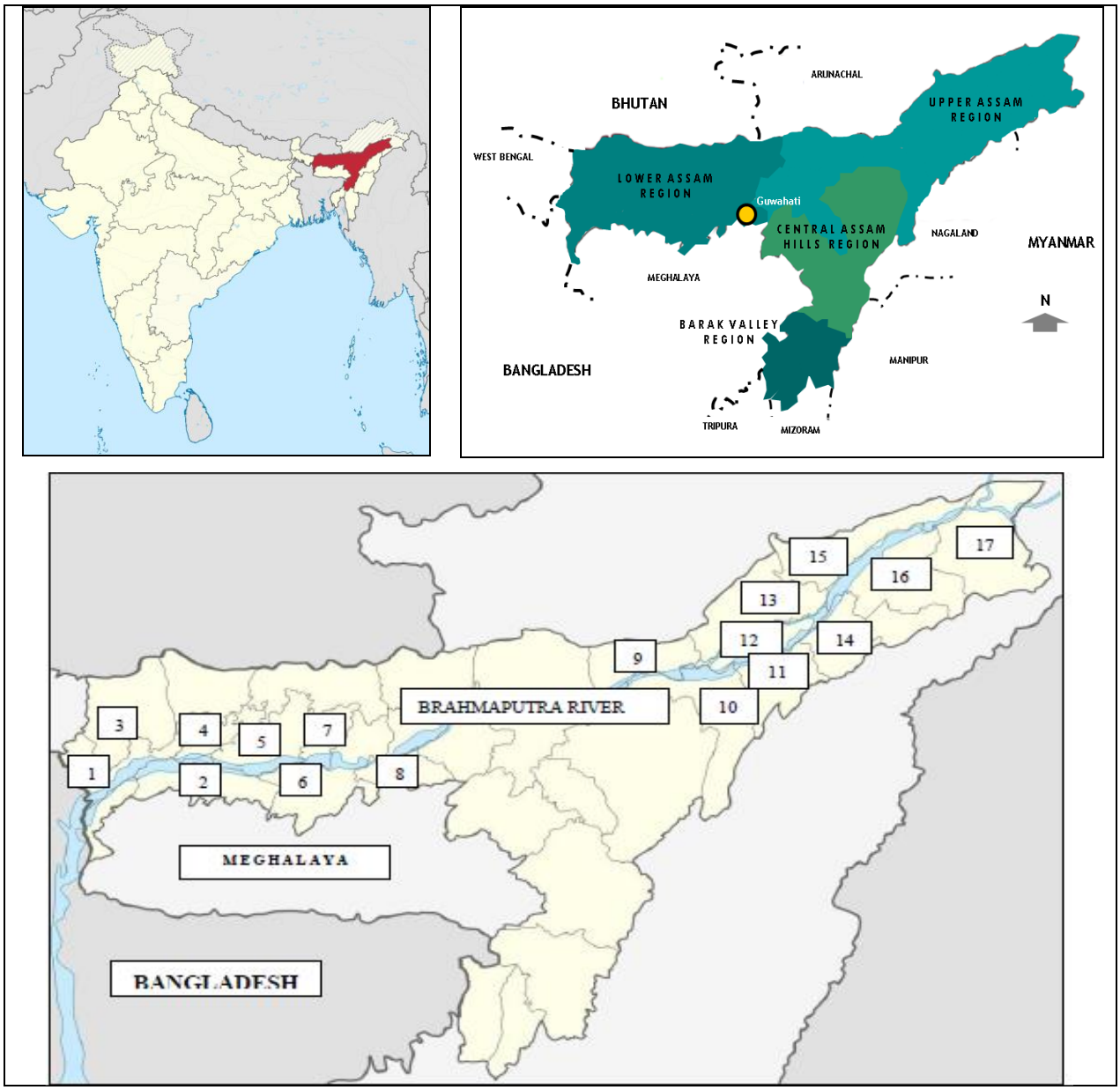

Figure 1. A-C: A, map of India indicating northeast India (NEI) and the state of Assam; B, map of Assam; C, map indicating the sampled districts of Lower Assam and Upper Assam [1-Dhubri; 2-Goalpara; 3-Kokrajhar; 4-Bongaigaon; 5-Barpeta; 6-Kamrup; 7-Nalbari; 8-Dispur; 9-Biswanath; 10-Golaghat; 11-Jorhat; 12-Majuli; 13-Lakhimpur; 14-Sibsagar;15-Dhemaji;16-Dibrugarh;17-Tinsukia]

Sibsagar, Dhemaji, Dibrugarh and Tinsukia districts of upper Assam (26°4′-27°8′N; 93°8′-95°6′E). All the collections were made by towing nylobolt plankton net (# size 50µm) and were preserved in 5% formalin. Individual samples were screened with a Wild stereoscopic binocular microscope; the rotifers were isolated and mounted in polyvinyl alcohol–lactophenol and were observed with Leica DM 1000 stereoscopic phase contrast microscope fitted with an image analyzer. Various rotifer taxa were identified following the works of Koste (1978), Segers (1995), Sharma (1998), Sharma and Sharma (1999, 2000, 2008), and Jersabek and Leitner (2013) except for two indeterminate species warranting more specimens. The biogeographic remarks were made by vide Segers (2007), Sharma and Sharma (2017) and Jersabek and Leitner (2013). The microphotographs were provided for interesting species and measurements were indicated in micrometers (µm). The percentage similarity between the rotifer communities was calculated vide Sørensen's index (Sørensen 1948). The voucher collections are submitted to the holdings of Zoological Survey of India, Kolkata.

Results

The plankton and semi-plankton collections examined from small wetlands of the Brahmaputra River floodplains of lower and upper Assam reveal 157 species belonging to 34 genera and 18 families. A detailed systematic list of the observed taxa is presented below:

Systematic list of Rotifera recorded from small wetlands of upper and lower Assam

Phylum : Rotifera

Class : Eurotatoria

Subclass : Monogononta

Order : Ploima

Family : Brachionidae

1 *Anuraeopsis fissa* Gosse, 1851
2 *Brachionus angularis* Gosse, 1851
3 *B. bernini* Leissling, 1924
4 *B. bidentatus* Anderson, 1889
5 *B. budapestinensis* Daday, 1885
6 *B. calyciflorus* Pallas, 1766
7 *B. caudatus* Barrois & Daday, 1894
8 *B. dichotomus reductus* Koste & Shiel, 1980 #@
9 *B. diversicornis* (Daday, 1883)
10 *B. durgae* Dhanapathi, 1974
11 *B. falcatus* Zacharias, 1898
12 *B. forficula* Wierzejski, 1891
13 *B. kostei* Shiel, 1983 #@
14 *B. mirabilis* Daday, 1897
15 *B. nilsoni* Ahlstrom, 1940@
16 *B. quadridentatus* Hermann, 1783
17 *B. rubens* Ehrenberg, 1838
18 *Keratella cochlearis* (Gosse, 1851)
19 *K. edmondsoni* Ahlstrom, 1943 #
20 *K. javana Hauer*, 1937 #@
21 *K. lenzi* Hauer, 1953
22 *K. tecta* (Gosse, 1851)
23 *K. tropica* (Apstein, 1907)
24 *Plationus patulus* (O.F. Muller, 1786)
25 *Platyias leloupi* Gillard, 1967
26 *P. quadricornis* (Ehrenberg, 1832)

Family : Euchlanidae

27 *Beauchampiella eudactylota* (Gosse, 1886)
28 *Dipleuchlanis propatula* (Gosse, 1886)
29 *Euchlanis dilatata* Ehrenberg, 1832
30 *E. incisa* Carlin, 1939
31 *E. triquetra* Ehrenberg, 1838
32 *Tripleuchlanis plicata* (Levander, 1894)

Family : Mytilinidae

33 *Lophocharis salpina* (Ehrenberg, 1834)
34 *Mytilina acanthophora* Hauer, 1938
35 *M. bisulcata* (Lucks, 1912)
36 *M. brevispina* (Ehrenberg, 1830)
37 *M. michelangellii* Reid & Turner, 1988
38 *M. ventralis* (Ehrenberg, 1830)

Family : Trichotriidae

39 *Macrochaetus longipes* Myers, 1934
40 *M. sericus* (Thorpe, 1893)
41 *Trichotria tetractis* (Ehrenberg, 1830)
42 *Wolga spinifera* (Western, 1894)

Family : Lepadellidae

43 *Colurella adriatica* Ehrenberg, 1831
44 *C. obtusa* (Gosse, 1886)
45 *C. uncinata* (O.F. Müller, 1773)
46 *Lepadella acuminata* (Ehrenberg 1834)
47 *L. apsicora* Myers, 1934
48 *L. apsida* Harring, 1916
49 *L. benjamini* Harring, 1916
50 *L. biloba* Hauer, 1958
51 *L. costatoides* Segers, 1992
52 *L. dactyliseta* (Stenroos, 1898)
53 *L. desmeti Segers and Chittapun, 2001**#@*
54 *L. discoidea* Segers, 1993 #
55 *L. ehrenbergi* (Perty, 1850)
56 *L. heterostyla* (Murray, 1913)
57 *L. minuta* (Weber & Montet, 1918)
58 *L. ovalis* (O. F. Müller, 1786)
59 *L. patella* (O.F. Muller, 1773)
60 *L. quinquecostata* (Lucks, 1912)
61 *L. rhomboides* (Gosse, 1886)
62 *L. triptera* Ehrenberg, 1830
63 *L. vandenbrandei* Gillard, 1952 #@
64 *Squatinella lamellaris* (O. F. Müller, 1786)

Family	:	**Lecanidae**
	65	*Lecane aculeata* (Jakubski, 1912)
	66	*L. aeganea* Harring, 1914
	67	*L. arcula* Harring, 1914
	68	*L. bifurca* (Bryce, 1892)
	69	*L. blachei* Berzins, 1973 #
	70	*L. bulla* (Gosse, 1851)
	71	*L. calcaria* Harring & Myers, 1926 #@
	72	*L. clara* (Bryce, 1892) *#
	73	*L. closterocerca* (Schmarda, 1898)
	74	*L. crepida* Harring, 1914
	75	*L. curvicornis* (Murray, 1913)
	76	*L. decipiens* (Murray, 1913)
	77	*L. dorysimilis* Trinh Dang, Segers & Sanoamuang, 2015 #@
	78	*L. doryssa* Harring, 1914
	79	*L. elegans* Harring, 1914
	80	*L. flexilis* (Gosse, 1886)
	81	*L. furcata* (Murray, 1913)
	82	*L. haliclysta* Harring & Myers, 1926
	83	*L. hastata* (Murray, 1913)
	84	*L. hamata* (Stokes, 1896)
	85	*L. hornemanni* (Ehrenberg, 1834)
	86	*L. inermis* (Bryce, 1892)
	87	*L. inopinata* Harring & Myers, 1926
	88	*L. lateralis* Sharma, 1978 #
	89	*L. leontina* (Turner, 1892)
	90	*L. ludwigii* (Eckstein, 1883)
	91	*L. luna* (O. F. Müller, 1776)
	92	*L. lunaris* (Ehrenberg, 1832)
	93	*L. monostyla* (Daday, 1897)
	94	*L. nitida* (Murray, 1913
	95	*L. niwati* Segers, Kotethip & Sanoamuang, 2004 #@
	96	*L. obtusa* (Murray, 1913)
	97	*L. papuana* (Murray, 1913)
	98	*L. ploenensis* (Voigt, 1902)
	99	*L. pusilla* Harring, 1914
	100	*L. pyriformis* (Daday, 1905)
	101	*L. quadridentata* (Ehrenberg, 1830)
	102	*L. rhenana* Hauer, 1929@
	103	*L. rhytida* Harring & Myers, 1926@
	104	*L. signifera* (Jennings, 1896)
	105	*L. stenroosi* (Meissner, 1908)
	106	*L. stichoclysta* Segers, 1993 # @
	107	*L. thienemanni* (Hauer, 1938)
	108	*L. undulata* Hauer, 1938
	109	*L. unguitata* (Fadeev, 1925) #
	110	*L. ungulata* (Gosse, 1887)
Family	:	**Notommatidae**
	111	*Cephalodella forficula* Ehrenberg, 1830
	112	*C. gibba* (Ehrenberg, 1830)
	113	*C. trigona* (Rousselet, 1895) # @
	114	*Monommata grandis* Tessin, 1890@
	115	*M, longiseta* (O.F. Müller, 1786)
	116	*Notommata pachyura* (Gosse, 1886)
Family	:	**Scaridiidae**
	117	*Scaridium longicaudum* (O.F. Müller, 1786)
Family	:	**Trichocercidae**
	118	*Trichocerca bicristata* (Gosse, 1887)
	119	*T. bidens* (Lucks, 1912) @
	120	*T. capucina* (Wierzejski & Zacharias, 1893)
	121	*T. cylindrica* (Imhof, 1891)
	122	*T. elongata* (Gosse, 1886)
	123	*T. flagellata* Hauer, 1938
	124	*T. hollaerti* De Smet, 1990#@
	125	*T. insignis* (Herrick, 1886) @
	126	*T. longiseta* (Schrank, 1802)
	127	*T. maior* (Hauer, 1935) #
	128	*T. pusilla* (Jennings, 1903)
	129	*T. rattus* (O.F. Müller, 1776)
	130	*T. scipio* (Gosse, 1886) @
	131	*T. similis* (Wierzejski, 1893)
	132	*T. tenuior* (Gosse, 1886)
	133	*T. tigris* (O.F. Muller, 1786)
	134	*T. weberi* (Jennings, 1903)
Family	:	**Asplanchnidae**
	135	*Asplanchna brightwelli* Gosse, 1850
	136	*A. priodonta* Gosse, 1850
Family	:	**Synchaetidae**
	137	*Polyarthra vulgaris* Carlin, 1943
Family	:	**Dicranophoridae**
	138	*Dicranophorus forcipatus* (O.F. Müller, 1786)
Order	:	**Floscullariaceae**
Family	:	**Flosculariidae**
	139	*Ptygura* sp.
	140	*Sinantherina socialis* (Linne, 1758)
	141	*S. spinosa* (Thorpe, 1893)
Family	:	**Conochilidae**
	142	*Conochilus unicornis* Rousselet,1892
Family	:	**Hexarthridae**
	143	*Hexarthra mira* (Hudson, 1871)
Family	:	**Testudinellidae**
	144	*Pompholyx sulcata* Hudson,1885
	145	*Testudinella amphora* Hauer, 1938 #@
	146	*T. brevicaudata* Yamamoto, 1951#@
	147	*T. dendradena* de Beauchamp, 1955 #@
	148	*T. emarginula* (Stenroos, 1898)
	149	*T. parva* (Ternetz, 1892)
	150	*T. patina* (Hermann, 1783)
	151	*T. tridentata* Smirnov, 1931@
	152	*Testudinella* sp. @
Family	:	**Trochosphaeridae**
	153	*Filinia camasecla* Myers, 1938
	154	*Filinia longiseta* (Ehrenberg, 1834)
	155	*F. opoliensis* (Zacharias, 1898)
Subclass	:	**Bdelloidea**
Order	:	**Philodinida**
Family	:	**Philodinidae**
	156	*Dissotrocha aculeata* (Ehrenberg, 1832)
	157	*Rotaria neptunia* (Ehrenberg, 1830)

* New record from the Oriental region; ** new record from Assam;
# species of global biogeopgraphic interest;
@ species restricted to Northeast India.

Lecane clara (Bryce) is a new record (Figure 2.A) from the Oriental region (marked as *) and

Lepadella desmeti Segers and Chittapun (Figure 2.B) is a new record from Assam state (marked as**). *Brachionus dichotomus reductus* (Figure 2.C), *B. kostei* (Figure 2.D), *Cephalodella trigona, Filinia camasecla* (Figure 2.E), *Keratella edmondsoni, K. javana, Lecane blachei* (Figure 2.F), *L. calcaria* (Figure 3.A), *L. niwati* (Figure 3.B), *L. dorysimilis* (Figure 3.C), *L. lateralis, L. stichoclysta, L. unguitata, Lepadella desmeti, L. discoidea, L. vandenbrandei, Testudinella amphora, T. brevicaudata* (Figure 3.D), *T. dendradena, Trichocerca hollaerti* (Figure 3.E) and *T. maior* are species of global bigeographic interest (marked as #). *Testudinella* (Figure 3.F) sp. is yet an un-determined species awaiting description pending examination of more specimens. Our collections indicate 23 species (marked as @) with their distribution in India known to be restricted to NEI.

We report 152 and 125 rotifer species from lower and upper Assam collections, respectively (Table 1) with consistent importance of Lecanidae (45 and 42 species), Lepadellidae (21 species each) and Trichocercidae (16 and 13 species). The comparison of Rotifera species composition of the two study areas records lower richness of the Brachionidae (14 species), *Brachionus* (6 species) and paucity of species of *Keratella* and *Mytilina* especially in our collections from upper Assam wetlands (Table 1).

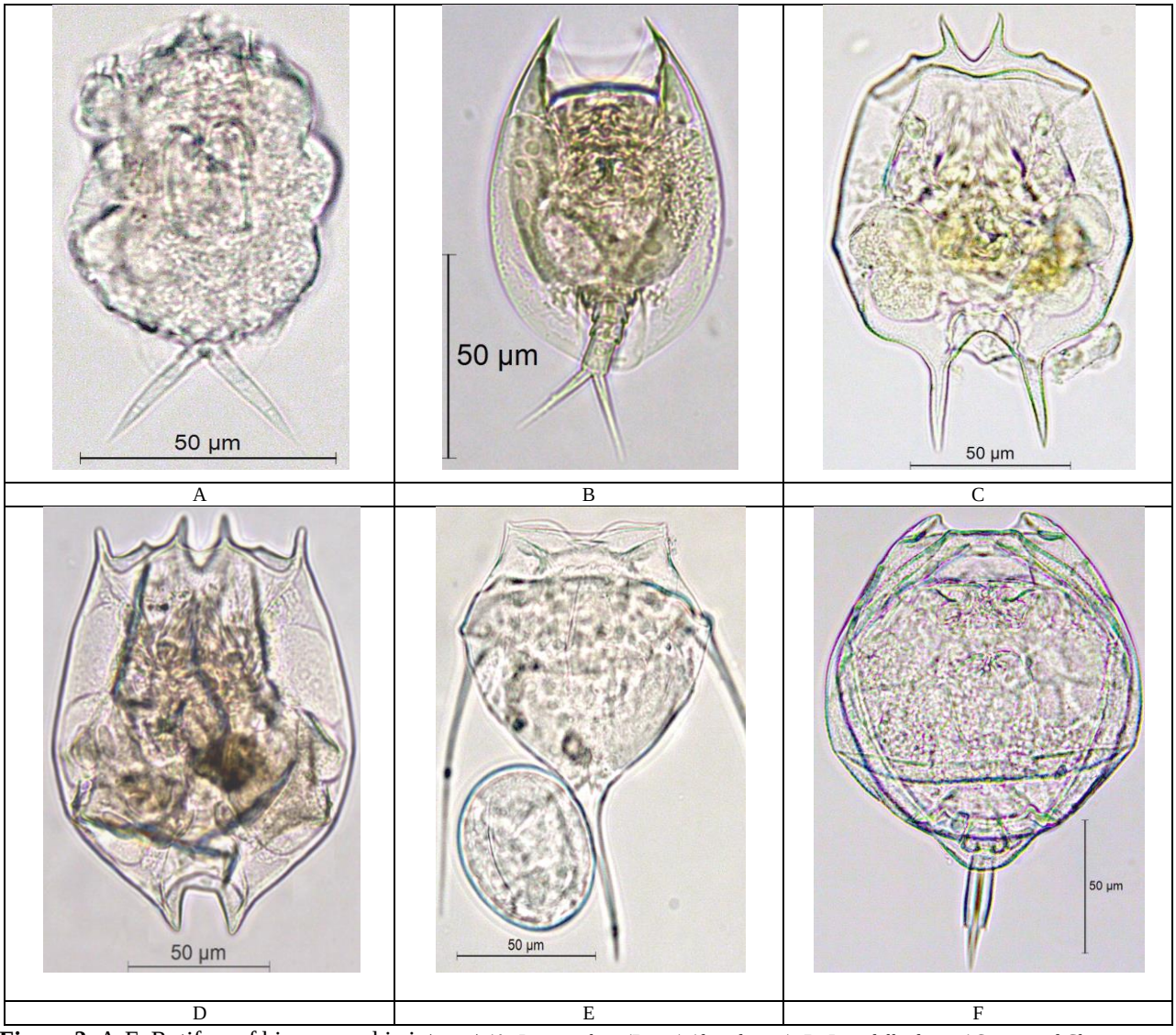

Figure 2. A-F: Rotifers of biogeographic interest (A, *Lecane clara* (Bryce) (dorsal view); B, *Lepadella desmeti* Segers and Chittapun (ventral view); C, *Brachionus dichotomus reductus* Koste & Shiel (ventral view); D, *Brachionus kostei* Shiel (ventral view); E, *Filinia camasecla* Myers with parthenogenetic egg (dorsal view); F, *Lecane blachei* Berzins (dorsal view).

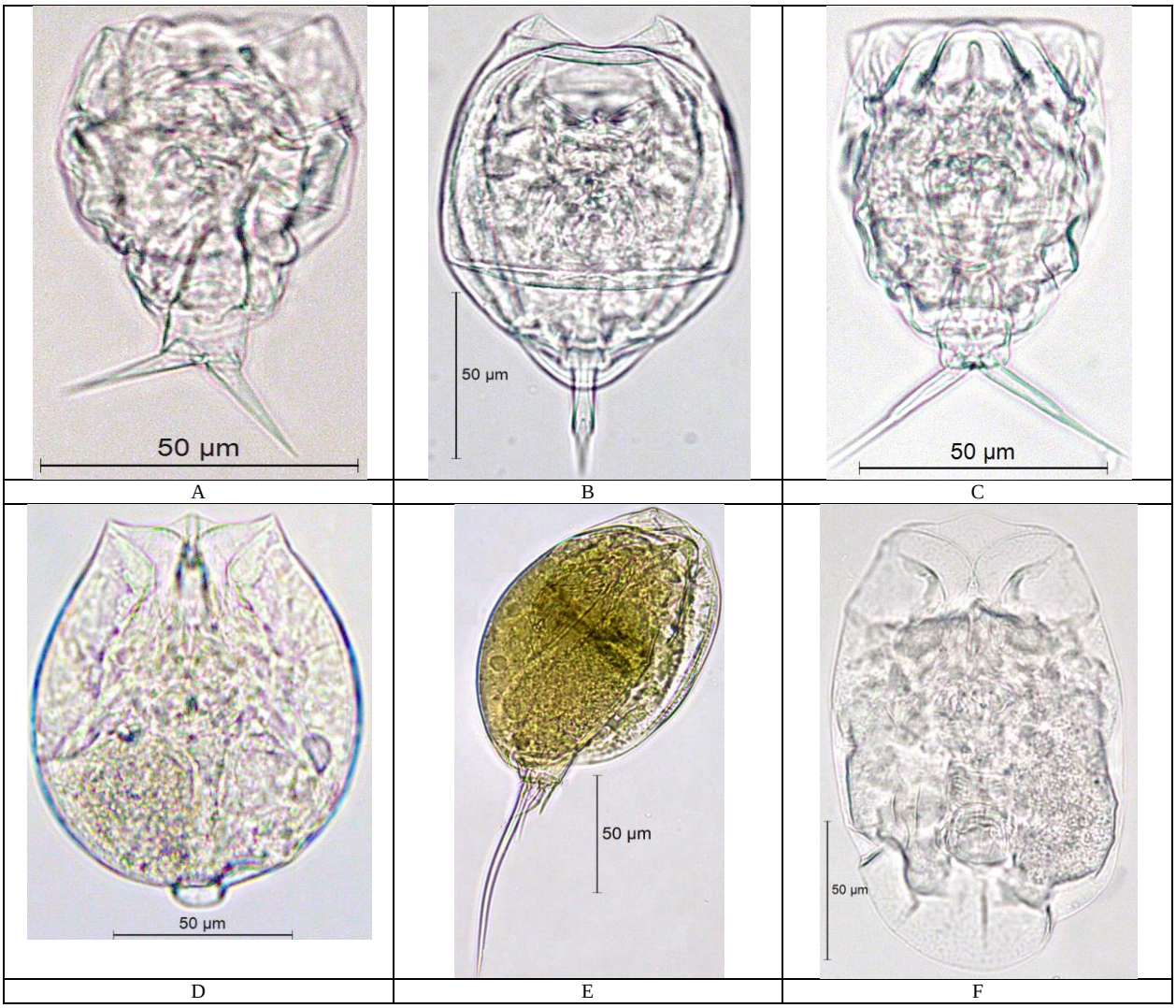

Figure 3. A-F: Rotifers of biogeographic interest (A, *Lecane calcaria* Harring & Myers (ventral view); B, *Lecane niwati* Segers, Kotethip & Sanoamuang (ventral view); C, *Lecane dorysimilis* Trinh Dang, Segers & Sanoamuang (ventral view); D, *Testudinella brevicaudata* Yamamoto (ventral view); E, *Trichocerca hollaerti* De Smet (lateral view); F, *Testudinella* sp. (ventral view)

Table 1. Comparison of Rotifera composition of small wetlands of lower and upper Assam

Taxa	This study	Lower Assam	Upper Assam
Species	157	152	125
Genera	34	34	30
Families	18	18	16
		Important families: species (%)	
Lecanidae	46 (29.3%)	45 (29.6%)	42 (33.6%)
Brachionidae	26 (16.6%)	26 (17.1%)	14 (11.2%)
Lepadellidae	22 (14.0%)	21 (13.8%)	21 (16.8%)
Trichocercidae	17 (10.9%)	16 (10.5%)	13 (10.4%)
	111 (70.8%)	108 (71.0%)	90 (72.0%)
		Other notable families: species (%)	
Testudinellidae	8	8	7
Euchlanidae	6	6	6
Notommatidae	6	5	5
Mytilinidae	6	6	3
	26 (16.6%)	25 (16.4%)	21(16.8%)
		Important genera: species (%)	
Lecane	46 (29.3%)	45 (29.6%)	42(33.6%)
Lepadella	18 (11.4%)	16 (10.5%)	17 (13.6%)
Trichocerca	17 (10.9%)	16 (10.5%)	13 (10.4%)
Brachionus	16 (10.2%)	16 (10.5%)	06 (4.8%)
	97 (61.8%)	93 (61.2%)	78 (62.4%)
		Other notable genera: species (%)	
Testudinella	7	7	6
Keratella	6	6	4
Mytilina	5	5	3
	18 (11.4%)	18 (11.8%)	13 (10.4%)

Discussion

Our collections from *dobas* or *dubies* of the Brahmaputra river basin reveal 157 species, belonging to 34 genera and 18 families; the richness comprises ~65%, ~52% and ~27% of species of Rotifera known from Assam state, NEI and India (Sharma and Sharma 2017), respectively and thus affirms biodiverse nature of the rotifer assemblage. Besides, 152 and 125 species observed from lower and upper Assam, respectively depict species-rich rotifers of the two study areas individually, while higher community similarity (85.2% vide Sørensen's index) depicts overall homogeneity in species composition but with certain differences.

Lecane clara, a new record from the Oriental region, is diagnosed by its soft lorica and characteristic elongate toes. This lecanid is yet known (Segers 2007) from the African, Neotropical. Nearctic, Pacific, and Palaearctic regions, while our report extends its distribution to the Oriental region. *Lepadella desmeti*, described from Thailand (Segers and Chittapun 2001), is known elsewhere from Neotropical and Pacific regions (Segers 2007). The only earlier record of this species from the Indian sub-region (Sharma and Sharma 2015b) relates to Loktak Lake (a Ramsar site), Manipur state; the present study further extends its distribution within NEI to Assam state. In addition, *Lecane calcaria* and *L. stichoclysta* deserve mention in view of the restricted reports from south Assam (Sharma and Sharma 2019a) and the eastern Himalayas (Sharma and Sharma 2019b).

Interestingly, Rotifera of *dobas* or *dubies* invariably record species consortia of maximum 30-35 species/sample while a few small wetlands from the Majuli River Island and the Dibru-Saikhowa Biosphere reserve of upper Assam indicate speciose constellations of up to 50 species/sample. We categorize these reports as `Rotifera paradox' following analogy to the classical `the paradox of the plankton` highlighted by Hutchinson (1961); the former, in turn, is hypothesized to the intriguing possibility of the co-existence of a number of species in `a relatively isotropic or unstructured environment of small wetlands`.

This study records biodiverse rotifers than the reports from *dobas* or *dubies* of the Majuli River Island (Sharma 2014), small lentic biotopes of Mizoram (Sharma and Sharma 2015a) and Nagaland (Sharma and Kensibo 2017, Sharma et al. 2017), and the Kashmir Himalayan floodplains (Sharma and Sharma 2018). The richness is marginally lower than the reports from small lentic biotopes of the eastern Himalayas (Sharma and Sharma 2019a) and the floodplains of Barak valley of south Assam (Sharma and Sharma 2019b). The comparisons highlight *dobas* or *dubies* of the Brahmaputra floodplains to be one of the biodiverse rotifer environs of India; this generalization is hypothesized to the function of habitat diversity of the sampled wetlands and sampling intensity vides Fontaneto et al. (2012). Interestingly, this study registers higher rotifer richness than certain global floodplain reports i.e., the Rio Pilcomayo National Park, Argentina (Jose de Paggi 2001); Oguta and Iyi-Efi lakes of the Niger delta (Segers et al. 1993) of Africa; Lake Guarana (Bonecker et al. 1994), and Lago Camaleao (Koste and Robertson 1983) of Brazil; Thale-Noi Lake, Thailand (Segers and Pholpunthin 1997); and it compares well with 151 species known from Rio Tapajos (Koste 1974) of Brazil.

Our collections reveal notable examples of global and regional biogeographic interest. The former include 21 species (13.4%) namely (i) the Australasian *Brachionus dichotomus reductus* and *B. kostei*; (ii) the Oriental endemics *Filinia camasecla, Keratella edmondsoni, L. blachei* and *L. niwati*; (iii) the paleotropical *K. javana, Lecane lateralis, L. stichoclysta, L. unguitata, Lepadella discoidea, L.vandenbrandei, Testudinella brevicaudata* and *Trichocerca hollaerti*; (iv) the Indo-Chinese *Lecane dorysimilis*; (v) the Holarctic *Trichocerca maior*; and (vi) five other species namely *Cephalodella trigona, Lecane calcaria, Lepadella desmeti, Testudinella amphora* and *T. dendradena*. The Australian elements affirm affinity of the rotifer assemblage with Southeast Asian and Australian faunas, while other categories impart affinities with Southeast Asian Rotifera (Sharma and Sharma 2014, 2017). In addition, 23 species (~15%) are known for their Indian distribution till date restricted to NEI, while ~12% species namely *Brachionus durgae, Colurella adriatica, Lecane aeganea, L. doryssa, L. elegans, L. haliclysta, L. hastata, L. pusilla, L. thienemanni, Lepadella benjamini, L. dactyliseta, L. discoidea, L. quinquecostata, Macrochaetus longipes, Mytilina michelangellii, Platyias leloupi, Testudinella parva, Trichocerca flagellata* and *Wolga spinifera* depict regional distribution interest vis-à-vis the Indian Rotifera (Sharma and Sharma 2017). We hypothesize overall biodiverse rotifer assemblage of *dobas* or *dubies* of the Brahmaputra river floodplains and occurrence of sizable fractions of specie of biogeographic interest to impact of `the Assam-gateway'- a vital biogeographic corridor of India that facilitated extensive interchanges between the Indian and Asian biota (Mani 1974), thus changing the modern biotic composition of the

epigean ecosystems of India into one of 'predominantly Oriental' nature (Ranga Reddy 2013).

Lecanidae > Brachionidae > Lepadellidae > Trichocercidae, collectively form large fraction (~71%) of the rotifer species known vide this study as well as from lower Assam wetlands. On the other hand, upper Assam wetlands indicate corresponding cumulative importance of these Eurotatoria families (72%) but are characterized by a notable paucity of Brachionidae (14 species). Testudinellidae, Euchlanidae, Notommatidae, and Mytilinidae also deserve attention for collective contributions of 16.4% and 16.8% from lower and upper Assam wetlands, respectively. The 'tropic centered' *Lecane* is most speciose genus both of lower (45 species) and upper Assam (42 species) rotifers. *Lepadella* = *Trichocerca* = *Brachionus* are notable in lower Assam wetlands (31.5%), while *Lepadella* > *Trichocerca* > *Brachionus* (29.8%) deserve attention in upper Assam wetlands but with a distinct paucity of *Brachionus* spp. (6 species; 4.8%). In general, the paucity of the brachionid Rotifera of upper Assam wetlands, concurs with the reports from the floodplains of the Majuli River Island (Sharma 2014) and our results from the states of Meghalaya (Sharma and Sharma 1999), Mizoram (Sharma and Sharma 2015a), Nagaland (Sharma et al. 2017), and Arunachal Pradesh (Sharma and Sharma 2019a) of NEI. Further, the collections from upper Assam are notable for relative paucity of *Trichocerca*, *Keratella* and *Mytilina* species; rare occurrence of *Asplanchna* and *Filinia* species and lack of species of Conochilidae and Hexarthridae. The richness of important Eurotatoria families and genera assigns the littoral-periphytonic nature to Rotifera of *dobas* or *dubies* of the Brahmaputra floodplains, in conformity with the reports from the floodplains of Africa (Segers et al. 1993; Green 2003), Brazil (Koste 1974; Koste and Robertson 1983), Thailand (Segers and Pholpunthin 1997), Argentina (Jose De Paggi 2001) and NEI (Sharma and Sharma 2008, 2014). The high richness of 'tropic centered' *Lecane* and that of *Brachionus* in lower Assam wetlands**,** large fraction of cosmopolitan species (~67%) and occurrence of several (~20%) pantropical and cosmotropical species imparts 'tropical character' to the rotifer assemblages of *dobas* or *dubies* following the reports on the tropical rotifer faunas (Fernando 1980; Segers 2008; Sharma and Sharma 2008, 2014, 2017, 2019a).

To sum up, this study is an important contribution to Rotifera biodiversity of small lentic habitats of India, Asia, and tropics and subtropics. The biodiverse rotifers affirm habitat and environmental heterogeneity of *dobas* or *dubies* of the Brahmaputra floodplains. `Rotifera paradox` hypothesizes niche diversification enabling co-existence of several species within an unstructured environment of small wetlands. The diverse species composition is attributed to the location the sampled study areas within the key biodiversity area of the Indo-Burma Hotspot as well as the historical influence of `the Assam-gateway' facilitating incursion of species from Asian and the Oriental faunas. Such studies need to be extended to other small lentic wetlands of NEI and elsewhere in India to explore ecosystem diversity value of small water bodies vs. biodiversity of Indian Rotifera.

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Aşağı Melet Irmağı (Ordu, Türkiye)'nda Yaşayan *Barbus tauricus* Kessler, 1877 Otolit Kütle Asimetrisinin Belirlenmesi

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ÖZ

Kemikli balıklar, otolit kütle asimetrisinin fizyolojik rolünü değerlendirmede en uygun organizmalardır. Asimetri, balığın vestibular ve işitsel fonksiyonları üzerinde şiddetli etkilere sebep olan bir faktördür ancak bu konuyla ilgili sınırlı sayıda çalışma bulunmaktadır. Türkiye sularındaki tatlı su türlerinin asimetrileri hakkında birkaç çalışma vardır. Bu çalışmanın amacı, *Barbus tauricus* Kessler, 1877'un otolit kütle asimetrisini belirlemektir. Balık örnekleri Temmuz 2010-Ekim 2011 tarihleri arasında serpme ağlar kullanılarak aşağı Melet Irmağı'ndan yakalanmıştır. *B. tauricus* bireylerinin sağ ve sol otolitleri arasındaki fark, paired-t testine göre istatistiksel olarak önemli bulunmuştur ($P<0,05$). Ortalama x ve $|x|$ değerleri 0,0685±0,0194 ve 0,2377±0,0135 olarak hesaplanmıştır. Balık boyu-x, balık boyu-$|x|$ ve balık boyu-(M_R-M_L) arasındaki ilişkiler sırasıyla $y=0,0011x+0,0434$; $y=0,0066x+0,1457$ ve $y=0,0000008x+0,00003$ olarak tespit edilmiştir. Otolit kütle asimetrisi değeri, balıkların yaşadıkları ortamla ilişkili olarak ağır metaller, pestisit, tarım ilaçları gibi kirlilik etkenleri, stres oluşturan faktörler, suyun fiziko-kimyasal özelliklerinde meydana gelebilecek değişimler hakkında bilgi verebilir. Diğer tatlı su türlerinde yapılan çalışmalarla karşılaştırıldığında, ortalama otolit kütle asimetrisi değerinin daha yüksek olarak belirlenmesi, ırmağın *B. tauricus* bireyleri üzerindeki çevresel etkisini de ortaya çıkarmıştır.

Anahtar kelimeler: Otolit, kütle asimetrisi, Cyprinidae, *Barbus tauricus*, Aşağı Melet Irmağı.

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Determination of Otolith Mass Asymmetry in *Barbus tauricus* Kessler, 1877 Inhabiting Lower Melet River (Ordu, Turkey)

Abstract: Teleost fishes are the most suitable organisms to assess the physiological role of otolithic mass asymmetry. The asymmetry is a factor which caused drastic effects on vestibular and auditory functions of fish. However, there are limited studies on this subject. There are few studies about the asymmetry of freshwater species in Turkish waters. The aim of this study is to determine otolith mass asymmetry of *Barbus tauricus* Kessler, 1877. Fish samples were caught from the Lower Melet River by using cast nets between July 2010 and October 2011. The difference between right and left otoliths of *B. tauricus* was statistically significant according to paired-t test result ($P<0.05$). The mean values of x and $|x|$ were calculated as 0.0685±0.0194 and 0.2377±0.0135. The relationships between fish length and x, fish length and $|x|$ and fish length and *MR-ML* determined as $y=0.0011x+0.0434$; $y=0.0066x+0.1457$, and $y=0.0000008x+0.00003$, respectively. The otolith mass asymmetry value can give information about pollution factors such as heavy metals, pesticides, stress factors, changes in the physico-chemical properties of water in relation to the environment in which fish habitat. Compared with other fresh water studies, the higher mean autolithic mass asymmetry value revealed the environmental impact of the river on *B. tauricus* individuals.

Keywords: Otolith, mass asymmetry, Cyprinidae, *Barbus tauricus*, Lower Melet River.

Alıntılama
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Giriş

Otolitler üç çift halinde, kalsiyum karbonat yapısında olan ve kemikli balıklarda iç kulakta bulunan kemiksi yapılardır. Türe özgü farklı morfolojilere sahip olan otolitler, balık boyu, beslenme, stres gibi farklı ekolojik faktörlerden etkilenmektedir. Aynı zamanda, balık türlerinin yaşam döngüsünü karakterize eden birçok olayı çalışmak amacıyla en çok kullanılan kemiksi yapılardır (Merigot vd. 2007). Yaş ve büyüme, taksonomi, şekil analizleri, av-avcı ilişkileri ve otolit kütle asimetrisi gibi birçok çalışma için otolitlerden faydalanılmaktadır.

Otolitler, hem deniz hem de tatlı su balık türleri için kolaylıkla ulaşılabilen kemiksi yapılardır. Otolitlerin kolayca elde edilebilmeleri, otolit kütle asimetrisinin balık üzerindeki fizyolojik rolünü belirlemede kullanılacak kemikli balıkları, uygun organizmalar durumuna getirmektedir (Lychakov vd. 2006). Genel olarak, balıklarla yapılan çalışmalarda sagitta ve asteriskus otolitleri incelendiğinde, deniz balıklarında sagitta otolitlerinin en büyük otolit çifti olduğu ve dolayısıyla en büyük kütleye sahip olduğu görülürken, tatlı suda yaşayan Cyprinidae türlerinde asteriskus otolitlerinin kütlesinin daha büyük olduğu görülmektedir (Secor vd. 1992).

Sağ ve sol otolit çiftleri arasındaki kütle farkından kaynaklanan otolit kütle asimetrisi, balıklardaki vestibüler boşluk bozukluklarının oluşumunda etkili olan önemli faktörlerden biri olduğu belirtilmektedir (Lychakov ve Rebane 2004). Asimetri, balıkların işitsel ve vestibüler fonksiyonları üzerinde güçlü değişimlere sebep olmakla birlikte (Lychakov vd. 2006), balıkların otolitlerinde kütle asimetrisi oluşması durumunda, genellikle bireylerin davranışlarında önemli değişikliklere yol açmaktadır (Lychakov ve Rebane 2004; Jawad 2013). Bu güne kadar konuyla ilgili olarak, tatlı su ve deniz balıklarında az sayıda çalışma yapılmış ve bu balık türlerinde bilgi elde edilmiştir (Lychakov vd. 2006; Jawad vd. 2010; Jawad vd. 2011; Jawad 2013; Jawad ve Sadighzadeh 2013; Bostancı vd. 2017b; Kontaş vd. 2017; Yedier vd. 2017; Yedier vd. 2018b).

Balık türlerinin sagitta ve asteriskus otolitleri, ülkemizin hem deniz hem de tatlı su habitatlarında yaşayan balık türlerinde, farklı tekniklerin kullanıldığı çeşitli amaçlarla yapılan çalışmalarda ana materyal olarak kullanılmıştır. Bunlardan bazıları şu şekilde özetlenebilir: a) balık boyu-otolit ölçümleri arasındaki ilişkilerin hesaplanmasında [*Barbus tauricus* (Kontaş ve Bostancı 2015), *B. grypus* (Düşükcan vd. 2015), *Alburnus* spp. (Tsagarakis vd. 2015), *Garra rufa* (Yedier vd. 2016), *Diplodus puntazzo* (Bostancı vd. 2016), *Atherina boyeri* (Bostancı vd. 2017a), *B. grypus* (Düşükcan ve Çalta 2018) ve *Capoeta trutta* (Düşükcan 2018)]; b) tür ayrımında *Alburnus* spp. (Bostancı vd. 2015); c) otolit morfolojisinde (Assis 2003; Tuset vd. 2008); d) stok ve popülasyon ayrımında *Atherina boyeri* (Bostancı ve Yedier 2018; Yedier vd. 2019). Deniz balıklarındaki otolit asimetrisi çalışmalarının (Lychakov vd. 2006; Jawad vd. 2010; Jawad vd. 2011; Jawad 2013; Jawad ve Sadighzadeh 2013; Bostancı vd. 2018; Kontaş vd. 2018; Yedier vd. 2018a, 2018b) yanı sıra, tatlı sularda yapılan çalışmalar da vardır (Lychakov vd. 2006; Bostancı vd. 2017b; Kontaş vd. 2017; Yedier vd. 2017).

Balık türleri, yaşadıkları sucul sistemlerin çevresinde yer alan tarım alanlarından gelen pestisit, tarım ilaçları, evsel, endüstriyel ve kentsel atıklara bağlı ağır metal ve diğer kirleticilerden etkilenir. Bunun sonucunda ise, bu etkiyi fizyolojik ve davranışsal olarak gösterirler. Bu çalışmanın amacı; maruz kaldıkları doğal ve antropojenik stresin önceden tayininde de kullanışlı bir metot olarak değerlendirilen otolit kütle asimetrisi (x) ve mutlak otolit kütle asimetrisini ($|x|$), Ordu ili içme su kaynağı olarak da kullanılan Melet Irmağı'nda yayılış gösteren *B. tauricus* türünde belirlemektir. Ayrıca türün total boyu ile otolit kütle asimetrisi, mutlak otolit kütle asimetrisi ve otolit kütle farkı (M_R-M_L) değerleri arasındaki ilişkiler de araştırılmıştır.

Materyal ve Metot

Bu araştırmada, Melet Irmağı (Ordu) üzerinde yer alan Topçam Barajı'nın alt bölgelerinde kalan aşağı havzasında yaşayan *B. tauricus* (n=269) örnekleri Temmuz 2010-Ekim 2011 tarihleri arasında serpme ağlar kullanılarak balıkçılar yardımıyla yakalanmıştır (Şekil 1). Yakalanan örnekler, Ordu Üniversitesi Fen-Edebiyat Fakültesi Hidrobiyoloji Laboratuvarı'na getirilmiş ve analizleri yapılana kadar derin dondurucuda saklanmıştır. Balık bireylerinin total boyları ±1 mm ve ağırlıkları ±0.1 g hassasiyetle ölçülmüştür. Diseksiyonları yapılan örneklerin, sağ ve sol asteriskus otolitleri dikkatli bir şekilde çıkarıldıktan sonra temizlenerek analizlere kadar muhafaza edilmiştir. Hesaplamalar kırılmış parçası olmayan sağ ve sol otolit çiftlerinde yapılmıştır (Şekil 2).

Sağ ve sol otolitlerin ağırlıkları Precisa XB220A marka hassas terazide ±0,0001 g hassasiyetle tartılmıştır. Balıkların sağ ve sol otolit ağırlıkları arasındaki farkın araştırılması amacıyla paired-t testi kullanılmıştır. Otolit değişkenlerinin tanımlayıcı istatistiklerinin belirlenmesi ve paired-t testi için MİNİTAB 17.0 istatistik programı kullanılmıştır.

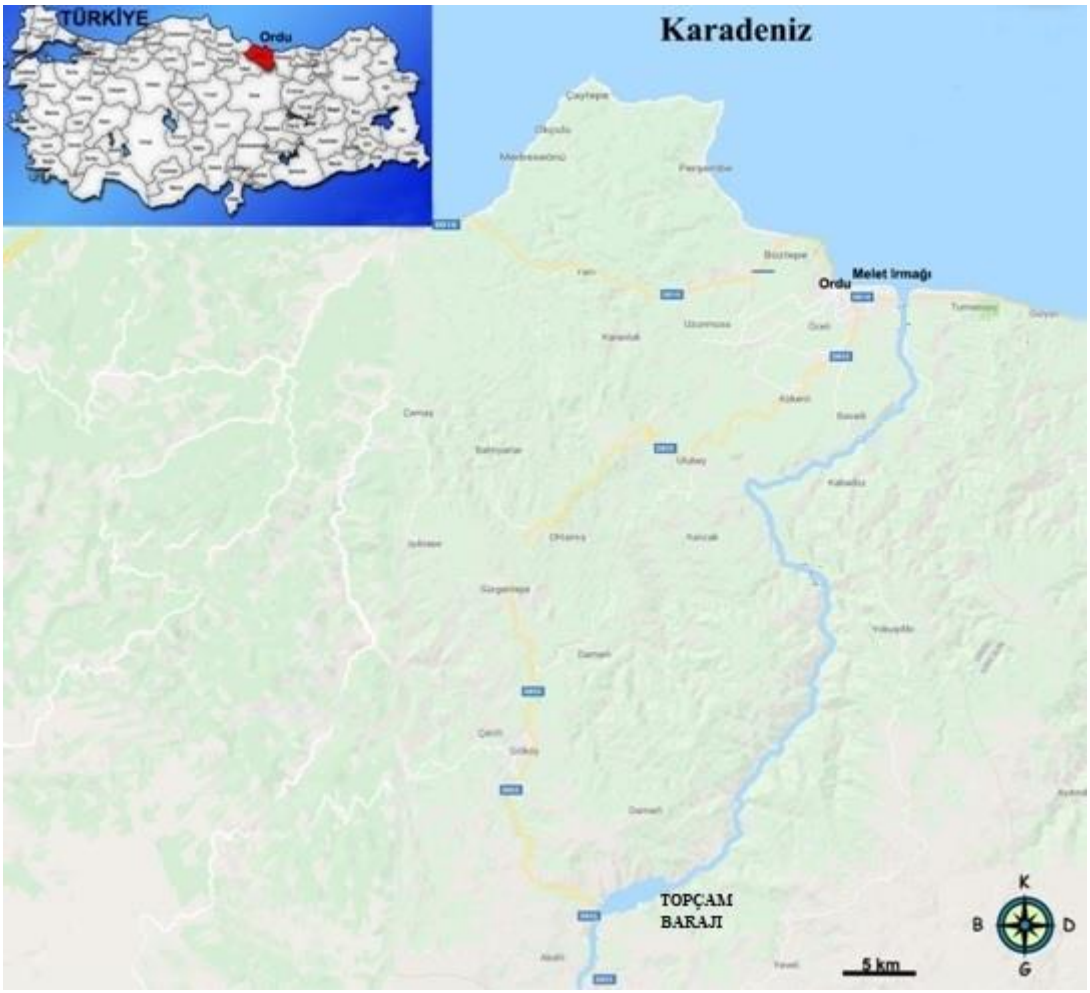
Otolit kütle asimetrisi (x), sağ ve sol otolitlerin kütlesi arasındaki farkın ortalama otolit kütlesine bölünmesiyle hesaplanır. Otolit kütle asimetrisi (x) değerinin hesaplanmasında;

$$x=(M_R-M_L)M^{-1}$$

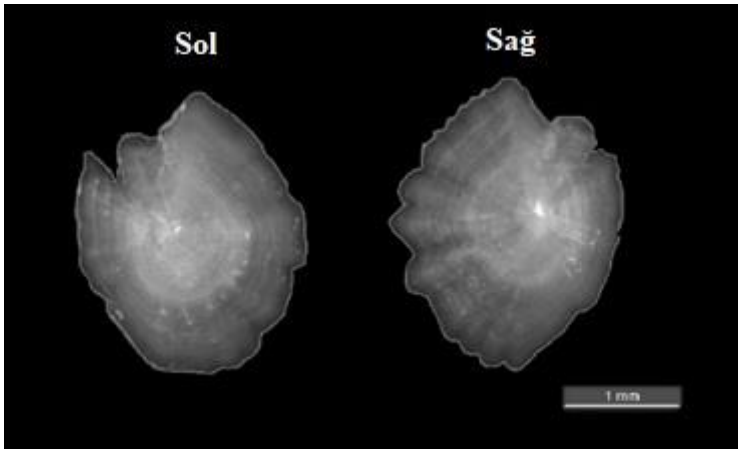
formülünden yararlanılmıştır. Formülde, M_R ve M_L değerleri sırasıyla sağ ve sol otolitlerin ağırlıklarını, M ise tüm otolitlerin ortalama ağırlığını ifade etmektedir. Mutlak otolit kütle asimetrisi ($|x|$) değeri, *B. tauricus* bireylerinde hesaplanan x değerlerinin ortalaması alınarak hesaplanmıştır. Türün total boyu ve otolit kütle asimetrisi (x) değeri arasındaki ilişki;

$$m=a\ x\ l+b$$

formülü kullanılarak hesaplanmıştır. Formülde, "l"; balığın total boyu, "a"; otolitin karakteristik büyüme oranı katsayısı ve "b"; denklemdeki tür için bir sabittir. Otolit kütle asimetrisi değeri (x), balık türleri arasında teorik olarak -0,2 ve +0,2 değerleri arasındadır. "0" değeri kütle asimetrisinin olmadığını ifade eder ($M_R=M_L$) (Lychakov vd. 2006). Ayrıca, balık boyu ve otolit kütle farkı (M_R-M_L) arasındaki ilişki de hesaplanmıştır.



Şekil 1. Melet Irmağı'nın genel görünüşü.



Şekil 2. *B. tauricus*'un asteriskus otolit çiftinin genel görünüşü

Bulgular

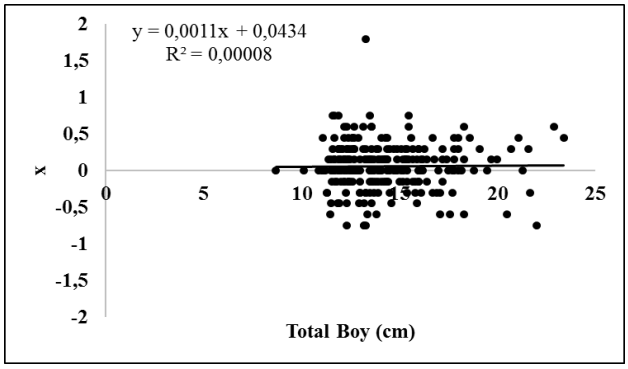
Bu çalışmada, Melet Irmağı'ndan örneklenen *B. tauricus* örneklerinin total boyları 8,7-23,4 cm, ağırlıkları ise 4,03-122,83 g aralığında değişmektedir (Kontaş 2012). Otolit kütle asimetrisi çalışılan ve bir tatlı su balığı olan *B. tauricus*'un asteriskus otolitleri 0,0001-0,0021 g aralığında değişim göstermektedir. Sağ ve sol otolit ağırlıkları karşılaştırıldığında, sağ otolitlerin sol otolitlerden daha büyük olduğu ve yapılan paired-t testi sonucunda aralarındaki kütle farkının da istatistiksel olarak önemli olduğu ($P<0,05$) belirlenmiştir (Tablo 1).

Yapılan hesaplamalar sonucunda, Melet Irmağı'nda yaşayan *B. tauricus* türünün, ortalama otolit kütle asimetrisi (x) değeri 0,0685±0,0194 ve ortalama mutlak otolit kütle asimetrisi ($|x|$) değeri 0,2377±0,0135 olarak tespit edilmiştir. Sağ ve sol otolitlerin arasındaki kütle farkının ortalama değeri ise 0,000046±0,000013 olarak hesaplanmıştır (Tablo 1).

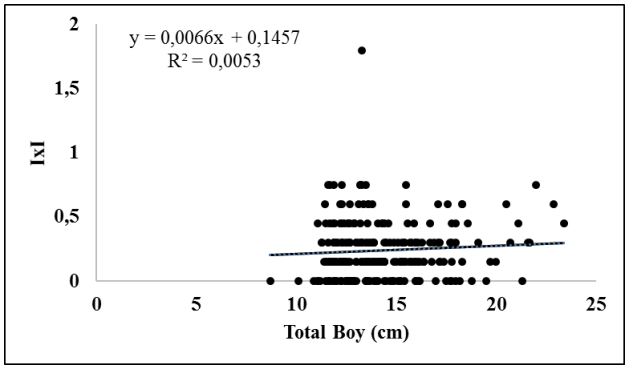
Tablo 1. *B. tauricus*'un otolit değişkenlerinin tanımlayıcı istatistikleri (n=269)

Değişkenler	Ortalama ± SH	Minimum - Maksimum	P Değeri
Sağ Otolit Ağırlığı (g)	0,000691 ± 0,000025	0,0001 - 0,0021	P<0,05
Sol Otolit Ağırlığı (g)	0,000645 ± 0,000024	0,0001 - 0,0020	
X	0,0685 ± 0,0194	-0,7487 - 1,7969	
$|x|$	0,2377 ± 0,0135	0,0000 - 1,7969	
(M_R-M_L)	0,000046 ± 0,000013	-0,0005 - 0,0012	

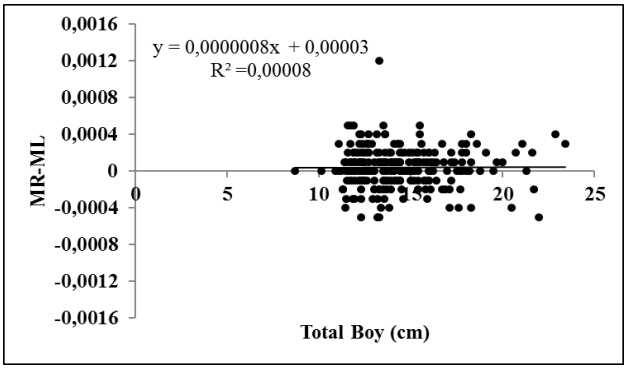
Regresyon analizi grafiği incelendiğinde, *B. tauricus* türü için total boy ile otolit kütle asimetrisi arasında bir ilişki bulunmadığı belirlenmiştir (*y=0,0011x+0,0434*, R^2=0,00008) (Şekil 3). Aynı şekilde, balık boyu ile mutlak otolit kütle asimetrisi ($|x|$) arasında da bir ilişki olmadığı tespit edilmiştir (*y=0,0066x+0,1457*, R^2=0,0053) (Şekil 4). Balık boyu ile otolit kütle farkı (M_R-M_L) arasında belirlenen ilişkinin denklemi de *y=0,0000008x+0,00003* (R^2=0,00008) olarak hesaplanmıştır (Şekil 5).



Şekil 3. Total boy ile otolit kütle asimetrisi (x) değeri arasındaki ilişki.



Şekil 4. Total boy ile mutlak otolit kütle asimetrisi ($|x|$) değeri arasındaki ilişki.



Şekil 5. Total boy ile otolit kütle farkı (M_R-M_L) arasındaki ilişki.

Tartışma ve Sonuç

Otolit kütle asimetrisi (x) değeri, genel olarak -0,2 ve +0,2 arasındadır (Lychakov vd. 2006). Yaptığımız çalışmada, *B. tauricus* için bu değer -0,7487 ile 1,7969 aralığında hesaplanmıştır. Bireysel olarak düşünüldüğünde, *B. tauricus* türü için hesaplanan otolit kütle asimetrisi (x) değeri, genel olarak balık türleri için beklenen sonuçlara benzer şekilde -0,2 ile +0,2 değerleri arasında tespit edilmemiştir. Ancak, ortalama otolit kütle asimetrisi değeri (x=0,0685) belirtilen aralıkta hesaplanmıştır. Diğer taraftan, birçok deniz balığı türünde otolit kütle asimetrisi 0,05 değerinden daha düşük hesaplanmıştır (Lychakov vd. 2006) ve hesaplanan x değeri de büyüme oranına bağlı değildir (Jawad ve Sadighzadeh 2013). Daha önce yapılan çalışmalarda hesaplanan ortalama mutlak otolit kütle asimetrisi değerleri incelendiğinde (Tablo 2), Lychakov vd. (2006)'nın bazı tatlı su türlerinde hesapladıkları mutlak otolit kütle asimetrisi değerleri *Carassius auratus* için 0,03209 ± 0,00634; *Cyprinus carpio* için 0,03615 ± 0,00361 olarak tespit edilmiştir. Bostancı vd. (2017b) ise Melet Irmağı'nda yaşayan *Capoeta*

banarescui türü için ortalama mutlak otolit kütle asimetrisi değerini 0,08558 ± 0,00322 olarak belirlemişlerdir. Tatlı su balıklarında hesaplanan *x* değerinin *B. tauricus*'ta daha yüksek olduğu göze çarpmaktadır. Lychakov vd. (2006), balığın kulağındaki akustik ve vestibular fonksiyonun kütle asimetrisinden dolayı azalabileceğini ve bu durumun daha önceki matematiksel modellemelerle gösterildiğini bildirmişlerdir. Aynı zamanda, balık türlerindeki otolit kütle asimetrisinin nispeten daha yüksek değerleri akustik fonksiyonu değiştirebilmekte ve anormal balık davranışlarına sebep olabilmektedir (Lychakov ve Rebane 2004, 2005). Ayrıca, Lychakov ve Rebane (2005)'nin çalışmalarında belirtildiği gibi, başın her iki tarafındaki otolitler arasında oluşan kütle farkı nedeniyle, otolitler arası hareket uyumsuzluğu ve uyuşmazlıktan dolayı sesin alınması ve dengede zorluk yaşanabileceği bildirilmiştir.

Tablo 2. Deniz ve tatlı su habitatlarındaki bazı balık türlerinde ortalama mutlak otolit kütle asimetrisi (|*x*|) değerleri.

	n	Lokasyon	Ortalama |*x*| ± SH	Kaynaklar
Deniz Türleri				
Merluccius merluccius	27	Akdeniz	0,09326 ± 0,0607	Lychakov vd. 2006
Pagellus erythrinus	18	Akdeniz	0,02374 ± 0,0063	Lychakov vd. 2006
Sciaenops ocellatus	196	Meksika Körfezi	0,03161 ± 0,0029	Lychakov vd. 2006
Beryx splendens	32	Umman Denizi	0,0177 ± 0,0142	Jawad vd. 2010
Rhynchorhamphus georgi	124	Umman Denizi	0,3529 ± 0,0081	Jawad vd. 2011
Carangoides caeruleopinnatus	150	Umman Denizi	0,0886 ± 0,2418	Jawad 2013
Liza klunzingeri	30	Basra Körfezi	0,16667 ± 0,0142	Jawad ve Sadighzadeh 2013
Solea solea	50	İskenderun Körfezi	0,04301 ± 0,0050	Yedier vd. 2018b
Solea solea	50	Mersin Körfezi	0,04558 ± 0,0079	Yedier vd. 2018b
Tatlı Su Türleri				
Carassius auratus	45	Japon iç suları	0,03209 ± 0,0063	Lychakov vd. 2006
Cyprinus carpio	103	Japon iç suları	0,03615 ± 0,0036	Lychakov vd. 2006
Poecilia reticulata	27	Japon iç suları	0,05797 ± 0,0095	Lychakov vd. 2006
Alburnus chalcoides	25	Curi Irmağı	0,1135±0,0577	Kontaş vd. 2017
Alburnus mossulensis	130	Munzur Nehri	0,07745±0,0098	Yedier vd. 2017
Capoeta banarescui	236	Melet Irmağı	0,08558 ± 0,0032	Bostancı vd. 2017b
Barbus tauricus	269	Melet Irmağı	0,2377 ± 0,0135	Bu çalışma

Bilateral simetrili kemikli balıklarda, otolit kütle asimetrisi değerinin düşük seviyede olması asteriskus otoliti için tipiktir (Lychakov vd. 2006). Yaptığımız çalışmada, tatlı suda yaşayan bilateral simetrili kemikli bir balık türü olan *B. tauricus*'a ait ortalama otolit kütle asimetrisi değerinin 0,0685 olduğu tespit edilmiştir (Tablo 1). Aynı akarsuda yaşayan *C. banarescui* türünde otolit kütle asimetrisini değerlendirmek amacıyla yapılan bir diğer çalışmada bu değer -0,00803 olarak hesaplanmıştır (Bostancı vd. 2017b). Aynı bölgede yaşayan farklı iki türe ait ortalama otolit kütle asimetrisi değerleri incelendiğinde, *x* değerinin *B. tauricus*'da *C. banarescui* türünden daha yüksek olduğu görülmektedir. Bu sonuçlar, otolit kütle asimetrisi değerinin asteriskus otoliti için düşük değerlerde olduğunu ve türler arasında değişiklik gösterdiğini belirtmektedir. Bunun sebebi, farklı türlerin çevresel kirlilik, stres, rekabet gibi faktörlerden farklı şekillerde etkilenmesidir. Aynı zamanda, -0,2 ile +0,2 değerleri arasında olması beklenen ortalama *x* değerinin, *B. tauricus* türünde de sınır değerler arasında olduğu ve yapılan diğer çalışmalarla uygunluk gösterdiği görülmektedir.

Önceki çalışmaların sonuçları değerlendirildiğinde, otolit kütle asimetri ve mutlak otolit kütle asimetrisi değerlerinin, balıkların büyümesi sırasında değişiklik gösterdiği sonucuna varılmıştır (Lychakov ve Rebane 2004, 2005). Melet Irmağı'nda yaptığımız çalışmanın sonuçları da bu durumla uygunluk göstermektedir. Yaptığımız bu çalışmada da *B. tauricus* için hem otolit kütle asimetrisi hem de mutlak otolit kütle asimetrisinin total boyla ilişkili olmadığı tespit edilmiştir. Bununla beraber, balığın total boyu ile otolitler arasındaki kütle farkı ilişkisi, total boy ile otolit kütle asimetrisi

arasındaki ilişkiden daha kompleks bir yapıya sahip olduğu ifade edilmiş ve otolit kütle farkının (M_R-M_L), deniz balıklarında balığın boyuyla arttığı bildirilmiştir (Lychakov vd. 2006). Tatlı su balığı olan *B. tauricus*'ta sağ-sol otolitler arasındaki ağırlık farkı ile total boy arasında anlamlı bir ilişki olmadığı belirlenmiştir. Araştırmanın sonuçlarına göre, balık boyu ve otolit kütle farkı arasında da bir ilişki olmadığı, türün otolit kütle farkının total boya bağlı olarak artmadığı belirlenmiştir.

Balıklarda otolit kütle asimetrisi, bireysel olarak balıkların büyümesi esnasında değişebilir. Melet Irmağı'nda yaşayan *B. tauricus* türünün bireysel anlamda otolit kütle asimetrisi değerleri -0,7487 ile 1,7969 değerleri arasında hesaplamıştır. Bu da boy aralığı 8,7-23,4 cm arasında değişen bireylerin oluşturduğu bir örneklemede, bireysel otolit kütle asimetrisinin değişiklik gösterdiğini açıkça belirtmektedir. Bununla birlikte, çoğu balık türünde gözlemlenen, sağ ve sol otolitler arasındaki nispi kütle farkının % 10 - 20 aralığında olduğudur. Yapılan bir çalışmada, deniz balıklarında sağ ve sol otolitler arasındaki nispi kütle farklarının çok büyük veya çok küçük olduğu belirli balık türlerine rastlanmadığı bildirilmiştir (Lychakov ve Rebane 2004). Tatlı su türleri olan *Carassius aurata* ve *Cyprinus carpio*'da (Lychakov vd. 2006) asteriskus otolit çiftleri arasındaki ortalama fark ise sırasıyla 0,03035 ve 0,03491 olarak bulunmuştur. *B. tauricus* için bu değer 0,000046 olarak hesaplanmıştır ve oldukça küçük bir değer olarak göze çarpmaktadır. Bu farklılığın farklı tatlı su habitatlarındaki fiziko-kimyasal yapılarının birbirinden farklı oluşundan, çalışılan türlerin genetik farklılıklarından ve bu habitatlarda yaşayan türlerin farklı ekolojik nişlere sahip olmalarından kaynaklı olduğu düşünülmektedir.

Orta ve Doğu Karadeniz bölümleri arasında doğal bir sınır oluşturan ve coğrafik öneme sahip olan Melet Irmağı'nda yapılmış bir çalışmada (Kontaş 2018), ırmağın doğal jeolojik konumunun, maden yataklarınca zengin bir bölgede oluşundan ve fındık tarımının yapıldığı arazilerde kullanılan pestisitlerden, ağır metaller ve diğer kirleticilerden dolayı tehdit altında olduğu bildirilmiştir. Bu bölgeden örneklenen *B. tauricus* türüne ait balık bireylerinde belirlenen ortalama mutlak otolit kütle asimetrisi değeri (0,2377) bu etkiyi göstermektedir. Otolit kütle asimetrisi değerinin bilinmesi hem çalışılan bu tür için hem de diğer balık türleri için maruz kaldıkları doğal ve antropojenik stresin önceden tayininde de kullanışlı bir metot olarak görülmektedir.

Aynı zamanda farklı türlerin her biri için de bu ilişkilerin araştırılması, yaşadıkları ortamla ilişkili olarak ağır metaller, pestisit, tarım ilaçları gibi kirlilik etkenlerinin, stres oluşturan faktörlerin, suyun fiziko-kimyasal özelliklerinde meydana gelebilecek değişimlerin önceden tespit edilmesinde kolayca faydalanılabilecek bir metot olarak görülmektedir.

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Morphological Diversification of *Mesocyclops leuckarti* (Claus, 1857)

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ABSTRACT

In this study, different populations of a copepod species, *Mesocyclops leuckarti*, were compared with their body size. Samples were taken from Asi River, Mehmetli Dam Lake, Yenişehir Lake, and Tahta Köprü Dam Lake, locating in east-south part of Turkey. Whole sexual dimorphism for the populations was changed from 1.56 to 1.62. Intra-population variability on body size by locations were changed as 3.85 % to 5.05 %. Intra-population variability was bigger in male than that of female populations in each water body. The same pattern was observed for inter-population variability among male populations (8.46 %) and female populations (8.04 %). Discriminant analysis (*DFA*) and *SIMPER* (Similarity Percentage) methods revealed that Cephalozom Length (*CL*) measurementwas the most distinguished measure leading discriminate among the male populations with 23.97 %. Abdomen (*ABD*) measurement was the most discriminative measure among female populations with 30.86 %. Based on the *MANOVA*, the differentiation in body size among the female populations was very significant ($p<0.001$), contrary to male populations ($p>0.05$).The Minimal Spanning Tree (*MST*) analysis showed that the specimen living in the pond and lake systems were closer than river systems in terms of body size diversification especially for female ones of copepods.

Keywords: Copepoda, *Mesocyclops leuckarti*, body size, variability, sexual dimorphism

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Mesocyclops leuckarti (Claus, 1857) nin Morfolojik Çeşitliliği

Öz: Bu çalışmada, bir kopepod türü olan *Mesocyclops leuckarti* nin vücut büyüklüğü farklı popülasyonları arasında karşılaştırıldı. Örnekler Türkiye nin Güney Doğusunda yer alan Asi Nehri, Mehmetli Baraj Gölü, Yenişehir Gölü ve Tahta Köprü Baraj Gölü'nden alındı. Popülasyonları için genel eşeysel dimorfizim 1,56 ile 1,62 arasında değişti. Lokasyonlara göre, popülasyonlar içi vücut büyüklüğündeki değişim % 3,85 ile % 5,05 arasında değişmiştir. Her bir su kütlesinde erkeklerin popülasyonlar içi değişimleri dişilere göre daha fazla idi. Aynı model erkek popülasyonları (% 8,46) arasında ve dişi popülasyonları (% 8,04) arasında da gözlemlenmiştir. Diskriminant analizi (*DFA*) ve *SIMPER* (Benzerlik Yüzdesi) yöntemleri, Cephalozom Uzunluk (*CL*) ölçümünün, erkek popülasyonları arasında % 23,97 ile ayrışmaya yol açan en belirgin ölçüm olduğunu ortaya koydu. Abdomen (*ABD*) ölçümü dişi popülasyonlar içinde % 30,86 ile en fazla ayrışmaya yol açan ölçüm idi. *MANOVA*' ya göre, dişi popülasyonları arasında vücut büyüklüğündeki farklılaşma, erkek popülasyonların aksine ($p>0,05$) çok anlamlıydı ($p <0,001$). Minimal Yayılma Ağacı (*MST*) analizi, vücut büyüklüğü farklılaşması açısından özellikle kopepod' ların dişileri için, gölet ve göl sistemlerinde yaşayan bireylerin nehir sistemlerinde yaşayan bireylere göre birbirlerine daha yakın olduğunu göstermiştir.

Anahtar kelimeler: Copepoda, *Mesocyclops leuckarti*, vücut büyüklüğü, değişkenlik, eşeysel dimorfizim

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Introduction

Animal species distributed over wide geographic areas are usually split into subspecies. According to current theory, geographic variation and isolation are important factors in the process of speciation (Elgmork and Halvorsen 1998). Therefore, expecting the influence of geographic isolation on any dynamics of species, including on body size, may reasonable. There is growing recognition that both inter-specific and intra-specific variations can have significant effects on population, community, and ecosystem dynamics. Morphological differences are likely a key component of this ecologically important variation (Hausch et al. 2013). As a fundamental

morphological parameter, body size largely determines species' functional and environmental characteristics, growth, life duration, population density, and species' place in food webs (Anufriieva and Shadrin 2014).

The Cyclopoida is the most species–rich group among copepod lineages and comprises, the largest group within the subclass Copepoda. Members of the copepods are important components of pelagic ecosystems. Several cyclopoid copepod species among the genus *Mesocyclops* have invaded different places of the world, each showing distinct distributional patterns (Reid and Saunders 1986; Reid and Pinto-Coelho 1994; Hribar and Reid 2008; Suárez-Morales et al. 2011).

Morphometric variability of some copepod species was studied in different water bodies in the world (Hausch et al. 2013; Anufriieva and Shadrin 2014; Anufriieva and Shadrin 2015) and the size of copepods is affected by a number of environmental factors and varies widely (Anufriieva and Shadrin 2015). However, little effort has been made, for copepods or any other taxonomic groups, to quantify morphological variation among populations relative to that among species. It was expressed as unexpected that both inter-specific and intra-specific diversity has important effects on species coexistence and ecosystem dynamics. While intra-specific diversity is maintained both within and between populations, comparisons of inter to intra-specific diversity appear to have focused exclusively on comparing diversity between species to that within local populations (Hausch et al. 2013).

The aim of our study, is to determine both inter-population and intra-population diversity on body size, as an indicator of morphological differentiation, of *M. leucarti* from the different water bodies, locating in the Southeast Region of Turkey. We analyzed both variability by partition the morphological variation into components by sexes and populations.

Materials and Methods

Sampling

Zooplankton samples were taken from four different localities in Turkey. *M. leuckarti* were collected from Tahta Köprü Dam Lake (36°52'22.19" K, 36°41'20.31" D, for 12 months), Mehmetli Dam Lake (37°30'31.79" K, 36°01'09.37" D, on the 7th of October 2009, 16th of July 2010, and 13th of April 2010), Yenişehir Lake (36°14'12.17" K, 36°34'08.38" D, for12 months in 2003 and 2004) and Asi River (36°13'00.54" K, 36°09' 44.42" D, for 12 months in 2005 and 2006) with horizontal and vertical draws by using 60 μm mesh size plankton net. Samples were replaced into a glass jar and fixed with 4% formaldehyde. Then, the selected adult *M. leuckarti* specimens were put in ethanol due to health concerns of formaldehyde. The copepod specimen examination, counting, and measurements were done by using an Olympus CH40 microscope and a micrometric ocular. The taxonomic literature were used to identify the zooplankton specimen (Scourfield and Harding 1966); Dussart 1969; Kiefer and Fryer 1978). Temperature (°C) and dissolved oxygen concentration (*DO, mgL^{-1}*) were measured in the field with a thermometer and a YSI 52 model oxygen meter, respectively.

Morphological measurement

Copepod body length was measured under the binocular microscope to the nearest 0.01 mm at a magnification of 10x and 40x. It was taken from the tip of the prosome to the end of the caudal rami, including the extremely long furcal setae. Some individuals were slightly bent due to fixation. Therefore, each specimen was placed laterally between 2 movable cover slips in a small droplet of lactophenol, which softened the exoskeleton (Böttger-Schnack 1989). By carefully moving the cover slips together, the specimens were straightened, and their total length could be measured. On the other hand, the width and length of each thorax segment were measured at its widest point.

Table 1. Measured distances (measurements) on the body with their corresponding abbreviations.

Measured Distance (measurement)	Abbreviation
Cephalozom Length	*CL*
Cephalozom-Cross	*CC*
Cephalozom- Width	*CW*
Thorax-1- Width	*TW1*
Thorax-1- Length	*TL1*
Thorax-2- Width	*TW2*
Thorax-2- Length	*TL2*
Thorax-3- Width	*TW3*
Thorax-3- Length	*TL3*
Thorax-4- Width	*TW4*
Thorax-4- Length	*TL4*
Abdomen	*ABD*
Furca- Width	*FW*
Furca- Length	*FL*
Total Length	*TL*

Table 2. The codes of populations with their number of samples (n)

Populations	Codes	N
Asi Female	*ASI-FM*	19
Asi Male	*ASI-ML*	10
Tahta Köprü Female	*TAHTA-FM*	20
Tahta Köprü Male	*TAHTA-ML*	10
Mehmetli Female	*MEH-FM*	20
Mehmetli Male	*MEH-ML*	10
Yenişehir Female	*YENİ-FM*	20
Yenişehir Male	*YENİ-ML*	10

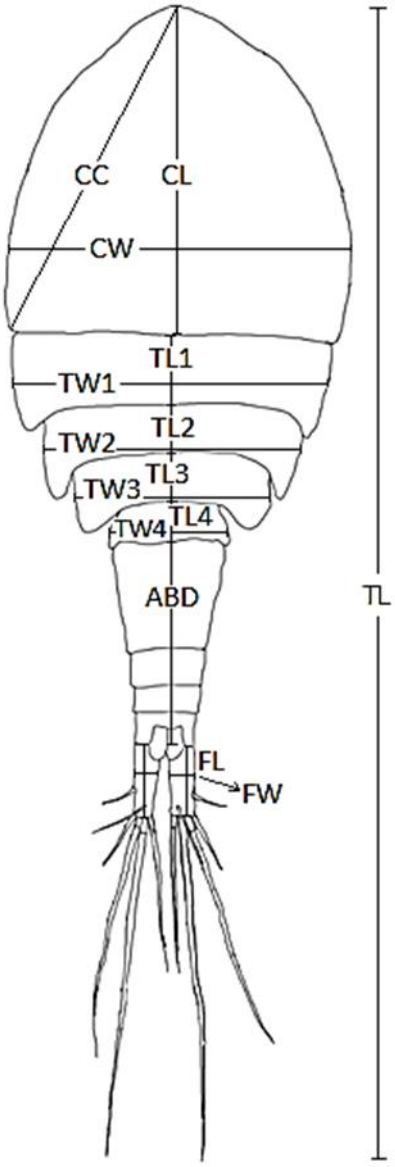

Figure 1. Measurement applications on the body of copepod

Data Analysis

In this study, two species have been studied in four water systems, so 8 populations have been formed. Before starting the main data analysis, every measurement was checked for outliers and missing values by simply plotting the data as xy pairs.

Inter and intra-population diversity were assessed in terms of coefficient of variation (*CV*, %), discriminate function analysis (*DFA*), *SIMPER* (Similarity Percentage), *MANOVA* (Multivariate Analysis of Variance) and, the Minimal Spanning Tree (*MST*) analysis.

Variability in each measured distances or measurements was qualified by the coefficient of variation (*CV*, %). Then, to find an overall variability value for any population, mean *CV* value calculated as fallowing;

$$CV_{Overall} = \frac{\sum CV_i}{n}$$

Where, CV_i: is the *CV* value of the i'th measurement and n: is the number of measurements.

The best model for the standardization of the morphometric data was the regression of Elliott et al. (1995). This model removes the size component from the shape measurements (allometry). Due to sexual dimorphism, standardization was applied for both sexes separately. The model is defined by the following equation:

$$Ms = Mo\left[\frac{Ls}{Lo}\right]^b$$

Where, *Ms* = standardized measurement, *Mo* = measured character length (mm), *Ls* = overall (arithmetic) mean standard length (mm) for all individuals from all populations of each sex, *Lo* = standard length (mm) of specimen, and "*b*" was estimated for each character from the observed data using the non-linear equation,

$M = a\,L^b$.

The standardized morphometric values of the populations were analyzed within each sex and among sexes and compared by means of discriminate function analysis (*DFA*). This multivariate analysis allowed us to determine which combinations of variables (distances) discriminated best among populations and detected which populations were the most different (Ruiz-Campos et al. 2003).

Along with the *DFA*, *SIMPER* (Similarity Percentage) was used for assessing which measurements are primarily responsible for an observed difference between groups of samples or populations (Clarke 1993).

MANOVA (Multivariate Analysis of Variance) was used to test the overall differences among the populations without separating sexes.

Generally, the level of sexual dimorphism is evaluated by the ratio of female to male length (Anufriieva and Shadrin 2014). In this study, as in case for *CV*, an overall sexual dimorphism value for each population have been calculated. To compare the rates of sexual dimorphism among the water systems one-way *PERMANOVA* analysis was used.

The Minimal Spanning Tree (*MST*) analysis finds the shortest possible set of lines connecting all points (Dussert et al. 1987). Therefore, it was used to detect link or links among the populations. All calculations and statistical analysis were conducted using MS Excel and *PAST* software (Paleontological statistics, Version 3.20) (Hammer et al. 2001).

Results

Some properties of studied locations

Temperature range mean dissolved oxygen level, fish presence, kind of environment, altitude, surface area, and the maximum depth of the studied areas were given into *MTS* analysis graph (Figure 4)**.**

Inter and Intra-population variability (diversity)

Some descriptive statistics of measured distances on the body of *M. leuckarti* specimen from the 8 populations were given in Table 3 and Table 4. Based on the these values, the overall variability within (intra-population diversity) *ASI-ML*(Asi-male), *ASI-FM* (Asi-female), *MEH-ML* (Mehmetli-male), *MEH-FM* (Mehmetli-female), *TAHTA-ML* (Tahta Köprü-male), *TAHTA-FM* (Tahta Köprü-female),*YENI-ML* (Yenişehir-male) and *YENI-FM* (Yenişehir-female) populations were calculated as 4.16 %, 3.85 %, 5.08 %, 4.34 %, 4.56 %, 3.65 %, 5.14 % and 4.70 %, respectively. Considering sexes, intra-population variability was bigger in male than that of female populations in each water body. Also, inter-population diversity among male populations was (8.46 %) bigger than inter-population variability (diversity) among female populations (8.04 %). In inter-population diversity among male populations, the Furca Width-*FW* (12.44 %), Abdomen-*ABD* (11.98 %) and Thorax4 Width-*TL4* (10.05 %) measurements were having three the biggest coefficient of variation (*CV*), whereas for female populations they were Thorax Width-*TL4* (11.66 %), Thorax2 Length-*TL2* (10.36 %) and Thorax3 Length-*TL3* (9.40 %) (Table 3 and Table 4).

Table 3. Some descriptive statistics of measured distances on the body of *M. leuckarti* specimen and Sexual Dimorphism rate by populations and locations.

		ASI-ML	***ASI-FM***	***MEH-ML***	***MEH-FM***	***TAHTA-ML***	***TAHTA-FM***	***YENİ-ML***	***YENİ-FM***
		n:10	**n:19**	**n:10**	**n:20**	**n:10**	**n:20**	**n:10**	**n:20**
	Mean	57.50	87.89	57.75	90.95	50.30	83.10	53.30	82.45
TL	*Sd*	1.18	3.86	1.96	1.73	1.16	2.40	1.95	2.39
	Mean	38.40	61.08	41.40	63.75	37.40	60.08	35.45	56.05
CL	*Sd*	1.07	2.32	1.17	1.37	0.74	2.07	1.38	1.96
	Mean	41.50	66.79	43.20	66.65	38.40	61.85	37.90	59.00
CC	*Sd*	0.71	1.99	1.62	1.53	0.66	1.46	0.74	1.81
	Mean	32.10	59.47	33.15	57.95	28.60	52.40	28.60	49.28
CW	*Sd*	0.74	1.98	1.81	1.00	0.70	1.50	1.35	1.27
	Mean	30.60	54.05	30.45	52.20	26.55	47.08	27.10	45.28
TW1	*Sd*	0.84	2.09	0.50	1.40	0.60	1.26	1.10	1.59
	Mean	11.30	18.24	10.35	17.28	9.60	16.13	10.00	14.73
TL1	*Sd*	0.59	0.73	0.75	0.82	0.61	0.76	0.00	1.06
	Mean	25.55	43.97	25.05	41.55	21.85	38.08	22.40	35.50
TW2	*Sd*	0.60	1.01	1.38	0.99	0.75	0.83	1.35	1.39
	Mean	10.55	16.11	8.80	15.28	9.60	14.23	9.25	12.70
TL2	*Sd*	0.37	0.83	0.79	0.97	0.84	0.77	0.42	0.70
	Mean	20.80	35.05	20.00	31.85	17.65	29.95	18.00	28.85
TW3	*Sd*	0.42	0.83	1.05	0.88	0.63	0.72	0.82	1.17
	Mean	8.20	12.95	7.90	11.45	8.10	11.55	6.90	10.78
TL3	*Sd*	0.63	0.96	0.74	0.89	0.74	0.51	0.52	0.70
	Mean	12.95	21.95	12.20	19.93	11.40	18.83	11.65	18.25
TW4	*Sd*	0.37	0.60	0.42	0.73	0.39	0.61	0.47	0.70
	Mean	4.90	6.84	5.05	5.90	4.60	5.80	4.55	6.08
TL4	*Sd*	0.57	0.55	0.44	0.72	0.39	0.52	0.37	0.59
	Mean	35.20	58.50	38.85	48.80	29.15	48.98	31.85	50.58
ABD	*Sd*	1.62	2.74	1.62	1.68	1.78	2.54	1.94	2.74
	Mean	2.95	4.00	3.00	4.30	2.55	4.00	2.30	3.90
FW	*Sd*	0.16	0.00	0.00	0.30	0.16	0.00	0.26	0.21
	Mean	8.35	13.76	7.30	11.55	7.05	12.00	7.45	11.68
FL	*Sd*	0.47	0.39	0.48	0.48	0.16	0.49	0.50	0.44
Sexual Dimorphism		1.61		1.55		1.62		1.56	

Table 4. Variation Coefficients (*CV, %*) of measurements by locations and sexes.

	ASI-ML	*ASI-FM*	*MEH-ML*	*MEH-FM*	*TAHTA-ML*	*TAHTA-FM*	*YENİ-ML*	*YENİ-FM*	*FM- CV*	*ML-CV*
TL	2.05	4.39	3.40	1.90	2.31	2.89	3.65	2.90	5.13	6.40
CL	2.80	3.80	2.84	2.15	1.97	3.44	3.90	3.50	5.63	6.36
CC	1.70	2.98	3.75	2.30	1.71	2.36	1.95	3.06	5.86	6.02
CW	2.30	3.33	5.46	1.72	2.44	2.86	4.72	2.58	8.02	7.81
TW1	2.76	3.87	1.63	2.68	2.25	2.68	4.06	3.50	7.93	7.09
TL1	5.19	4.02	7.22	4.74	6.40	4.71	0.00	7.18	9.40	8.12
TW2	2.34	2.29	5.52	2.37	3.42	2.18	6.03	3.90	8.59	8.14
TL2	3.50	5.13	8.96	6.33	8.78	5.41	4.59	5.48	10.36	9.40
TW3	2.03	2.37	5.27	2.75	3.55	2.42	4.54	4.06	8.01	8.00
TL3	7.71	7.38	9.34	7.75	9.11	4.42	7.48	6.47	9.40	10.60
TW4	2.85	2.73	3.46	3.67	3.46	3.26	4.07	3.82	7.86	6.00
TL4	11.58	8.10	8.67	12.17	8.57	9.02	8.11	9.73	11.66	10.05
ABD	4.60	4.69	4.16	3.45	6.11	5.19	6.10	5.42	8.98	11.98
FW	5.36	0.00	0.00	6.96	6.20	0.00	11.23	5.26	5.79	12.44
FL	5.68	2.81	6.62	4.19	2.24	4.06	6.67	3.75	8.09	8.55
Overall *CV*	4.16	3.85	5.08	4.34	4.56	3.65	5.14	4.70	8.04	8.46

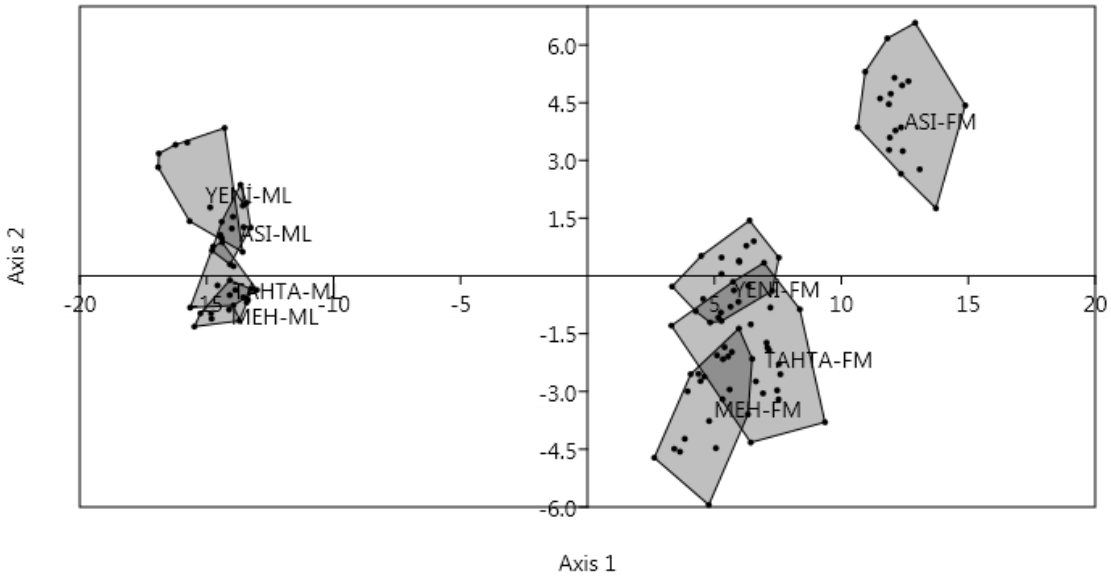

Figure 2. *DFA* results for *M. leucarti* populations.

Based on the *DFA* analysis, two main groups, one was located on the left X-axe (male) and other was located on right X-axe (female) of *DFA* graph, were clearly observed. In *DFA* the first two functions (axes) were accounted for 98.49 % variance (Figure 2).

The number of the re-assigned specimen based on Jackknife estimation procedure (group assignment) were shown into a confusion matrix (Table 5). In that matrix, specimens were reorganized by leave one out cross validation. For example, 14 out of 20 specimens belong to *MEH-FM* were remained or re-assigned (*RS*) by Jackknife estimation procedure again into the *MEH-FM* populations. So, the ratio of *RS* (n: 14) to sampled number (*T*, n: 20) for *MEH-FM* was 70 % (14*100/20). By the same approach, the ratio of *RS* to *T* for the populations were (%); *ASI-FM:* 100, *MEH-FM:* 70, *TAHTA-FM:* 70, *YENI-FM:* 85, *ASI-ML*: 90, *MEH-ML:* 80, *TAHTA-ML:* 40, *YENI-ML:* 60.

MANOVA analysis showed that there are no significant differences ($p>0.01$) among the male populations; whereas, there is statistical significance ($p<0.01$) among the female populations with changing p-values (Table 6). In accordance with the *DFA* and the Jackknife procedure the *ASI-FM* was found as the most different populations among others (Figure 2, Table 5).

The contribution of measurements to discriminate for all populations, among female populations and among male populations were given in Figure 3. Based on *SIMPER* analysis the most

distinctive measurements contribute the diversity for all populations were CW (19.32 %), *CC* (17.92 %), *CL* (16.13 %); for female populations; *ABD* (31.95 %), *CW* (12.25 %), *TW1* (12.10 %), *CC* (9.87 %) and for male populations; *CL*- (23.97 %), *ABD* (17.75 %), *CC* (14.45 %), *CW* (11.61 %) and so on (Figure 3).

Table 5. The confusion matrix for *M. leucarti* populations based on the Jackknife estimation procedure.

Populations	***ASI-FM***	***ME-FM***	***TAHTA-FM***	***YENİ-FM***	***ASI-ML***	***MEH-ML***	***TAHTA-ML***	***YENİ-ML***	***Total (T)***
ASI-FM	19								19
MEH-FM		14	6						20
TAHTA-FM		3	14	3					20
YENİ-FM		1	2	17					20
ASI-ML					9		1		10
MEH-ML						8	2		10
TAHTA-ML					3	3	4		10
YENİ-ML					2		2	6	10
Remained specimen (n)-RS*	19	14	14	17	9	8	4	6	119
Re-assignment (n)	19	18	22	20	14	11	9	6	119
Ratio of *RS* to *T* (%)**	100	70	70	85	90	80	40	60	

* *RS* refers to number of specimens that remained after *DFA* and then Jackknifed assignment for each population. For example, 14 out of 20 specimens belong to *MEH-FM* were remained or re- assigned by Jackknife estimation procedure again into the *MEH-FM* population.

Ratio of *RS* to *T* refers to ratio of *RS* to the number of total specimen (*T*) before *DFA* and Jackknife procedure for every populations.**

Table 6. *MANOVA* analysis for all populations

	Populations with *P* values*							
	ASI-FM	***MEH-FM***	***TAHTA-FM***	***YENİ-FM***	***ASI-ML***	***MEH-ML***	***TAHTA-ML***	***YENİ-ML***
ASI-FM		0.000	0.000	0.000	0.000	0.000	0.000	0.000
MEH-FM	0.000		0.001	0.000	0.000	0.000	0.000	0.000
TAHTA-FM	0.000	0.001		0.000	0.000	0.000	0.000	0.000
YENİ-FM	0.000	0.000	0.000		0.000	0.000	0.000	0.000
ASI-ML	0.000	0.000	0.000	0.000		0.193	0.715	0.443
MEH-ML	0.000	0.000	0.000	0.000	0.193		0.844	0.263
TAHTA-ML	0.000	0.000	0.000	0.000	0.715	0.844		0.428
YENİ-ML	0.000	0.000	0.000	0.000	0.443	0.263	0.428	

* $p<0.05$ indicates a significant difference, $p>0.05$ indicates non-significant difference among the populations.

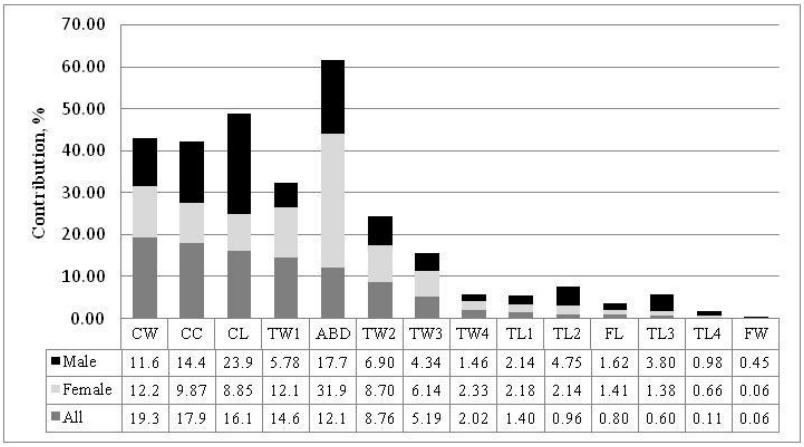

Figure 3. The contribution of measurements to discriminate for all populations, among female populations, and among male populations.

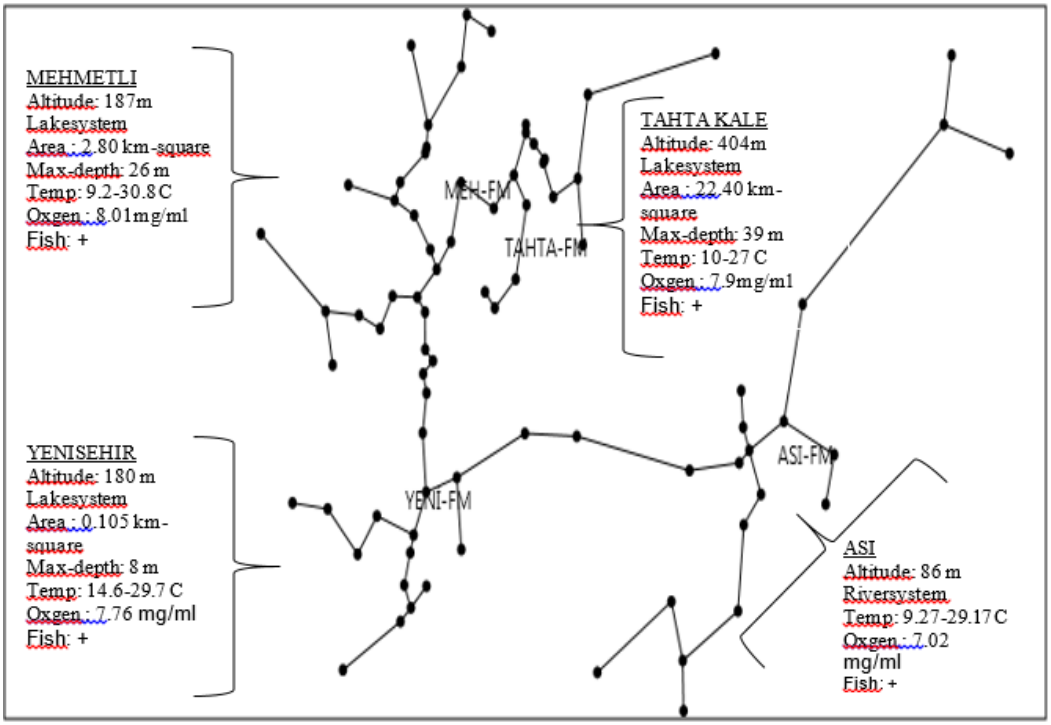

Figure 4. *MST* analysis graph with some properties of studied locations.

The Minimal Spanning Tree (*MST*) analysis for females showed that in general *ASI-FM* population links with *YENI-FM*, *MEH-FM* links with *TAHTA-FM* populations, *YENI-FM* links with *MEH-FM* and *TAHTA-FM* (Figure 4). To understand the factors effecting on the diversity of body size the *MST* graph was combined with some properties of four water systems. Since there were no statistical differences among the male populations, the *MST* analysis was not conducted for male populations.

Sexual dimorphism in body size

In all water systems, male copepods were smaller than females in overall size. The mean rate of sexual dimorphism for the populations of *ASI*, *MEH*, *TAHTA*, and *YENI* were 1.61, 1.55, 1.62 and 1.56, respectively (Table 3), and there was no significant difference among them (One-way *PERMANOVA*: $F = 1.03$, $p = 0.3932$).

Discussion

There were both intra-specific and inter-specific variations in the measured distances of the different populations for each sex of *M. leuckarti*. At intra-population level, these measurements were also having different variable patterns for all populations. But, the overall variations in all male populations were bigger than that of the female populations for each water systems (Table 4). Interestingly, the three consecutive measurements namely *TL4*, *ABD*, and *FW* had the highest *CV* values among male populations, and other three consecutive measurements having the highest *CV* values among male populations were *TL2*, *TL3*, and *TL4*.

It was found that there is inconstant variability on body size in all populations. In addition, *DFA* analysis (Figure 2) shows explicitly two different groups, namely female and male populations, most probably due to sexual dimorphism, which is discussed later in detail, observed in copepod species.

The inter-population variability was assessed with *DFA*, *MANOVA* and *MST* analyses. From those analyses, there is a phenomena which there was no significant differences among the male populations in terms of morphological diversification. Although *DFA* and Jackknife estimation procedures showed that there was specimen assignment tendency with changing numbers among the female populations, except *ASI-FM*, all female populations were statistically different from each other. In all analyses, it was clearly seen that *ASI-FM* population was very different from other female populations as well ($p<0.0000$).

In that study, the question has been emerged that why only significance differences were observed among female populations In order to evaluate the morphological difference between populations, some properties of water systems were examined. These properties were temperature range and mean dissolved oxygen, fish presence, kind of environment, altitude, surface area and maximum depth (Figure 4).

Anufriieva and Shadrin (2014), explained the constant linear dimensions and the level of their variation in *A. salinus* populations as to a certain extent they depended on temperature, salinity, and density of population. Their results lead to the

general conclusion that the impacts of factors on linear morphological characteristics and their variability can manifest itself in different ways at intra-population and inter-population levels. They also stated that in copepods, variability can increase when animals are near limit values of factors, such as temperature, salinity or increased population density, but, inter-population differences of *A. salinus* cannot be explained by only studied factors, and it is assumed that there are some overlooked factors as well as differences in the genetic architecture of populations.

Fifty-two freshwater planktonic copepod populations, *Cyclops scutifer* G.O. Sars, from Eurasia and North America, were studied to detect variations in morphology by means of morphometric analysis. It was revealed that many morphological relations were correlated with environmental factors such as depth, temperature, and trophic condition. Therefore, it was suggested that variations in body proportions were related to environmental factors rather than geographic distance (Elgmork and Halvorsen 1998).

Hausch et al**.** (2013), stressed that a wide variety of environmental gradients, including nutrient levels, temperature conditions, and predation pressure are likely to influence on the relationships between copepod body size and lake size and location. They also expressed that from studies of copepod body size along altitudinal and latitudinal gradient it is apparent that copepods are generally smaller at higher temperatures.

It was found that zooplankton body size decreases with temperature, increasing with latitude, elevation, and lake depth and decreases with visual predation threat, which is also expected to decrease with lake depth due to the presence of piscivores and a larger deep-water refuge (Hausch et al. 2013).

In most Calanoida species, other than very few exceptions, females are always bigger than males. The smaller size of calanoid males is generally attributed to their shorter developmental span of copepodite stage, which enables males to fertilize females as soon as molting. In zooplankton, the smaller size and the reduced feeding activity of males could maximize female fecundity by decreasing intra-specific competition for food. In unpredictable environments where generalism is favored, the sexual size-dimorphism may represent a way to widen the ecological niche of the species (Anufriieva and Shadrin 2014).

Gilbert and Williamson (1983) emphasized that copepods inhabit a remarkable diversity of habitats that range from small temporary pools to the abyssal depths of the ocean. The wide range of environmental conditions undoubtedly contributes to the variability in the taxon's patterns of sexual dimorphism. There are several possible adaptive advantages of having larger females than males. Perhaps the most obvious one is that females have a greater investment in offspring, both in terms of biomass contributed and, in those species that carry their eggs, of energy used to carry them.

The mean manifestation level of sexual dimorphism (mean ± *SD*, range: min-max) observed in family cyclopoida, considering 16 generas and 86 species, was 1.48 ± 0.367 (range: 0.72-3.36), and the difference in sexual dimorphism levels was explained as a function of variation in the environmental temperature or food conditions under which the distinct generations of multi-voltine populations develop (Gilbert and Williamson 1983)**.**

For a copepod species, *Arctodiaptomus salinus* (Daday, 1885), populations in the Crimean water bodies, the average index of sexual dimorphism was1.11 (1.00−1.3, *CV*=7.51), whereas, those of "small" and "large" sized populations were 1.10 and 1.13, respectively. Data obtained from different periods of a single lake (Lake Yanyshskoye) showed that this index could vary widely within a population. Sexual dimorphism of *A. salinus* manifests not only in linear dimensions and proportions of the body but also in variability level and reactions to the fluctuations of environmental conditions (Anufriieva and Shadrin 2015).

Considering temperature, latitude and lake depth regarding the mean body size of female populations, our findings were not compatible with previous studies (Table 3 and Figure 4). Capture success by planktivorous fish depends largely on prey visibility and the ability of the prey to escape. Body transparency of copepods decreases their susceptibility to visual predators, but this trait has only limited significance because many vital processes interfere with it. For example, the gut of a feeding copepod is usually distinctly colored, oocytes developing in the gonads of females are clearly visible and are often dark-colored, reserve lipids are often pigmented, and movements of feeding appendages make copepods more conspicuous (Pasternak et al. 2006). So, it was expected to interactions of some environmental factors on body size variations, but the reason of why there were only significant variations between body sizes of female populations may be due to interaction of sexual dimorphism, physiology of female copepod and fish predations. Fish predation on copepod is affected by fish species, fish population density and the structure of age classes. In this manner, it would be reasonable to expect the fishing could be a latent factor effecting variations on body size of copepods.

In conclusion, both intra- and inter-specific variability on body size were observed in the

populations of *M. leuckarti*. Intra-population diversity was bigger in male than that of female populations in each water body. Also, inter-population diversity among male populations was bigger than inter-population diversity among female populations. There was statistically significance among the female populations; whereas, no significant differences were detected among the male populations. The variability observed most probably due to the combination of environmental factors, sexual dimorphism, physiology of female copepod, fish predation and the characteristics of water systems.

As other copepod species, in general, sexual dimorphism was observed in all water body. It was clear evidence that the specimen living in the pond and lake systems were closer than river systems in terms of body size especially for female ones of copepods. Also, regarding the significant variations between female populations, it would be reasonable to expect the fishing could be a latent factor effecting variations on body size of copepods.

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An Invasion Report of The New Zealand Mud Snail, *Potamopyrgus antipodarum* (Gray, 1843) in Turkish Freshwaters: Delice River and Kocabaş Stream

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ABSTRACT

This study is combined the data from two different studies that carried out different regions and time, presenting some information on the population structure (dominance, local distribution etc.) of *Potamopyrgus antipodarum* (Gray, 1843). According to our data, this species was found in the four different localities in the Delice River with various population densities. However, only of small population was found in the Kocabaş Stream. This species was the second dominant species in the Delice River with 31.43 % after *Physella acuta* (Draparnaud, 1805) (46.88%). On the other hand, the species was not reached noticeable aggregates in the Kocabaş Stream. In this paper, supporting factors that paving the way *P. antipodarum* invasion are discussed for the study area.

Keywords: *Potamopyrgus antipodarum*, invasive population, Delice River, Kocabaş Stream, Turkey

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Türkiye içsularında Yeni Zelanda Çamur Salyangozu, *Potamopyrgus antipodarum* (Gray, 1843)'un bir istila raporu: Delice Nehri ve Kocabaş Çayı

Öz: İki farklı bölgede ve zamanda yürütülen çalışmalardan elde edilen verileri kapsayan bu çalışma, *Potamopyrgus antipodarum* (Gray, 1843)'un popülasyon yapısı ile ilgili bazı verileri (baskınlık, bölgedeki dağılımı gibi) sunmaktadır. Verilerimize göre, Delice Nehri'nde *P. antipodarum* dört farklı noktada ve farklı populasyon yoğunluklarında tespit edilmiştir. Ancak Kocabaş Çayı'nda sadece küçük bir populasyonun varlığına rastlanmıştır. Bu tür % 31,43 değeri ile Delice Nehri'nde *Physella acuta* (Draparnaud, 1805)'dan (% 46,88) sonra ikinci en baskın türdür. Diğer taraftan, bu tür Kocabaş Çayı'nda çok yüksek sayıda bir popülasyona sahip değildir. Bu makalede, *P. antipodarum*'un istilasını destekleyen faktörler tartışılmıştır.

Anahtar kelimeler: *Potamopyrgus antipodarum*, istilacı populasyon, Delice Nehri, Kocabaş Çayı, Türkiye

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Introduction

The New Zealand mud snail, *Potamopyrgus antipodarum* (Gray, 1843), is a truncatelloidean gastropod that can be able to tolerate a wide range of abiotic conditions from coastal estuaries to freshwater ecosystems (Gérard et al. 2003). It is known as an invasive worldwide that the current expansion comprises several continents including Europe, mainland Australia and Tasmania (Ponder 1988) and North America (Bowler 1991).

According to Ponder (1988), introduction of this species date back to 19th century in southern Australia, Tasmania and Europe, then the species has been reported in North America in 1987 (Bowler 1991), Japan (Shimada and Urabe 2003), and more recently in Canada (Davidson et al. 2008). Shell remains of the species have also been found in Lebanon and Iraq (Naser and Son 2009). In Turkey, occurrence of *P. antipodarum* has been known from various freshwater ecosystems and only one coastal

marine area in the western and southern Anatolia since 1980 (Bilgin 1980; Ustaoğlu et al. 2001a, b; Ustaoğlu et al. 2003; Özbek et al. 2004; Kalyoncu et al. 2008; Kılıçarslan and Özbek 2010; Yıldırım et al. 2006; Kebapçı and Yıldırım 2010; Gürlek 2015; Odabaşı and Arslan 2015). Although, its global spread and high infestation rate, all the documentations rely on several shells or specimens and the present records are not related with its population and invasion status on their habitats in Turkey. In this study, we have combined the data sets of two different studies carried out in different running waters: Delice (a branch of the River Kızılırmak) and Kocabaş (running water located in Biga Peninsula), which are located in central and north-western Anatolia, respectively. This study aimed to evaluate the mollusc fauna and demonstrate the population structures of *P. antipodarum* inhabiting in these streams, we also evaluated the invasive characteristics of this species in studied areas.

Materials and Methods

Study Area

The first study area is Delice River that is one of the major tributaries of the Kızılırmak River which is the longest running water across Turkey. The Kızılırmak River flows for a total of 1355 km, rising from Eastern Anatolia and flows into the Black Sea. The Delice River, one of the main tributaries of the Kızılırmak River, flows along 430 km with a high flow rate approximately 30352 m^3/s annually. It has many small tributaries while passing through the Çankırı, Yozgat, Kırşehir and Kırıkkale cities before the joining to the Kızılırmak River (Gül and Yılmaz 2002). Samplings were carried out monthly between July 2007 and August 2008 in the preselected sampling sites in Delice River.

On the other hand, the second study area is Kocabaş Stream (also called as Biga Stream) that rising from the extension of the Kaz Mountain, the ancient name is known as Mount Ida, flows into the Sea of Marmara at Dardanelles. It is one of the most important watercourses in the Biga Peninsula with an 80 km in length and 30 m^3/s annual flow rate. Seasonal samplings were carried out between May 2012 and November 2013 at two sites located both at the upper and lower regions of the dam lake in the Kocabaş Stream (Table 1).

Table 1. Species Content of the freshwater Mollusca in the study area both Delice River and Kocabaş Stream. Legends: St.: Sampling Station, S: Status, N: Native, NC: Non-Native – Cosmopolitan.

	Delice River										Kocabaş Stream		
	St. 1	**St. 2**	**St. 3**	**St. 4**	**St. 5**	**St. 6**	**St. 7**	**St. 8**	**St. 9**	**St. 10**	**Downstream**	**Upstream**	**S**
Borystenia naticina				+									N
Valvata kebapcii				+				+	+				N
P. antipodarum	+			+	+			+			+		NC
Pseudamnicola natolica	+				+								N
Theodoxus fluviatilis	+												N
Ancylus fluviatilis												+	N
Galba truncatula					+								N
Physella acuta	+			+	+	+		+	+	+		+	NC
Radix labiata	+				+			+	+	+			N
Gyraulus piscinarum	+			+	+			+	+			+	N
Dreissena polymorpha	+												N
Musculium lacustre												+	N
Euglesa casertana	+												NC
Pisidium subtruncatum				+						+			N
***Sphaerium* sp.**				+									N

Benthic samples were collected with a surber net from the different habitats in the stream including aquatic vegetation, stone-gravel, and sand. The samples were sieved with a series of strainer mesh sizes of 1, 0.5 and 0.25 mm, and then the snails were put into 75 % lab-grade ethanol in the field. Individuals counted one by one under stereo microscope to determine density in a unit area. A random sub-sampling was performed for shell measurements including shell height (SH), shell

width (SW) according to Glöer (2015). Shell measurements carried out by means of imaging system consisting of stereo microscope (Stemi 508, Zeiss) and a camera (Axiocam 105 color). At least 26 snails were included to measurements by sub-sampling. We also inspected to brood pouch contents of the sub-sampled snails in order to reveal seasonal reproductive efficiency. Randomly 15 snails per sub-sample unit were inspected for brood pouch regardless of their size.

Results and Discussion

Mollusca fauna and population structure of invasive species, *P. antipodarum*, inhabiting in two distinct areas were evaluated. Results showed that 12 and 5 taxa inhabiting in the Delice River and Kocabaş Streams respectively (Table 2, 3). The members of Mollusca fauna were assessed as nativeand non-native. There are 12 native and 3 non-native species were recorded in the two study areas (Table 1).

Potamopyrgus antipodarum originated from The New Zealand and *Physella acuta*, North American origin, are known as global invaders (Dillon et al. 2002; Semenchenko et al. 2008), so we can describe them as a non-native one in this study. In the stations of Delice River, a population with a higher number of individuals was detected (Figure 1, Table 2).

Table 2. Individual numbers per square meter of Mollusca found in the sampling stations (St.) of the Delice River.

Individual numbers	St. 1	St. 4	St. 5	St. 6	St. 8	St. 9	St. 10
Borystenia naticina	0	44	0	0	0	0	0
Valvata kebapcii	0	222	0	0	88	44	0
Potamopyrgus antipodarum	677144	44	133	0	89	0	0
Pseudamnicola natolica	44	0	88	0	0	0	0
Theodoxus fluviatilis	1909	0	0	0	0	0	0
Galba truncatula	0	0	1909	0	0	0	0
Physella acuta	16561	89	12318	14340	133	18958	44
Radix labiata	44	0	177	0	44	177	443
Gyraulus piscinarum	177	133	44	0	44	44	0
Dreissena polymorpha	44	0	0	0	0	0	0
Euglesa casertana	133	0	0	0	0	0	0
Pisidium subtruncatum	0	266	0	0	0	0	177
***Sphaerium* sp.**	0	44	0	0	0	0	0

Table 3. Individual numbers per square meter of Mollusca found in Kocabaş Stream

Individual numbers	Lw*	Up*
Potamopyrgus antipodarum	142	0
Ancylus fluviatilis	0	44
Physella acuta	0	22
Gyraulus piscinarum	0	22
Musculum lacustre	0	22

Lw: Lower part of the dam lake, Up: Upper part of the dam lake.

According to the community parameters in the Delice River, *P. antipodarum* predominated over the other mollusca taxa in the sampling site 1 (St. 1) (Figure 1). It has reached of the densest population in St. 1 with 56429/m^2 (Table 2). Among the sampling stations of the Delice River including 4th, 5th and 8th, *P. antipodarum* rarely established with a sparsely population (Table 2).

On the other hand, *P. antipodarum* was sampled from only one sampling site (lower dam Lake of Bakacak) with a few numbers of individuals (142/m^2) but predominated over associated species in Kocabaş Stream (Figure 4, Table 3).

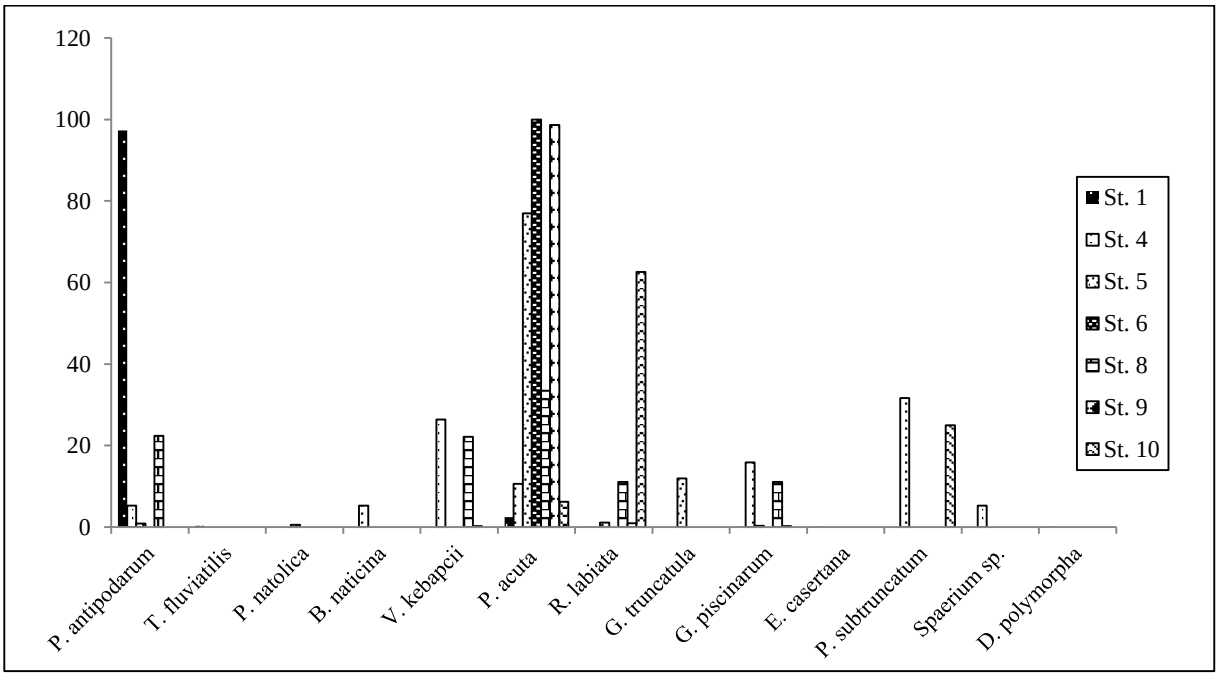

Figure 1. Dominance percentage of the Mollusca in the stations of the Delice River.

Considering the habitat structure and characteristics of the stations in which inhabiting of a *P. antipodarum* population in the study area, in the Delice River, the first station can be placed into a distinguished position among others as it is mainly fed by groundwater springs from the bottom (personal observation). *Theodoxus fluviatilis* and *Pseudamnicola natolica* are associated taxa with *P. antipodarum* supporting this claim because of their special habitat preference; springs (rheocrenes) and groundwater inhabitants (Yıldırım 1999; Falkner et al. 2001) that indicating the groundwater sources in the sampling region. According to the data, *P. antipodarum* might be considered as invasive for the region (1st station of the Delice River) due to a well-established population. Owing to the constant water quality regime throughout the year occurring in the spring-fed streams, they are suitable for growthof introduced species. The report of Hamada et al. (2013) also supports our findings that hot spring discharges are suitable habitats of *P. antipodarum* in Japan.

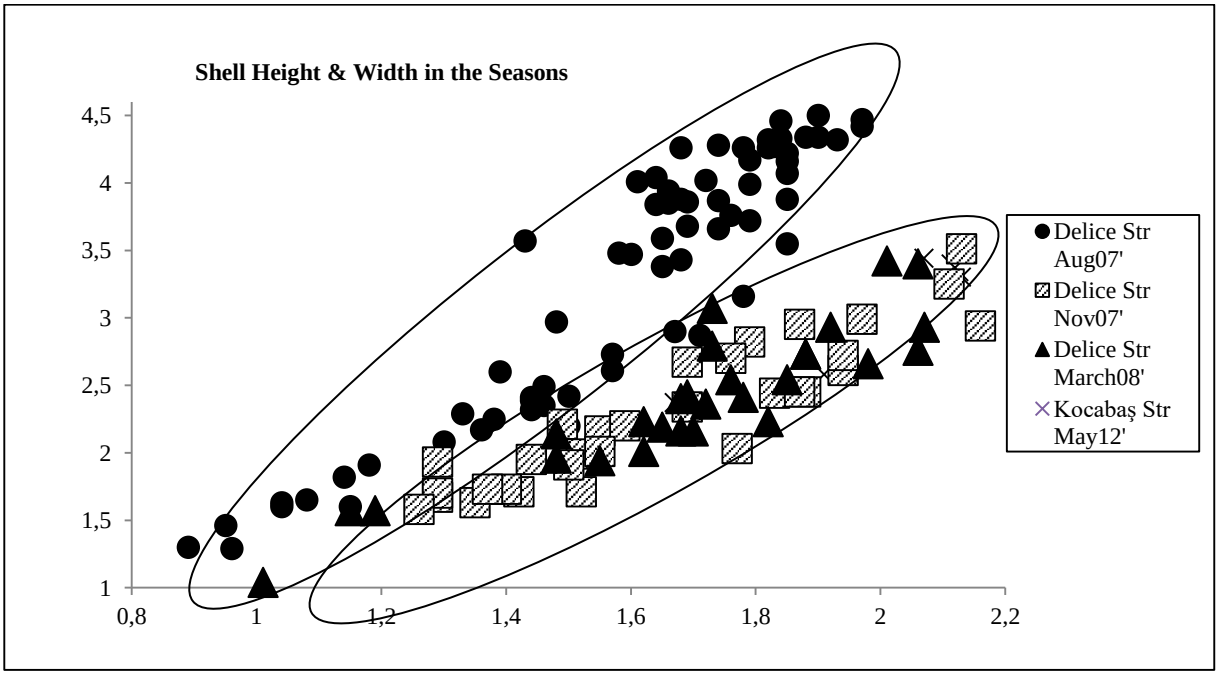

Figure 2. Seasonal shell height and shell width distributions of *P. antipodarum* in Delice River and Kocabaş Stream.

Shell height (SH) and width (SW) distributions of *P. antipodarum* revealed for streams in different seasons (Figure 2). As can be seen in the diagram, higher shells of *P. antipodarum* were observed in the season August 2007 in Delice River (Figure 2). Other seasons' SH and SW distributions of *P. antipodarum* belong to the streams were overlapped.

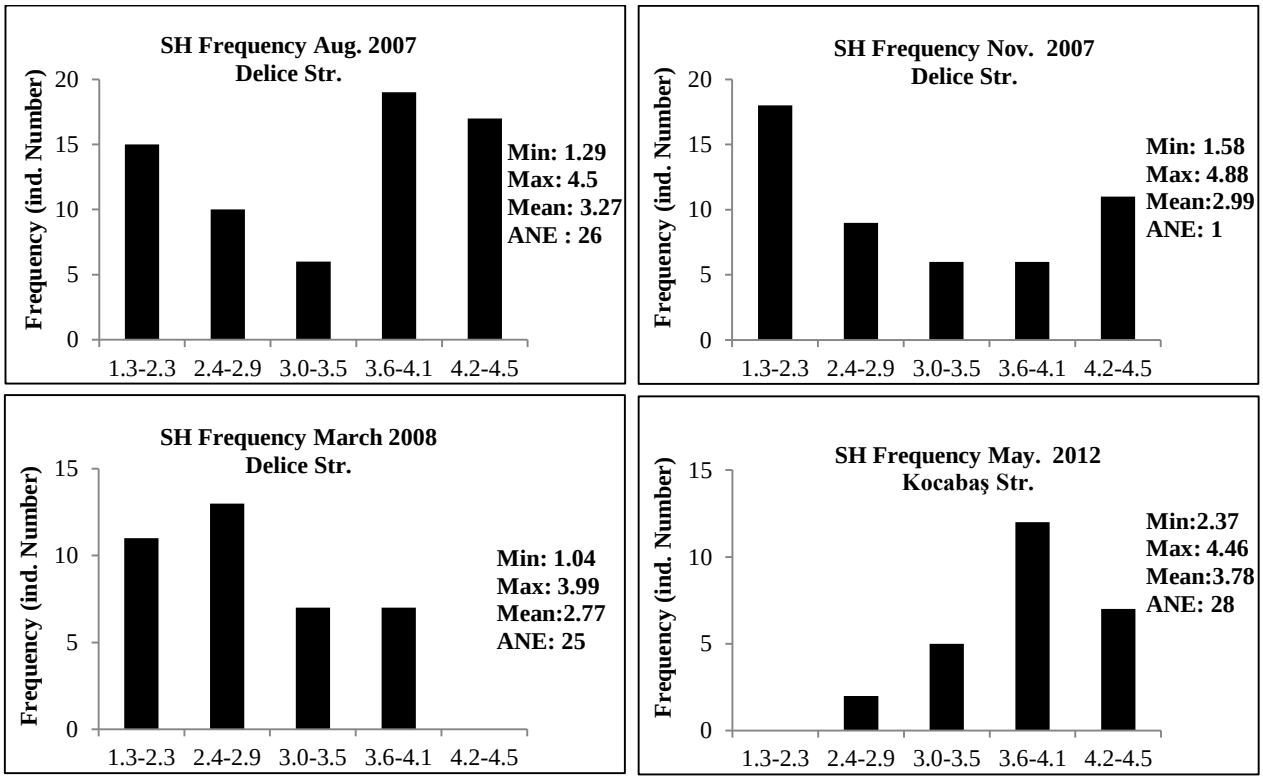

Figure 3. Seasonal shell height frequencies with maximum, minimum, mean values and the average number of embryo (ANE) of *P. antipodarum* in Delice River and Kocabaş Stream.

Snails were grouped into five SH ranges between minimum of 1.3 mm and maximum 4.5. Similar to SH distributions, higher frequency of appearance of longer shells were detected in the August 2007 of Delice River. On the contrary, the smaller SH ranges of shells assembled in the months March, May, and November, which are representing spring and autumn seasons. Minimum and maximum SH (mm) values, as expected, were in March and August 2007 respectively. Appearance frequency of the SH groups might be indicating that reproduction and individual growing occurred in seasons. Our data showed an annual life cycle for the seasons except winter. Accordingly, the first group range of the population, it can be considered as juveniles, were dominant group in November 2007 and March 2008 while the adults that can be placed into the last two SH groups were higher in number in August 2007. Although the sampling at the Kocabaş Stream coincided in early summer, growing population is visible by rising numbers of adult groups. As can be seen in charts, average number of embryo production was interrupted at Delice River only in November 2007 (Figure 3, ANE). The most productive season was early summer (in Kocabaş Stream) and summer (in Delice River). According to the brood pouch inspection, the minimum size of reproductive maturity was 3.47 mm in SH and 1.6 mm in SW bearing 21 embryos and neonates (Figure 3).

Dorgelo et al. (2014), revealed the dynamics of *P. antipodarum* populations inhabiting in two different lakes which have different trophic levels. The study showed that size distributions followed a regular annual pattern in the eutrophic lake. In this study, similar to our findings, smaller individuals that made a dense contribution to population size in one season, increased larger shells in the next season as reducing the number of smaller individuals. According to pouch analysis, embryo and neonates production was interrupted between November and February. Taking into account of the reproduction time and first reproduction maturity, a close similarity is observed with our study.

Before a species invade to an ecosystem, it has to overcome several challenges (Kolar and Lodge 2001; Sakai et al. 2001; Alonso and Castro-Díez 2008).

After the introduction of the species to a new environment, it must survive, grow and reproduce under the new environmental conditions (establishment). Furthermore, it must compete with the other organisms especially their ecologically similar taxa in order to reach a high population growth rate (invasion or spread). After that, the exotic species must alter the structure and functioning of the occupied ecosystem (impact) (Parker et al. 1999). In our study, as can be seen in Figure 1, *P. antipodarum* is successfully colonized at the 1st station of Delice River with an enormous number of individuals dominating the native mollusk community. Thus, there is a possible risk of impact on the native ecosystem in this site due to establishment success of the invader. Besides, *P. acuta* predominated over existing taxa at the other sites of Delice River. When we consider two non-native invaders, it is obvious that *P. antipodarum* is predominating *P. acuta* at the 1st station of Delice River. In Kocabaş Stream, there is no certain evidence about invasion processes of *P. antipodarum*. However, the species seems to reach an establishment success in the lower part of the Bakacak Dam in Kocabaş Stream (Figure 4).

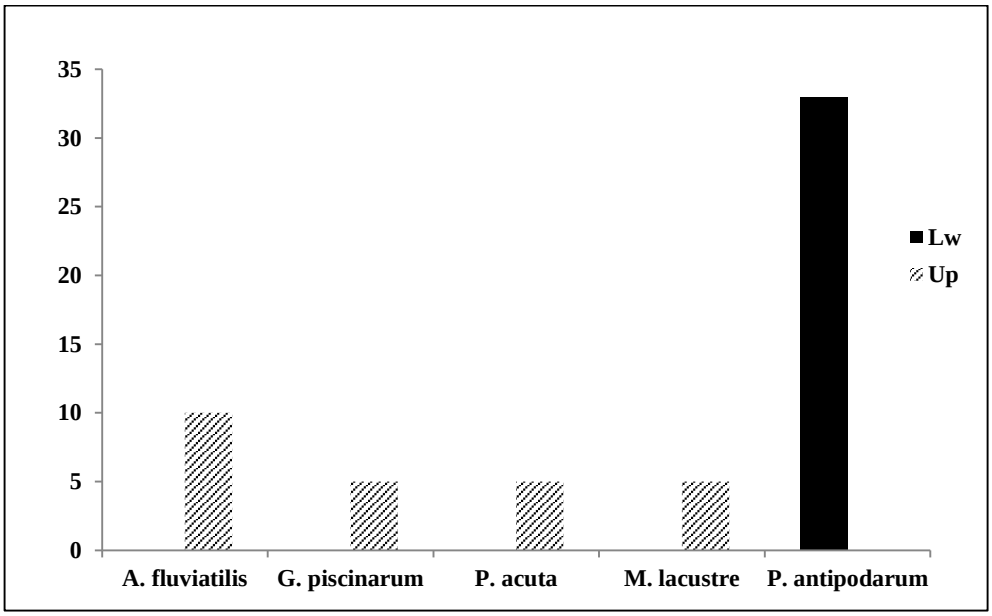

Figure 4. Dominance percentage of the Mollusca in the lower (Lw) and upper parts of the Dam Lake on the Kocabaş Stream.

In conclusion, further investigations and monitoring programs should be applied in order to clarify the influence of this non-native species on structure and functional ecology of the native benthic community.

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Akyaka Kadın Azmağı'nda Tespit Edilen *Salaria fluviatilis* (Asso,1801) (Familya: Blenniidae) Türünün Yumurta ve Larvalarının Morfolojik Özellikleri

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ÖZ

*Salaria fluviatilis (*Asso,1801), Blenniidae familyasının Akdeniz'de temsil edilen tek tatlısu türüdür. Bu çalışmada, ülkemiz içsularında ilk kez doğadan örneklenen bir kemikli balığa ait yumurta ve larvaların morfolojik özellikleri verilmiştir. 22 Temmuz 2012'de Gökova Körfezi'ne açılan Akyaka Azmak Nehri'nden S. *fluviatilis* türüne ait çok sayıda aynı gelişim evresindeki (çoğunluğu son safhaya erişmiş) yumurtalar tespit edilmiştir. Demersal özellikteki yumurtaların çapları; 0,89-1,31x0,78-1,23 mm, bu yumurtalardan çıkan prelarvaların boyları; 2,4-3,8 mm, postlarvalarınki 3,3-4,2 mm olarak ölçülmüştür.

Anahtar kelimeler: Blenniidae (Horozbina), demersal yumurta, Gökova Körfezi.

MAKALE BİLGİSİ

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Morphological Characteristics of Eggs and Larvae of *Salaria fluviatilis* (Asso,1801) (Family: Blenniidae) Collected from Akyaka Azmak Creek

Abstract: *Salaria fluviatilis (*Asso,1801) is the only freshwater species of Blenniidae represented in the Mediterranean Sea. This study stands as the first one to provide the morphological characteristics of one of Teleost fish eggs and larvae sampled from their natural habitats in Turkey's inland waters. A great majority of eggs (mostly at late stage) belonging to *S. fluviatilis* found in Akyaka Azmak creek flowing into the Gökova Bay on 22 July 2012. Diameters of these demersal eggs were 0.89-1.31x0.78-1.23 mm, and lengths of these hatched prelarvae were recorded as 2.4-3.8 mm, postlarvae 3.3-4.2 mm.

Keywords: Blenny, demersal eggs, Gökova Bay.

Alıntılama
Çoker T. 2019. Akyaka Kadın Azmağı'nda Tespit Edilen *Salaria fluviatilis* (Asso,1801) (Familya: Blenniidae) Türünün Yumurta ve Larvalarının Morfolojik Özellikleri. LimnoFish. 5(3): 220-225. doi: 10.17216/LimnoFish.484462

Giriş

Salaria fluviatilis (Asso,1801), erginleri sahile yakın göllerde veya az derin akarsuların taşlık ve kayalık zonlarında yaşayan, küçük boylu bir demersal balıktır (Whitehead vd.1989). Tuzlu suda da yaşayabilirler, kendi bölgesinden fazla uzaklaşmazlar, 5 yaşına kadar yaşarlar (Kottelat ve Freyhof 2007). Akdeniz'de Avrupa Baseni, Kuzeybatı Afrika ve Türkiye sahillerinde yayılış gösterirler (Anonim 2018). İznik Gölü'nden Özuluğ vd. (2005); Ceyhan Nehri'nden Kara vd. (2010), Seyhan Baraj Gölü'nden Ergüden ve Göksu (2012), Marmara, Küçük Menderes, Batı Karadeniz, Antalya, Doğu Akdeniz, Seyhan ve Ceyhan havzalarına ait akarsulardan İlhan vd. (2013) ergin kayıtlarını vermişlerdir.

S. fluviatilis bireyleri birinci yaşın sonunda üremeye başlar ve bir dişi bir seferde 1200 kadar yumurta bırakabilir (Anonim 2018). Dişi balık 2-30 kez olmak üzere, her seferde ortalama 200-300 yumurta bırakır. Bir yuvadaki yumurta sayısı 500-8000 civarındadır (Gasith ve Goren 2009). Dişi taşların altında yumurtalarını yığın olarak tek tabaka halinde yapıştırır ve döllenme olduktan sonra yuvayı terk eder (Vinyoles vd. 2002). Yumurtlanan yumurtaları erkek bireyler korur (Kottelat 2004), yuvanın temizlenmesini, gerekirse taşınmasını ve pectoral yüzgeçlerini hareket ettirerek yumurtaların

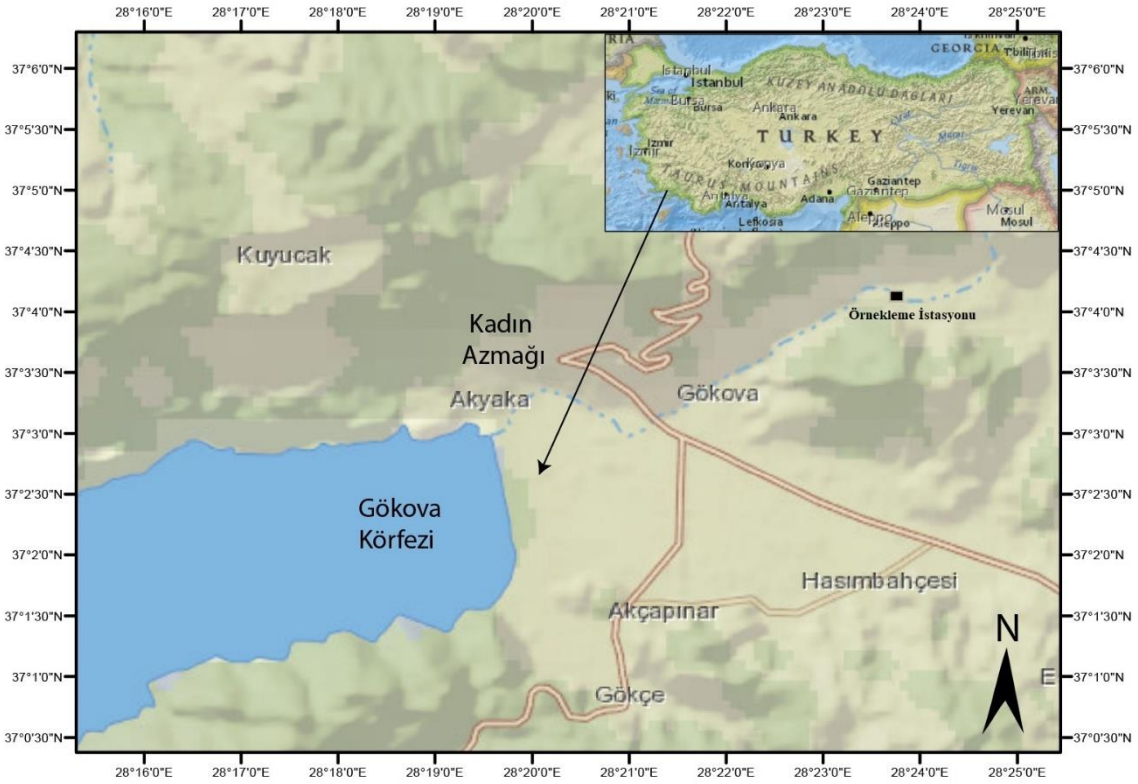
oksijenlenmesini sağlar. Çoker (1996; 2017) İzmir Körfezi'nde Blenniidae familyasına ait denizel 6 türün; (*Parablennius gattorugine* (Linnaeus,1758), *Parablennius tentacularis* (Brünnich,1768), *Salaria pavo* (Risso, 1810), *Parablennius sanguinolentus* (Pallas,1814), *Blennius ocellaris* Linnaeus,1758, *Aidablennius sphynx* (Valenciennes,1836), *Coryphoblennius galerita* (Linnaeus,1758) ve Karadeniz'de Gordina vd. (2005) 1 türün; *Parablennius zvonimiri'*nin (Kolombatović, 1892) larvalarını bildirmişlerdir.

İlk olarak (Gill vd. 2010) *S. fluviatilis* türünü laboratuvar koşullarında yetiştirerek embriyonik gelişimlerini izlemiş, yumurta ve larval dönemlerini morfolojik olarak tanımlamış ve fizyolojik koşullarını kaydetmişlerdir.

Gökova Körfezi iç kesimi tatlısu, acısu ve denizel ortam karakterlerini gösterir ve bu üç ekosistem bir kısım balıkların üreme ve beslenme göçleri açısından geçiş bölgelerini oluşturur. Dolayısıyla bu alanda türlerin üreme özelliklerinin ve yaşam ortamlarının belirlenmesinde yeni bulgular önemlidir. Bugüne kadar Akyaka Kadın Azmağı'nda *S. fluviatilis* türünün gerek erginleri gerek erken evre düzeyinde yumurta ve larvalarına dair bilgi veya kayıt bulunmamaktadır. Bu çalışmada nehirde tesadüfi olarak bulunan türün yumurta ve larvaları rapor edilerek, morfolojik özellikleri kaydedilmiş, çap ve boy ölçümleri ile bazı morfometrik oranları verilmiştir.

Materyal ve Metot

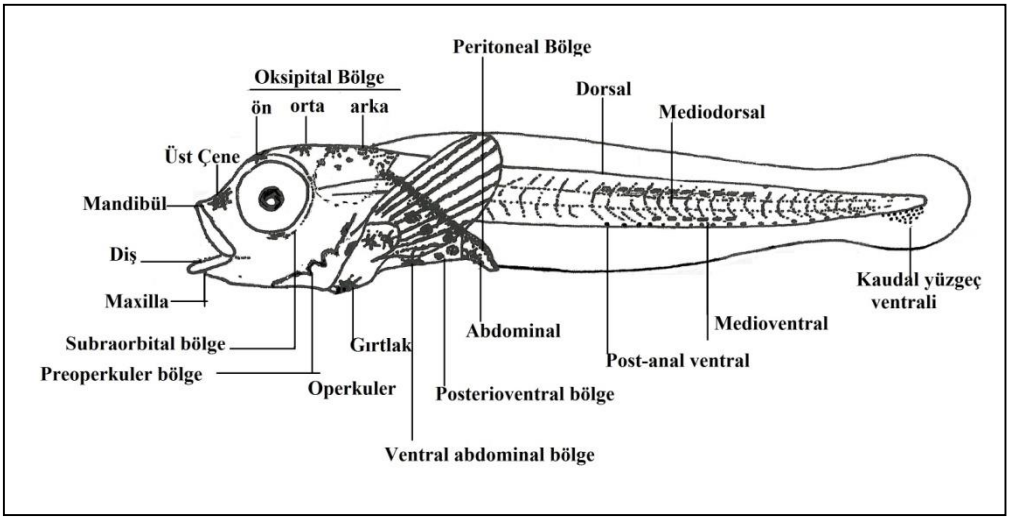
22 Temmuz 2012 tarihinde Gökova Körfezi'ne dökülen 1700 m uzunluğundaki Akyaka Kadın Azmağı'nda, denizden 1200 m mesafede (37°02'36"K, 28°20'12"D) bir iskele ayağının yakınına atılmış bir tahta parçası üzerine bırakılmış bir yığın halinde bulunan balık yumurtaları 1,5 m derinlikten dalarak çıkarılmıştır (Şekil 1).



Şekil 1. Örnekleme Alanı ve Örnekleme İstasyonu.

Bulunan yumurtalar, plastik bir kapta Azmak suyu içinde kendi haline bırakılmış ve yaklaşık 24 saat içinde yumurtaların pek çoğunun açıldığı gözlenmiştir. Sonrasında yumurta ve larvalar %4 formaldehit çözeltisi içine konularak muhafaza edilmiş, SZ-61 Olympus marka binoküler mikroskop altında teşhis ve tanımları, ölçümleri (4x1) yapılarak, fotoğraf çekimleri gerçekleştirilmiştir. Yumurtadan çıkan larvalar Şekil 2'de belirtilen morfolojik karakterler dikkate alınarak tanımlanmıştır.

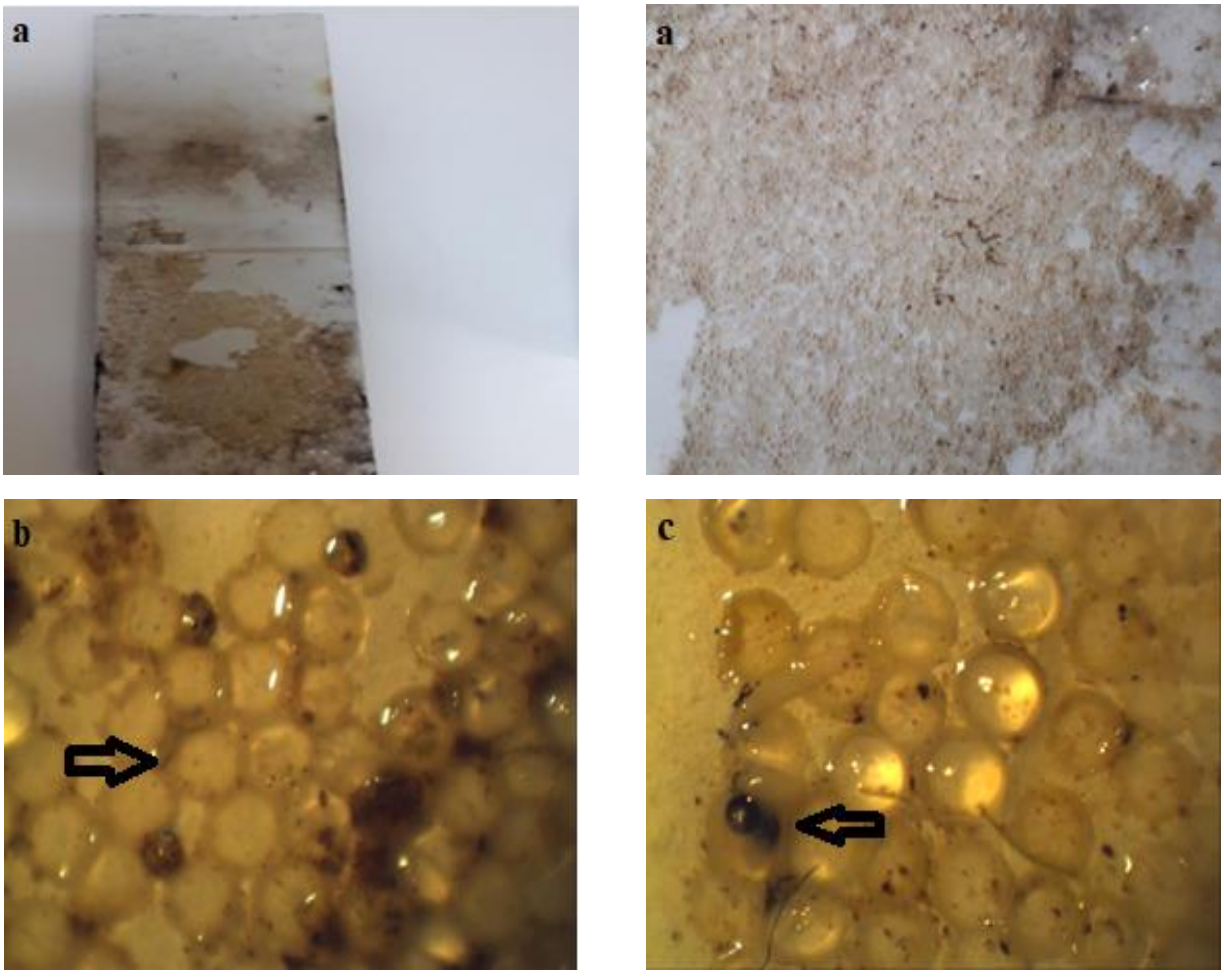
Türün örneklenmesini takiben YSI 550 ölçüm cihazı kullanılarak sıcaklık ve oksijen ölçümleri yapılmıştır.



Şekil 2. Bir Blenniid larvasının morfolojik karakterleri ve pigmentasyon görülen vücut bölümleri (Çoker 1996).

Sonuçlar

Akyaka Azmak Nehri'nden, *S. fluviatilis* türüne ait safhası olmayan (embriyonik gelişimin henüz başlamadığı) ve çok sayıda son safhaya erişmiş olan yumurtalar tespit edilmiştir (Şekil 3a). Yumurtaların bulunduğu derinlikte sıcaklık ve oksijen değerleri; 17,0 °C ve 5,80 mg/L olarak ölçülmüştür.



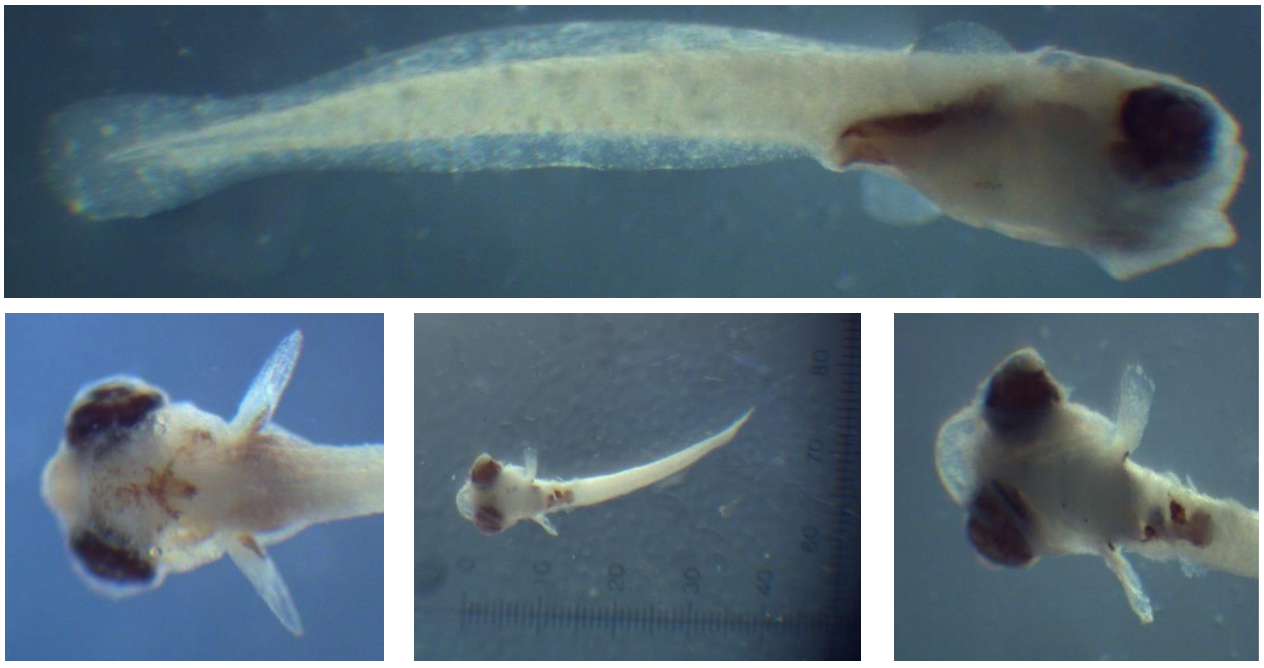
Şekil 3. a: Tahta üzerinde yüzeye ve birbirine yapıştırılmış halde çok sayıdaki *S. fluviatilis* yumurtaları **b:** Embriyonik safhası olmayan ve **c:** Embriyonik gelişimi son safhaya erişmiş haldeki yumurtalar.

Demersal özellikteki yumurtalar, basık küresele yakın şekillidir. Yumurta çapları; genişliği ve yüksekliği; 0,89-1,31x0,78-1,23 mm'dir. Bir kısım yumurtalarda embriyonik segmentasyonun henüz başlamadığı görülmüştür (Şekil 3b). Embriyosu gelişmiş yumurtalarda vitellüs kesesi üzerinde koyu kahve renkli küçük yıldız şekilli kromatoforlar embriyonik eksenin posteriorü etrafında yoğunlaşmıştır. Gözleri büyük (0,3 mm) ve belirgindir. Vitellüs homojendir. Yumurtalarda vitellüs üzerinde çok sayıdaki yağ damlaları gözlenmemiştir (Şekil 3c).

Yumurtadan çıkan prelarvaların boyu 2,4-3,8 mm'dir. Ağız açılmamıştır ve vitellüsleri homojen ve açık sarı renklidir. Preanal vücudun;1/3'de yer alır. Göz çapı baş boyunun yaklaşık yarısı kadardır. Vitellüs şeffaf ve belirgindir, tüm abdominalin yarısını veya yarısından çoğunu kaplamıştır. Baş üzerinde kalın nokta şeklinde yoğun ve dağınık kromatoforlar yer alır. Post-anal ventral sıra kuyruğa kadar 18-19 adet küçük noktalar şeklinde uzanır. Pektoral yüzgeç kaidesinde nokta şekilli kromatofor yer alır. Peritoneal kesikli kalın bant şeklindedir, barsak ventrali üzerinde belirgin bir yıldız kromatofor ve abdominalde küçük bir yıldız kromatofor yer alır (Şekil 4).

Postlarvaların boyları; 3,3-4,2 mm'dir. Baş vücuda oranla biraz büyük, vücut kuyruk kısmına doğru daralmıştır. Anüs vücudun ½,5'da yer almaktadır. Pektoral yüzgeçler orta büyüklükte ve yelpaze şeklindedir. Miyomer sayısı; 7+25 (32)'dir. Postlarvaların post-anal ventrali üzerinde 7. miyomerden başlamak üzere kuyruğa kadar uzanan 18 adet nokta şeklinde, baş üzerinde 3-4 adet yıldız şeklinde büyük belirgin, barsak posterioründe tek yıldız şekilli, abdominal bölge üzerinde birkaç adet olmak üzere kromatoforlar görülür ve peritoneal bölgede de yoğun pigmentasyon vardır (Şekil 5).

Şekil 4. 3,6 mm boyundaki *S. fluviatilis* prelarvası.



Şekil 5. 4,1 mm boyundaki *S. fluviatilis* postlarvası

Tartışma

Daha önce ülkemiz içsularında birkaç türün yumurta ve larvaları yetiştiricilik yoluyla tanımlandığı halde (Şahinöz vd. 2007; Doğu vd. 2013), bu çalışmada ilk kez doğadan örneklenen yumurta ve larvaların morfolojik özellikleri belirlenmiştir.

Yumurta yığını içinde farklı gelişim evrelerinin görülmesi türün poligamik üreme özelliklerine ilişkindir. Vodynitskii ve Kazanova (1954) erkek Blenniid'in yumurta bırakma yerini seçtiği ve birkaç dişiyi buraya çektiğini belirtmiştir. Böylelikle bir yuvada birkaç dişiden gelen yumurtalar farklı zamanlarda döllenmekte, aynı yuvadaki bireylerin yumurta çapları ve gelişim hızları farklılıklar göstermektedir. Örneklerimizde ortalama yumurta çapları 1,13x1.03 mm'dir. Yumurtaların yüksekliği mevcut değerlerin biraz üzerindedir. Psarras vd. (1997) yumurta çapını 0,95x0,68 mm, Gil vd. (2010) 1,1-1,2x0,70-0,80 mm olarak ölçmüşlerdir. 3,2-3,8 mm aynı boylardaki larvaların bir kısmı prelarva bir kısmı postlarva olarak ayırt edilmiştir. Gil vd. (2010) çalışmamızda 32 olarak kaydedilen miyomer sayısını 36-37 olarak vermişlerdir. Bu farklılıkların ayrı popülasyonlardan gelen ergin bireylerin gelişim özelliklerinden ve farklı sıcaklıklardan kaynaklandığı düşünülmektedir. Çoker (1996) denizel türlerin larvalarının yumurtadan çıkma boyunun kaynaklarda belirtilenden daha kısa olduğunu tespit etmiş, embriyonik gelişimin sularımızda daha hızlı olduğunu belirtmiştir.

Gil vd. (2010) embriyonik gelişimin 20-21 °C'de 12-14 gün, Gasith ve Goren (2009) 21-23 °C'de 11-13 gün sürdüğünü belirtmişlerdir. Çalışmamızda 17 °C Azmak Nehri'nden alınan embriyosu gelişmiş olan yumurtalar oda sıcaklığında yaklaşık 24 saat sürede yumurtadan çıkmışlardır.

Türün laboratuvar denemelerinde 14-31 °C'lerde yumurtladığı belirtilmiştir (Gasith ve Goren 2009). Çalışmamızda tespit edilen yumurtalar Temmuz ayında elde edilmiştir. Üremenin yıl boyunca olduğu ancak, türün yoğun olarak Mart veya Temmuz'da yumurtladığı bilinmektedir (Psarras vd. 1997; Gasith ve Goren 2009). Dekhnik (1973), denizel türlerde su sıcaklığı 15-16 °C'de iken Blenniid larvalarına nadir rastlandığı, 18-20 °C'de sayılarının birkaç kat arttığı, 25-26 °C'de yoğun yumurta bırakmaya başladıklarını belirtmiştir.

Gil vd. (2010) Türün yumurtaları için çözünmüş uygun oksijen değerini; 5,44 mg/L olarak vermişlerdir. Demirak vd. (2012) Azmak Nehrinde sıcaklıkları yıl boyunca 15-17 °C'lerde, oksijen değerlerini 7,17-9,51 mg/L, Döndü (2015) bir yıllık ortalama su sıcaklığı ve oksijen değerlerini; 16,40-16,51 °C ve 4,2-9,4 mg/L, Yabanlı vd. (2014) mevsimsel ortalama sıcaklık ve oksijen değerlerini; 14,81-16,71 °C ve 7,89-8,61 mg/L, Cesur vd. (2014) pH değerini 7,27, sıcaklığı; 16 °C, Çoker vd. (2015) Azmak sularında sonbahar ayları için sıcaklık; 16-18°C, tuzluluk; 2,69-2,70 ppt, oksijen değerlerini; 6,38-7,03 mg/L olarak ölçmüşlerdir.

Okuş vd. (2006) Akyaka kıyı ve Azmaklar mevkiinden türlerin en önemli yumurtlama alanlarından biri olarak bahsetmişlerdir. Çoker vd. (2015) sonbahar döneminde Gökova İç Körfez ihtiyoplanktonunda 5 familyadan 9 denizel kemikli balık türünün yumurta ve larvasına rastlamışlar, Kadın Azmağı üzerinde yer alan üç istasyondan hiç yumurta larva kaydetmemişlerdir. Araştırıcılar İç Körfez'de ihtiyoplanktonda birey ve tür sayılarındaki azlığı, sonbaharda üreyen balık tür sayısının fazla olmaması ve körfezin üretkenliğindeki mevsimsel düşmeye bağlamışlardır. Kontrolsüz avcılığın yanında hızla artan turizm ve onunla birlikte artan çevre kirliliğinin olumsuz etkilerinin sistemin verimsizleşmesine yol açacağı ve ortamdaki balıkların üremesi için uygun ortamların yok olma tehlikesi ile karşılaşacağı belirtilmiştir (Top vd. 2013). Demirak vd. (2012) bu alanda giderek artan su kirlenme sorununu; yasal olmayan atıksu boşaltılması, gezi ve balıkçı teknelerinin varlığına bağlamışlar, Yabanlı vd. (2014) tekne tamiri ve boyalarının civarı kirletmesine ve inceledikleri makrofitin köklerinde maksimum miktarda ağır metal bulunduğuna dikkat çekmişlerdir. *S. fluviatilis* Red-List'te LC (Least concern) kategorisinde yer alan türlerden biridir, Akdeniz'de bu tür üzerindeki asıl tehditler; ötrofikasyon, habitat bozulmaları ve istilacı türlerin predasyonu olarak belirtilmiş, popülasyonlarını sınırlayıcı asıl faktörün ise pelajikte geçirdikleri larval evreleri olduğu vurgulanmıştır (Anonim 2018). Azmak nehrinde halen üreme işlevini sürdüren bu türün larval dağılım ve bollukları, erginlerin üreme özellikleri ve populasyonları ile ilgili ileride ayrıntılı çalışmalar yapılmasına ihtiyaç vardır.

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