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Selçuk Altınsaçlı
İstanbul University, Faculty of Fisheries, Ordu Street. No: 200 34310 Laleli-İstanbul- Turkey

Juan Rueda Sevilla
Department of Microbiology and Ecology, University of València, Av. Dr. Moliner, 50, E-46100, Burjassot, Spain

Songül Altınsaçlı
Department of Biology, Faculty of Science, 34459, Vezneciler, Istanbul-Turkey

Correspondence:
Selçuk Altınsaçlı
İstanbul University, Faculty of Fisheries, Ordu Street. No: 200 34310 Laleli-İstanbul-Turkey

On occurrence of *Spongilla lacustris* (Linnaeus, 1759) from Lake Saklıgöl, Kandıra District, Turkey

Selçuk Altınsaçlı, Juan Rueda Sevilla & Songül Altınsaçlı

ABSTRACT
This study was carried out to determine porifera fauna of the freshwater Lake Saklıgöl. A single freshwater sponge species (*Spongilla lacustris*, Porifera, Spongillidae) has been observed in alkaline, well oxygenated and clear hard water with low electrical conductivity of the karstic and shallow Lake of Saklıgöl, Turkey. *Spongilla lacustris* became more dominant in June 2013 than in other months. Results showed that Lake Saklıgöl is an important habitat for *Spongilla lacustris* in this region.

Keywords: Kocaeli, Lake Saklıgöl, *Spongilla lacustris*, Sponge, Turkey.

1. Introduction

Freshwater sponges show a worldwide distribution from the Polar Regions to the tropics except in the Antarctic region and at the North Pole [1]. Sponges (Porifera) are generally believed to be ancient metazoans due to sponge fossils dating back 580 million years to Precambrian times [2, 3]. According to another opinion, the oldest known fossils of sponges in inland waters are from the Mesozoic Era [4, 5, 6]. The geographic distribution of the *Spongilla lacustris* (Linnaeus, 1759) covers the Holarctic region. Genus *Spongilla* Lamarck, 1816 includes 15 species [7]. Also, endemic species belonging to genus *Spongilla* such as *Spongilla prespensis* (Hadzische, 1953) and *Spongilla stankovici* (Arndt, 1938) have been reported from different lakes in Balkans (Europe).

Sponges are divided into three classes mainly according to the composition of their skeletons including Hexactinellida, Calcarea and Demospongiae [1]. 5000 species of sponges have been described across the world [8]. 8100 sponge species have been defined throughout the world and these species have been listed in the World Porifera Database [9]. Freshwater sponges live in standing and running fresh waters. Most of the sponges (excluding larvae) live as sessile forms in the sea, but small portion of their (Family Spongillidae) are found in fresh waters. Most sponges are found in coastal water and in deep waters on stones, coral reefs and plants or on the surface of any hard substrate. Most sponges prefer hard substrate such as rocks, metal or cement port, logs, macrophytes, wood sticks, shell of mollusc, rock and stones [10, 11], whereas, *Spongilla lacustris* can live on the soft muddy substrate of lakes. Sponges, despite being important water filter-feeding organisms, nevertheless, importance of theirs are not understood by mankind. Common freshwater sponge species is not accepted neither threatened or endangered species in animal conservation species categories.

Freshwater sponge fauna is poorly-known from Turkey due to the fact that sponges have been ignored in many benthic studies. The freshwater sponge species *Ephydatia fluviatilis* (Linnaeus, 1759) has been recorded in Çubuk Dam [12] and Lake Köyceğiz [13]. Other freshwater *Spongilla alba* (Carter, 1849) was recorded in Lake Köyceğiz by Gugel [14]. The freshwater sponge species *S. lacustris* and *Ephydatia* sp. were listed without taxonomic details contrary to scientific expectations from Lake Terkos, Kızılırmak Delta Wetlands, Lake Gala and Lake Melen [15]. The faunal assemblage of the sponges was investigated in detail in Lake Saklıgöl. During the study, *S. lacustris* samples were collected, identified and reported from Lake Saklıgöl. The aims of the present study are to identify the freshwater sponge species and determine the environmental factors affecting the distribution of *S. lacustris* in Lake Saklıgöl.

2. Material and Method

The karstic freshwater Lake Saklıgöl is located in the northwest of the district of Kandıra (Kocaeli Province) and to the north of the village Pınarlı in Turkey (41° 07' 25. 81''- N 29° 55' 19.27'' E; 77 m. a.s.l.) (Fig.1). The lake is a mesotrophic with a mean depth of 150 cm, maximum depth of 4 m and surface area of 3.9 ha [16]. This karstic freshwater lake is situated in the Upper Cretaceous-Paleocene age limestones of the northwestern part of the Kandıra District (Kocaeli, Turkey) [17]. The lake basin was formed by the solution of these carbonated rocks. The lake is fed by an underground water source and rain water source into a depression with no surface inlet or outlet. The lake water outlet can only be accounted for by discharge through a karst aquifer.

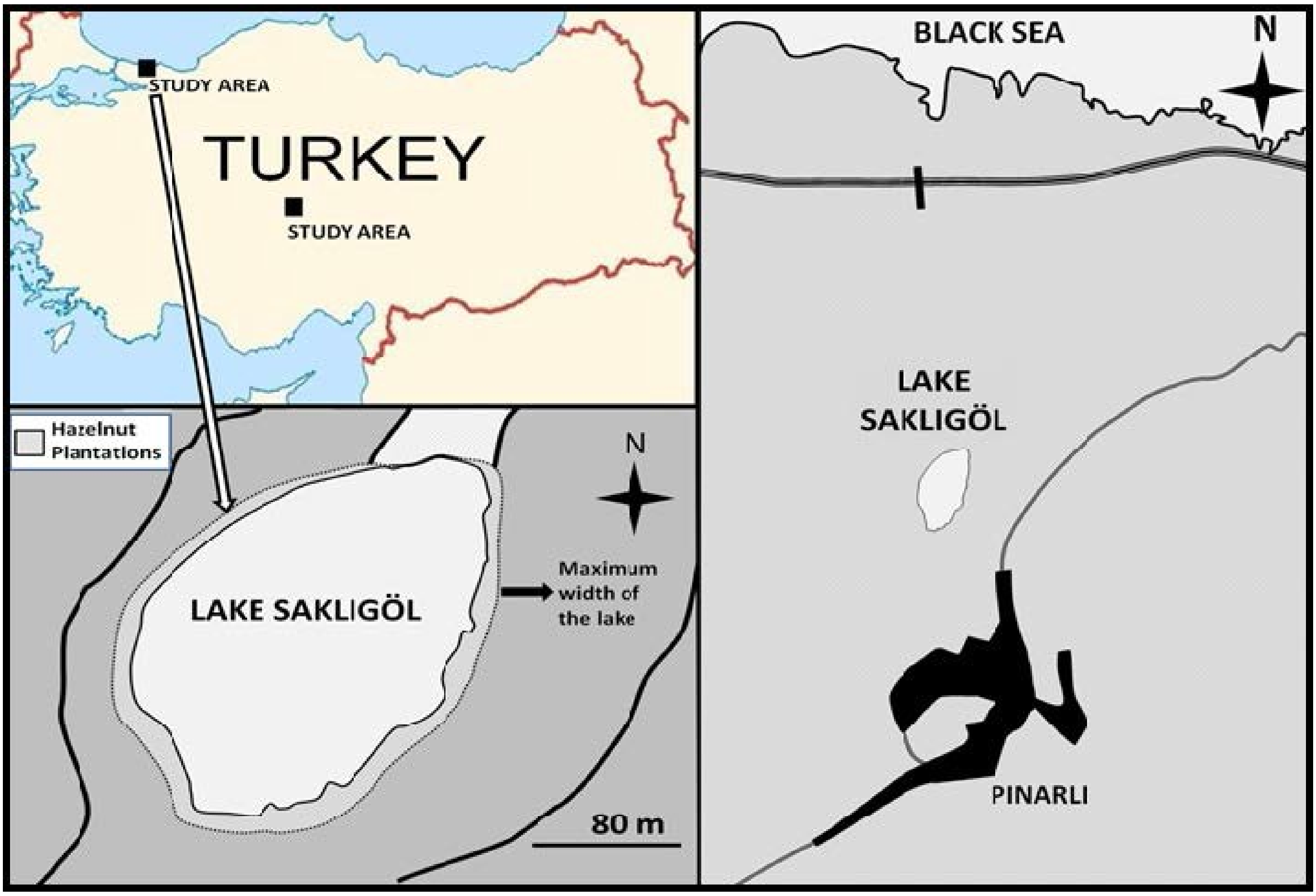


Fig 1: Collection sites of sponge species investigated in this study; Turkey

The climate of the Kandıra District is influenced by the climate of the Western Black Sea and Marmara regions. Therefore, the climate is affected by the Mediterranean climate and the Black Sea climate in Kandıra. According to Akman [18], the climate is generally temperate in the Kandıra District. There is significant rainfall throughout the year in Kandıra. Even the driest month still has a lot of rainfall. The mean annual temperature is 14.6 °C and annual precipitation is approximately 816.4 mm [18].

Eleven macrophyte species were recorded form lake Saklıgöl [16]. Submerged macrophytes are dominated in Saklıgöl [16]. The lake is surrounded by a cover of *Corylus avellana* L. (Common Hazel), *Laurus nobilis* Linnaeus (bay Laurel) and *Salix* (willow) shrubs. There is a small *Juglans regia* Linnaeus (Common walnut) plantation on eastern banks of Lake Saklıgöl [16]. Therefore, the proportions of present arable lands around the lake are quite low. *Cyprinus carpio* Linnaeus 1758 (Common carp), *Cyprinus carpio* Linnaeus 1758 (Mirror carp) and *Carassius gibelio* Bloch 1782 (Crucian carp) and *Gambusia affinis* Baird & Girard 1859 (Western mosquitofish) have been observed in the lake [16].

Samples were collected from Lake Saklıgöl between January-December 2013. The coordinates of the lake were obtained with a Garmin Etrex 12-channel GPS. *S. lacustris* samples were collected from hard substrates (at a depth of 50 cm attached on the karstic rock and at a depth 170 cm attached on a wooden stick) of the Lake Saklıgöl.

Gemmules and gemmoscleres play an important role in the identification of the freshwater sponge species. Therefore, gemmules were collected on fixed specimens of *S. lacustris* using a 0.025 mm hand net from the clear water of the lake. Samples were preserved in 95 % ethanol for taxonomic identification. Specimens were stored in 95% ethyl alcohol for taxonomic studies. The morphological characteristics of the specimens were studied by stereo microscope. One sample, which was dried and fixed to a slide was taken for spicule and tissue observations. Photographs of *S. lacustris* were taken from its natural habitat for facilitating the identification of species. A light microscope is usually sufficient for the morphological analysis of sponge spicules and gemmules. Moreover, photographs of gemmules and spicules photographs were taken under a light microscope for species identification. Main physico-chemical parameters (dissolved oxygen, saturation, water temperature, pH, salinity and electrical conductivity) were measured in situ with the help of a multimeter WTW 340i (Table 3). Light penetration (Water transparency) was measured using a 25 cm diameter Secchi disk. In this study, spicule preparations for sponge identification were obtained as follows: temporary preparations (for rapid or preliminary identification) were made by heating a mixture of sponge spicules and Perchloric acid ($HClO_4$) on a microscope slide. Spicules (megasclere, microsclere and gemmuloscleres) and gemmules were used for the description of *S. lacustris* [19, 20, 21, 22].

3. Results

The following references were reviewed for taxonomical determinations of the specimens: Penney & Racek [19] Ricciardi & Reiswig [20] Smith [21] and Manconi & Pronzato [22].

Phylum: Porifera Grant, 1836

Class: Demospongiae Sollas, 1885

Order: Haplosclerida Topsent, 1928

Family: Spongillidae Gray, 1867

Genus: *Spongilla* Lamarck, 1816

Species: *Spongilla lacustris* (Linnaeus 1759)

Morphological characters, skeletal architecture and the spicule composition of spicules have traditionally been of particular importance in the identification of sponges. Some of the parts of live samples are yellow. Fixed samples are yellowish green.

Greenish colored photosynthetic symbionts were observed on *Spongilla lacustris*. Because of this feature, greenish colored samples of *S. lacustris* quickly lost their color after fixation with ethanol. Performed macroscopic observations on morphological characteristics of *S. lacustris* samples have shown that specimens of *S. lacustris* have shape an encrusting, branched (tree branches), fragile and soft consistency in Lake Saklıgöl. We were observed that gemmules of *S. lacustris* water surface of the Lake Saklıgöl. Gemmules of *S. lacustris* can be mixed easily with plants pollens. Therefore, colors of gemmules were described in this study. Samples have green colour due to presence of zoochlorellae, inconspicuous scattered oscula, uneven surface and weak, fragile skeletons. Yellow colored sections have been observed on some parts of live sponges. Yellowish colored sections of sponge body can be traumatic sections of sponge body. However, its fixed samples were a yellowish green colour.

Smooth megascleres and small spines microscleres are present in *S. lacustris*. The structure of gemmules and their morphological traits are used to determine the family, genus and species [7]. Figure 2 shows gemmules, megascleres, microscleres and gemmuloscleres both matching the description of *S. lacustris* [19, 20, 21, 22].

The shape of the gemmules ranged from spherical to oval (Fig. 2 A, B, C). Gemmules colors can vary from a white to a white to yellow-orange or light green after alcohol fixation; light brown or brown after formalin fixation. Megascleres were straight sharply pointed and slightly curved smooth amphioxea (Fig. 2 F-H); gemmuloscleres (see also Fig. 2) were slightly to strongly curved acanthosis or strongyles, covered with large curved spines. Microscleres (Fig. 2 H) were slightly curved amphioxea, entirely covered with small spines.

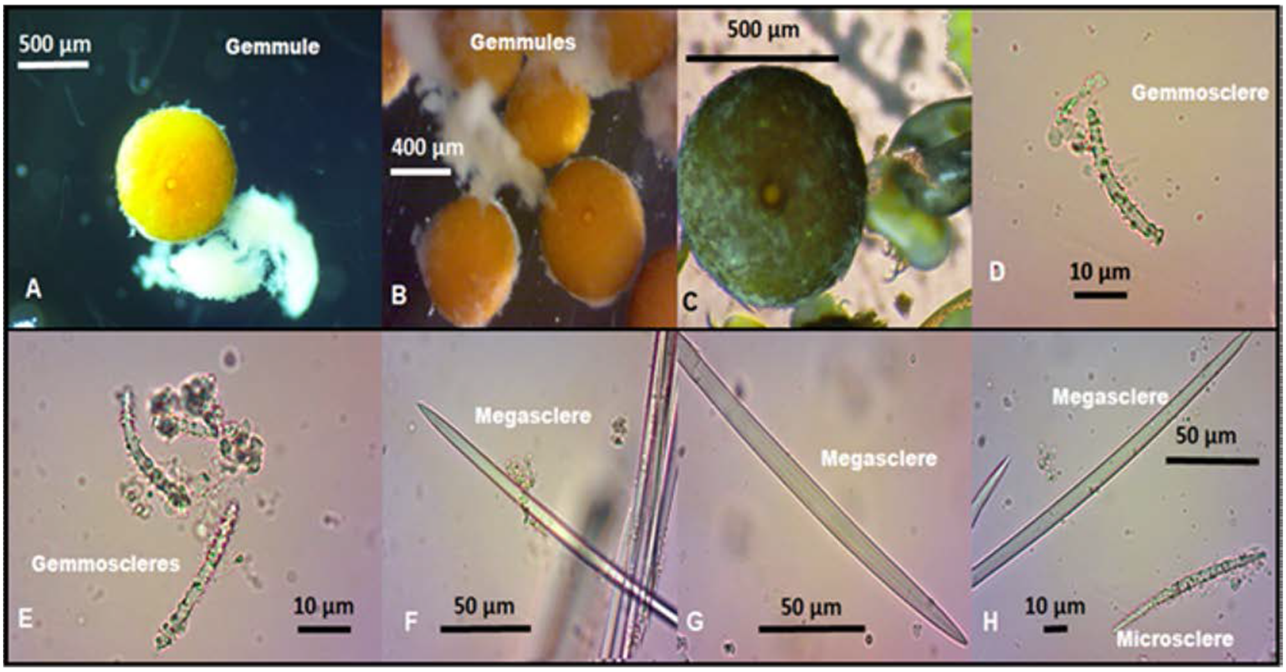


Fig 2: Spicules and gemmules of *Spongilla lacustris* (Linnaeus 1759) collected in Lake Saklıgöl **A-C:** gemmules; **D-E:** spiny gemmosclere; **F-G**: megasclere smooth oxea, **H:** megasclere and microsclere (acanthoxeas)

Also, the pictures of live samples of *S. lacustris* were taken (Figure 3). The gemmules allow sponges to survive extreme environmental conditions [23, 24]. Therefore, gemmules are a successful strategic structure for the continuation of the generation in the life cycle of *S. lacustris*. In the present study, the presence of gemmules inside sponge body was ascertained with an exhaustive dissection (see figure 3).

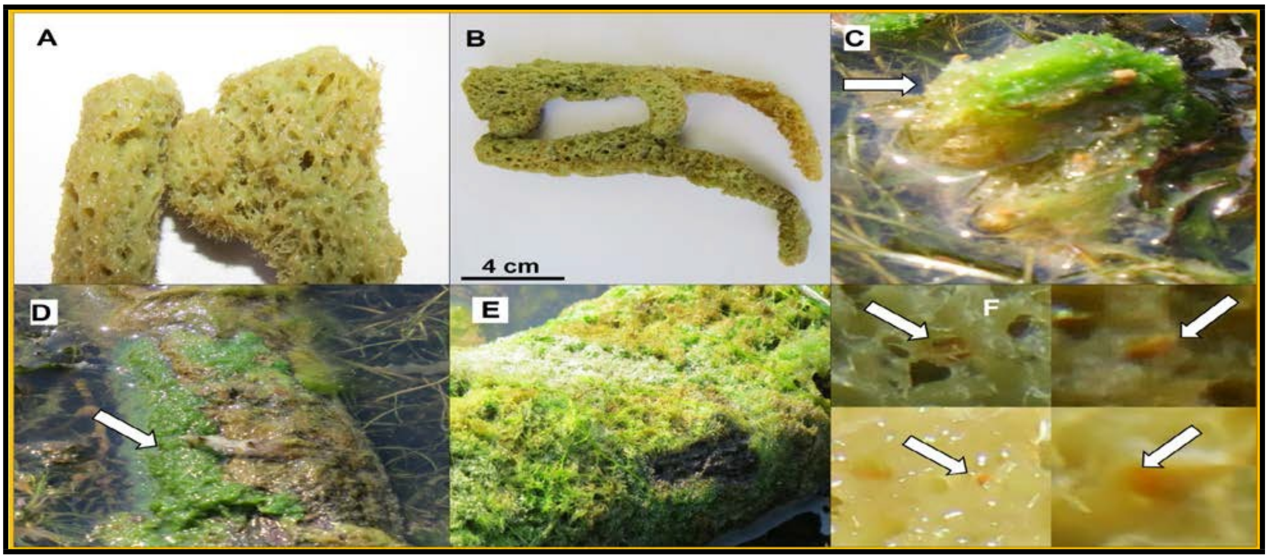
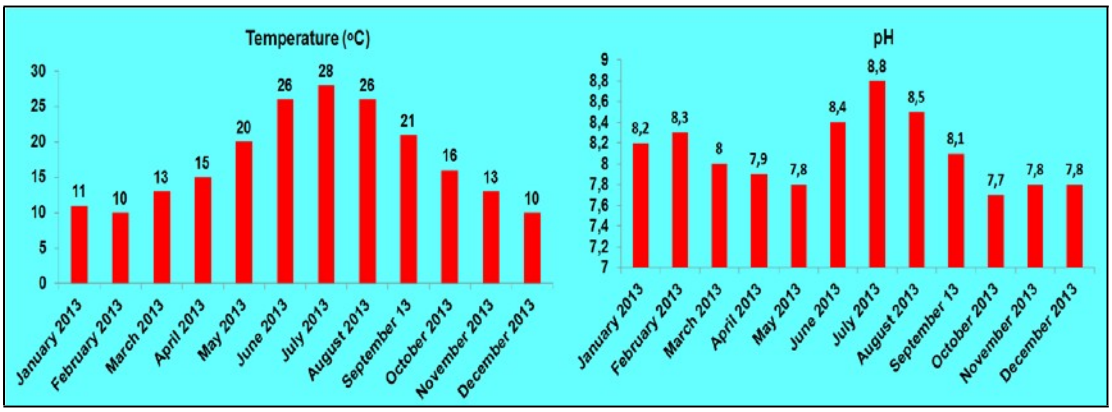


Fig 3: *Spongilla lacustris* (Linnaeus, 1759), **A-C:** the fixed specimens of the *Spongilla lacustris*; D-E: arrows shows live samples of *S. lacustris* located on woody sticks; **F:** gemmules seen on the fixed samples of *Spongilla lacustris*

Seasonal variations of physicochemical parameters measured in Lake Saklıgöl are shown in Table 1 and figure 4.

Table 1: Seasonal variation of some physicochemical parameters determined in Lake Saklıgöl. The scientific abbreviations shown are dissolved oxygen (DO, mg/L), water temperature (Temp, °C), oxygen saturation (Sat, %), standard hydrogen electrode (SHE, mV), electrical conductivity (EC, µS/cm), pH, salinity (Sal, ‰), Secchi depth (Secchi, cm); Standard Deviation (SD).

Months	Temp. (°C)	pH	SHE (mV)	Sal. (‰)	EC (µS/cm)	DO (mg/L)	Sat. (%)	Secchi (cm)
January 2013	11	8.2	-85	0.0	119	7.5	77	330
February 2013	10	8.3	-87	0.0	131	6.7	65	310
March 2013	13	8.0	-91	0.0	124	6.8	62	315
April 2013	15	7.9	-92	0.0	125	6.9	67	300
May 2013	20	7.8	-92	0.1	125	6.9	70	300
June 2013	26	8.4	-107	0.1	137	7.1	73	260
July 2013	28	8.8	-99	0.5	1370	6.7	67	200
August 2013	26	8.5	-90	0.1	167	6.5	61	190
September 2013	21	8.1	-69	0.1	233	8.3	96	200
October 2013	16	7.7	-58	0.1	156	7.8	85	280
November 2013	13	7.8	-59	0.0	141	7.2	75	300
December 2013	10	7.8	-57	0.0	133	7.4	76	310
SD	**6,6**	**0,3**	**17,0**	**0,1**	**355,1**	**0,5**	**10,1**	**50,2**
Maximum	**28**	**8,8**	**-57**	**0,5**	**1370**	**8,3**	**96**	**330**
Minimum	**10**	**7.7**	**-107**	**0**	**119**	**6.5**	**61**	**190**
Mean	**17**	**8.1**	**-82**	**0.0**	**246**	**7.1**	**72**	**275**



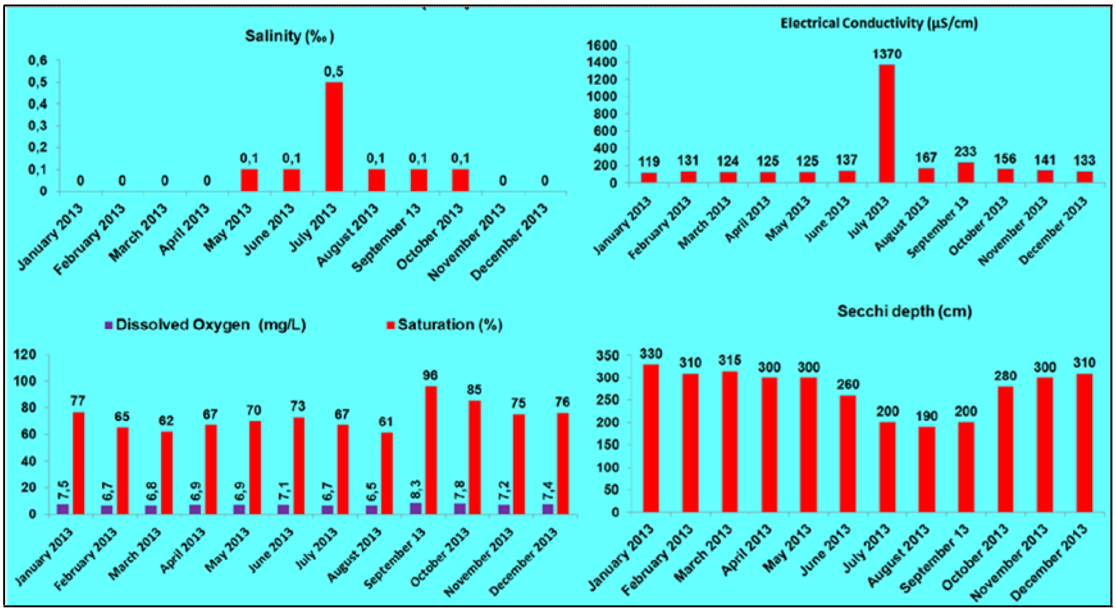


Fig 4: Seasonal fluctuations of some physicochemical parameters determined in Lake Saklıgöl.

4. Discussion

S. lacustris are an acid (soft) water form seldom occurring where the pH is above 7.0 or 7.3 [21]. *S. lacustris* is apparently the most successful species in surviving conditions high in calcium bicarbonate [25]. *S. lacustris* tolerate wide range in bound CO_2 and hydrogen ion concentration although they fare better under alkaline conditions. The results of the measured physicochemical variables show that the lake is alkaline (pH 7.7-8.8), well oxygenated (6.5-8.3 mg/L), freshwater (0.0-0.5‰), with low electrical conductivity (119- 1370 µS/cm), clear water (Secchi disc depth 190-330 cm) and hard water (see table 1).

S. lacustris may often be found in both the branching and encrusting form in the same water body [21]. According to Smith [20], the encrusting form is merely the earlier growth form that becomes branching as the sponge matures. The branching and encrusting form of *S. lacustris* were observed in Lake Saklıgöl. Even so, while branching forms of *S. lacustris* were more frequently observed in Lake Saklıgöl in the spring period on permanently submerged bedrock, its encrusting form were more frequently observed in lake in the spring period on roots, stems, branches and even leaves of trees or macrophytes.

Non-collectible organic particles from other organisms are quickly and effectively filtered from water by sponges [26]. For these reasons sponges can be valuable tools in monitoring aquatic systems, providing insight into both water quality and nutrient availability. According to Frost [27, 28, 29], finger sized *S. lacustris* can filter more than 125 litres per day. This feature of *S. lacustris* can explain why Lake Saklıgöl has clear lake water.

Species belonging to some Insecta orders (such as Neuroptera, Trichoptera, and Diptera) are predators on sponges [7]. Predator Caddisflies (Trichoptera) larvae were observed in samples from the bottom of Lake Saklıgöl. This situation suggests that this lake is attractive to predatory organisms fed by freshwater sponges.

The presences of Proteobacteria, Actinobacteria and Chloroflexi on *S. lacustris* have been reported [30]. According to Frost and Williamson [31], zoochlorellae make a major contribution to the growth of *S. lacustris*. *S. lacustris* are greenish color in light exposed habitats of Lake Saklıgöl, because, algal or cyanobacterial symbionts lives on *S. lacustris*. Due to the activity of its algal symbionts, *S. lacustris* may contribute significantly to the primary production of small lentic habitats [28]. Therefore, lakes (both where freshwater sponge is absent and where freshwater sponge is present) should be compared regarding primary productivity in Turkey.

S. lacustris is very common in Lake Saklıgöl, Kandıra District. Many studies have been carried out on the benthic fauna of wetlands of Turkey. The majority of those studies are on specific benthic groups, and many of them were performed for the identification of species belonging to some taxonomic groups. Freshwater sponges have been largely ignored in many benthic studies. Because, materials have been collected from previously determined stations during these types of studies. Bottom sediment grab sampling and hand nets have been used effectively for many decades to sample the top few centimeters of soft sediments in benthic studies. Also, the collection of a certain volume solely of benthic material can be obtained using bottom grab sampling and hand nets. Therefore, the probability of finding freshwater sponge is low with these techniques. Nevertheless, freshwater sponges can incidentally be found using these techniques. At the same time studies aimed at detecting all flora and fauna species present in wetland have been carried out to a limited extent in Turkey. Therefore, freshwater sponge species have only been observed in studies intended to determine all the fauna and studies were carried out for investigating and monitoring purposes by government agencies. In contrast, even though sponges were out of scope of these studies, freshwater sponges were not reported even if by chance in very comprehensive limnological studies [32] that were simultaneously carried out in many wetlands of Turkey. Scuba diving techniques are not usable in freshwater faunal studies due to the presence of many shallow and turbid lakes in Turkey. Conditions of the above mentioned gives and explains answer of this question: Why are freshwater sponges frequently not recorded from freshwater habitats by the researchers? According to Hutchinson [33], the species of the genus *Spongilla* can be found in deeper sections of the high light-transmission oligotrophic lakes, but they can also be found at a depth of a few centimeters in the less light-transmission lakes. *S. lacustris* was found to be abundant at depth of several centimeters in coastal parts of the Aslantaş Dam Lake by Fındık [34]. Both our data and the data of Fındık [34] show the existence of *S. lacustris* in shallow littoral sections of Lake Saklıgöl. Living sponges were found during this study in May, June, July and August in 2013. The specimens of *S. lacustris* collected in autumn were without gemmules and only those collected in summer had developed gemmules.

Also, unchecked urbanization, dam construction, draining of wetlands, poaching, and excessive irrigation are the most widespread threats to biodiversity of Turkey [35]. Therefore, researchers should give more importance to determine the sponge fauna in the all limnological and faunistic studies

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