Ecological requirements of Ostracoda (Crustacea) in Deştin, Dipsiz and Pınarbaşı karst springs (Yatağan, Muğla, Turkey)

Selçuk Altınsaçlı, Songül Altınsaçlı, Ferda Perçin-Paçal

Abstract

The karstic freshwater springs of Pınarbaşşı, Dipsiz, and Deştin are located in the Yatağan District of Muğla in the southwest of Turkey. Habitat features and ostracod populations were monitored during January to December, 2008 at monthly intervals in three karstic springs (one limnocrene, two rheocrene). We analyzed the diversity and distribution of ostracod assemblages associated with the three springs and the ecological factors reported to affect ostracod occurrence. For this purpose, the main physicochemical parameters of the habitat, including salinity, electrical conductivity, temperature, pH, dissolved oxygen, and oxygen saturation were recorded. Similar to previous findings, measurement results of physicochemical parameters indicate that springs are stable and resource-rich systems due to their physicochemical characteristics remaining stable. Eight species were recorded (Candona neglecta, Heterocypris incongruens, Heterocypris salina, Herpetocypris chevreuxi, Psychrodromus olivaceus, Prionocypris zenkeri, Ilyocypris gibba, Pseudocandona marchica) in two types of springs: limnocrene and rheocrene, while Candona neglecta, Herpetocypris chevreuxi, and Psychrodromus olivaceus were found in the all three springs. Candona neglecta was common in both types of springs.

Keywords: Ostracoda, spring, Distribution, Ecology, Candona neglecta

1. Introduction

Karstic rocks and its groundwaters are common in the eight circum-mediterranean countries of Turkey, Greece, Albania, Montenegro, Croatia, Slovenia, Spain, Algeria, Tunisia, Syria, Lebanon, Italy and Southern France. The most important karst features in Turkey are developed in Mesozoic limestones (partly dolomites) which are characterized by sporadic primary permeability and most often by a great secondary permeability. Drinking and agricultural irrigation water is supplied from karstic aquifers to the millions of people worldwide who live in karstic areas, whereas, these valuable water resources have been impacted by large-scale human activity and contamination. Drinkable freshwater springs are scarce resources in the world at large and in Turkey specifically. Groundwater resources are controlled and preserved for a sustainable supply of drinkable and irrigation water because of rapid population growth, global climate change, industrialization, and dwindling water resources. Sustainable use of freshwater (agriculture, industry, domestic use), huge savings, and effective water management is possible. Young alluvial deposits in the Yatağan Plain contain permeable and high permeable strata [1]. The uppermost section of rocks belonging to the Mesozoic age around springs in the Menderes Massif region in western Anatolia, Turkey have formed schists and faulted and fractured marble. Dipsiz and Pınarbaşşı springs are accepted as being karstic springs [2], Deştin Spring is a karst spring according to our observations. Therefore, Dipsiz and Pınarbaşşı resources in the area are accepted as being karstic springs [3]. Dipsiz, Deştin and Pınarbaşşı resources are a karst springs according to findings of this study. According to Baba [4], the high flowrate of water sources is a result of the impact of tectonic movement and karstification, producing such springs as Dipsiz Spring (1000/sec) and Pınarbaş Spring (450/sec). These aquifers are used for both domestic consumption and agricultural irrigation. Yatağan District is located in the western part of the Aegean region, near the city of Muğla, and has a Mediterranean climate: a hot and arid climate exists in the summer and thus drinking and agricultural irrigation water use peak in the summer months in this water-scarce area. The chemical and physical properties of the three springs are controlled primarily by the aquifer. Small differences in chemical parameters have been determined as a result of studies.
Springs are important for ecological studies. Aquifer discharge was divided into three classes: rheocrene, limnocrene, helocrene by Hynes [3]. Most springs are of the helocrene type and are relatively small in size. According to Särkkä et al. [3], springs can be considered as an ecotone between hypogean and epigeic aquatic environments and as discharge points of groundwater. Therefore, springs have relatively constant physical and chemical conditions. According to Helfrich et al. [4], most springs generally exhibit uniformity in physicochemical conditions throughout the year, but they should be monitored over time to understand their physicochemical conditions. The Yatağan thermal power plant is in the north-west of the agricultural Yatağan flat plain (37° 20’ N latitude, 28° 06’ E longitude, elevation 300 m), 23 km from Muğla, Turkey (in the south of the Aegean region) [5]. The thermal power plant was established for the evaluation of low-calorie lignite coals. One unit of the thermal power plant was activated in 1984, a second unit in 1981, and three more units in 1983 [1]. Karstic springs and groundwater resources found in Yatağan District and around it have faced the dangers of pollution due to the activities of the thermal plant as well as other anthropogenic activities [1]. According to Demirak et al. [6], low metal concentrations were determined in all caught fish samples from Dipsiz Spring, although it seems this does not have any toxicological effects on human health. Ostracoda a widespread micro-crustacean group found in almost all types of aquatic habitats, have been shown to be important biological, ecological, and geochemical indicator. The present study focused on the distribution of ostracod species and environmental factors affecting their distribution in three springs. Recent studies have clearly shown that ostracod species can be used as an indicator of water quality, and that there are close relationships between ostracod species diversity and distribution with abundance and nutrient richness [e.g. 7-12]. Ostracodes are important indicators of the structure and function of freshwater ecosystems and their ecological status [7]. The ecology of freshwater ostracods living in springs has been studied by numerous researchers [e.g. 7, 13-19]. Many ostracod species have been determined in springs of Turkey that flow into lakes and wetlands [10, 20-29]. This present study sought to find connections between previously studied springs and recently studied springs in terms of environmental factors shaping the ostracod community. Ostracod diversity is one of the most important ecological parameters in water quality and meiobenthic biodiversity assessment, because it is strongly affected by environmental conditions. The investigated three springs are important habitats for host various forms of life outside water use of agricultural irrigation and drinkable. The springs are needs to protection wherefore is an important habitat for many species. All kinds of scientific studies performed on these springs will protection of springs and will contribute to the use of springs in a conscious way. The main aim of this study was to collect ostracod samples from three of karstic springs of Yatağan, Muğla (Southwestern Turkey) to obtain both an insight into the factors affecting ostracod distribution as well as evaluated to their occurrence in different habitats. In other words, during the present study we investigated to: (1) study the ostracod community composition in a three karstic spring; (2) discuss ecological requirements of ostracods, focusing on the values of physicochemical variables; and (3) increase our knowledge of the these springs conservation status.

2. Material and Methods
This study was performed in during January to December, 2008 at monthly intervals in three karstic springs in Yatağan District.

2.1 Description of the site
The distribution of freshwater ostracod populations was studied together with physicochemical variables in three karstic springs in Yatağan District. The modern climate in Yatağan is Mediterranean, with hot, dry summers and cool, wet winters. Mean January, July, and annual temperatures in the basin center are 5.5, 33.3, and 14.9 °C, respectively [30]. Mean annual precipitation is 94.4 kg/m² in the basin [10]. The natural vegetation exists in the basin. Herbaceous plants and shrubs fill the open spaces between trees, while the area surrounding the springs is used for agriculture and grazing. The three springs studied are located around Yatağan District (Fig. 1, 2, 3). Of the three springs sampled, one spring is limnocrene and two are rheocrene (Fig. 1, 2, 3). All three have a Ca-HCO₃ water type, since calcium is one of the most abundant elements in the limestone rock surrounding springs, is a major constituent in groundwater, and is a primary contributor of water hardness. Pınarbaşı Spring, particularly, is a well-known tourist attraction, and is consequently under pressure due to heavy touristic activity.

Pınarbaşı spring: The Pınarbaşı spring is a calcium carbonate-rich, rheocrene –type freshwater spring located at an altitude of 364 m a.s.l. in the southwestern Turkey. Rheocrene spring Pınarbaşı is situated (37° 17’42.95” N 28° 07’37.14” E, 364 m a.s.l) 5 km southwest of Yatağan District (Fig 1, 2, 3). Pınarbaşı Spring is located vicinity of the ancient settlement Boziyüük belonging to Ottoman Empire Times and it is still the most important natural drinking water sources at present. Flowrate of Pınarbaşı spring is 450 l/sec [1]. This aquifer is used for domestic and irrigation. The flow velocity rate of spring water falls due to the lack of rain in the summer or almost completely dry up during the summer months in recent year. According to statement of managers of Pınarbaşı Touristic facilities, Pınarbaşı spring was completely dried in summer of 1994, 1996 and 2008 years. There is meaningful relationship between rainfall and spring discharge in Pınarbaşı spring. Spring water is Ca-HCO₃ type. Spring water to discharge into a stream channel and this stream is flowing to Yatağan Creek.

Dipsiz Spring: The Dipsiz spring is a calcium carbonate-rich, limnocrene –type freshwater spring located at an altitude of 304 m a.s.l in the southwestern Turkey. Limnocrene spring Dipsiz is situated (37° 22’29.61” N 28° 05’31.85” E, 304 m a.s.l) 5 km Northwest of Yatağan District (Fig 1, 2, 3). Dipsiz spring has the highest flow velocity rate (1000 l/sec) between all of three springs (see Fig. 1) [7, 31]. The main fish species are Barbus plebejus, Barbus capito, C. capoeta, Lepomis gibbosus [6]. Discharge water of Dipsiz spring used for the water supply to the Yatağan Thermal Power Plant, and provision of drinking water to five villages. Spring water is Ca-HCO₃ type. According to our observation that flow velocity rate of discharge water does not decrease in summer in Dipsiz Spring. Drinking water are supplied to five villages from Dipsiz spring. There is a depth pool of Limnocrene Dipsiz spring. Accumulated water in this spring pool creates a source brook, and this brook is flows into Dipsiz Creek. This spring is also known as the Bottomless Spring. Spring is also called the Dipsiz (Bottomless) spring due to its deep pool.

Deştin Kaynağlı: The Deştin spring is a calcium carbonate-rich, rheocrene –type freshwater spring located at an altitude of 638 meters above sea level in the southwestern Turkey. Rheocrene spring Deştin is situated (37° 20’54.62” N 28°16’00.38” E, 638 m a.s.l) 9.5 km southwest of Yatağan District.
(Fig. 1, 2 and 3). Flow velocity rate of Deştin spring is 1000 l/sec Discharge waters of Deştin spring use for provision of drinking water to three villages and agricultural irrigation. Spring water is Ca-HCO₃ type. According to our observation that flow velocity rate of discharge water does not decrease in summer in Deştin Spring. Drinking water is supplied to three villages from Destin spring. Spring water to discharge into a stream channel and this stream is flowing to Yatağan Creek

Fig 1: Distribution of springs in the study area.

Fig 2: Location of three springs (the dashed-line circle= study site; dashed straight lines: flow direction of the spring waters). Digital image is produced from Google Earth 4.3.

Fig 3: Photographs of Pınarbaşı Spring (A–B), Dipsiz Spring (C) and Deştin Spring (D)
The area around the springs is dominated with Calabrian pine (*Pinus brutia* Tenore), oriental plane (*Platanus orientalis* L.), field elm (*Ulmus minor* L.) white willow (*Salix alba* L.), Persian walnut (*Juglans regia* L.) and black poplar (*Populus nigra* L.) trees. Shrubby plants contain more than 40 species in Yatagan such as *Nerium oleander* L., *Pistacia terebinthus* L., and *Arbutus unedo* L. [5]. Macrophyte species found in the three springs are shown in Table 1.

### Table 1: List of macrophyte species found in the in three springs.

<table>
<thead>
<tr>
<th>Macrophyte species</th>
<th>Pınarbaşı Spring</th>
<th>Dipsiz Spring</th>
<th>Destin Spring</th>
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<td>Fontinalis antipyretica Hedw.</td>
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<td>Nasturtium officinale W.T. Aiton</td>
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<td>Mentha aquatica L.</td>
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<td>Potamogeton nodosus Poir.</td>
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<td>Lemna minor L.</td>
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<td>Mentha aquatica L.</td>
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<td>Phragmites australis (Cav.) Trin. ex Steud.</td>
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#### 2.2 Sampling

Ostracods were collected from the springs (< 1 m) using a hand net (mesh size 250 μm) at each site. Two hundred milliliters of sediment (with submerged aquatic plants) was collected from a depth of 10 to 60 cm (over an area of 1 m²), and the collected samples were kept in polyethylene jars (250 ml containers) containing 4% formaldehyde solution and fixed in situ. In the laboratory, each sample was filtered through four standard sized sieves (0.5, 1.0, 1.5 and 2.0 mm mesh size) under tap water and then preserved in 70% alcohol for further studies. Subsequently, specimens were preserved in 70% ethanol and glycercine (1:1 ratio) and the retained material transferred to Petri dishes. Ostracod dissections were prepared according to the methodology of Namiotko et al. [32]. The number of adult and juvenile individuals to each identified ostracod species was counted under a stereomicroscope. Specimens were determined using the taxonomic publications by Bronshtein [33], Hartmann and Puri [34], Meisch [35] and Karanovic [36]. Seven major water variables (temperature, pH, electrical conductivity, dissolved oxygen, saturation, salinity and redox potential) were measured in situ before sampling with a WTW340i multimeter at all study sites. Table 2 includes both ecological and environmental data during sampling from each spring.

### Table 2: Taxonomical and environmental data collected during sampling from each spring. Abbreviations of species are as follows: HC: *Herpetocypris chevreuxi*, HS: *Heterocypris salina* HI: *Heterocypris incongruens*, PZ: *Prionocypris zenkeri*, IG: *Ilyocypris gibba*, CN: *Candona neglecta* Sars, 1887, PM: *Pseudocandona marchica* and PO: *Psychrodromus olivaceus*. Environmental variables measured include: Water redox potential (Eh), pH, percent oxygen saturation (% S), Dissolved oxygen (DO (mg/L), Electrical conductivity (EC (mS/cm), Salinity (Sal (‰)) and water temperature (T(w)). NSS: Number of specimen in station. TNS: Total number of specimen in all stations.

#### Pınarbaşı Spring

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<th>Time (h)</th>
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#### Destin Spring

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2.3 Statistical analysis

Correlations between species, environmental variables, and species and environmental variables were analyzed by a two-tailed nonparametric Spearman coefficient correlation performed with the SPSS 10.0 software program [37]. Significant results were determined at 0.01 and/or 0.05 critical levels.

Classification of ostracod species and sampling stations were achieved using the Bray–Curtis similarity coefficient to construct dendrograms. Species richness and diversity of sampling stations were calculated using the Shannon–Weaver diversity index.

Binary (presence–absence) data were used to show relationships among species by means of a Bray–Curtis UPGMA (Unweighted Pair Group method with Arithmetic Mean) analysis as provided by the Multivariate Statistical Package (MVSP) program, version 3.1 [38]. Relationships between ostracod site assemblages were examined using UPGMA hierarchical clustering based on the Bray–Curtis similarity coefficient MVSP, version 3.1 [38].

A Bray–Curtis cluster analysis was used to obtain the species-station and station-species similarity in stations, (with log (x+1) transformation performed before the analysis) [39].

The Shannon–Weaver index, which combines information on species richness (number of species) and how individuals are distributed among species, was calculated. A living ostracod species database of seasonal samples from the three springs was calculated by means of the (log 2) Shannon–Weaver index (H) [40].

3. Results

A total of 733 ostracod specimens, consisting of 8 species from seven genera and three families, were identified from three springs in two types of springs (Limnocrene, Rheocrene) in the Yatağan District (Muğla, Turkey). The 8 species were: Herpetocypris chevreuxi (Sars, 1896), Heterocypris salina (Brady, 1868), Heterocypris incongruens (Ramdohr, 1808), Prionocypris zenkeri (Chyzer and Toth, 1858), Ilyocypris gibba (Ramdohr, 1808), Candonia neglecta Sars, 1887, Pseudocandona marchica (Hartwig, 1899) and Psychrodromus olivaceus (Brady and Normal, 1889). C. neglecta, H. chevreuxi and P. olivaceus were found in all three springs. C. neglecta was common in all types of springs. Relationships between eight species and seven environmental variables has been analysed by Spearman Correlation analysis in Table 3.

### Table 3: Spearman correlation analyses showing significant relationships between eight species and seven environmental variables. For abbreviations, see Table 2. *P<0.05, **P<0.01.

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</tr>
<tr>
<td>Rheocrene</td>
<td>733</td>
<td>0.855</td>
<td>0.745</td>
<td>0.585</td>
<td>0.617</td>
<td>0.565</td>
<td>0.556</td>
<td>0.510</td>
<td>0.510</td>
<td>0.510</td>
<td>0.510</td>
<td>0.510</td>
<td>0.510</td>
<td>0.510</td>
<td>0.510</td>
</tr>
</tbody>
</table>

The Shannon–Weaver index, which combines information on species richness (number of species) and how individuals are distributed among species, was calculated. A living ostracod species database of seasonal samples from the three springs was calculated by means of the (log 2) Shannon–Weaver index (H) [40].

3. Results

A total of 733 ostracod specimens, consisting of 8 species from seven genera and three families, were identified from three springs in two types of springs (Limnocrene, Rheocrene) in the Yatağan District (Muğla, Turkey). The 8 species were: Herpetocypris chevreuxi (Sars, 1896), Heterocypris salina (Brady, 1868), Heterocypris incongruens (Ramdohr, 1808), Prionocypris zenkeri (Chyzer and Toth, 1858), Ilyocypris gibba (Ramdohr, 1808), Candonia neglecta Sars, 1887, Pseudocandona marchica (Hartwig, 1899) and Psychrodromus olivaceus (Brady and Normal, 1889). C. neglecta, H. chevreuxi and P. olivaceus were found in all three springs. C. neglecta was common in all types of springs. Relationships between eight species and seven environmental variables has been analysed by Spearman Correlation analysis in Table 3.
The results of the Spearman coefficient correlation analysis show a meaningful positive correlation in the relationships between all ostracods species living in the three springs. A positive correlation was also observed between ostracod species and environmental variables in the Deştin and Pınarbaşı springs (Table 3). Except for pH, a positive correlation was observed between ostracod species and physicochemical variables in Destin Spring (Table 3). Salinity and conductivity were not a remarkable environmental variable in all three springs. Shannon-Weaver diversity index \( (H') \) values of the three springs has been shown in Fig 4.

![Fig 4](image1.png)

**Fig 4:** Seasonal Shannon–Weaver diversity index \( (H') \) values of the three springs January-December 2008.

According to the Shannon–Weaver index, the highest level of diversity (2.938) was found in August 2008 and the lowest (0) in January and February 2008 in Dipsiz Spring. The highest level of diversity (2.298) was found in August 2008 and the lowest (0) at January, February, and December 2008 in Pınarbaşı Spring. For Deştin Spring, the highest level of diversity (1.971) was found in September 2008 and the lowest (0) in January, February, March, April, November, and December 2008 (Fig. 4). For all stations, the highest diversity was found in the summer and autumn months. The number of individual ostracods increased due to the reduced water flowrate in the summer.

The similarity between the three springs was demonstrated in binary (presence-absence of ostracod species) data using the Bray-Curtis similarity index and the UPGMA method (Fig. 5). Based on the species binary (presence-absence) data, the UPGMA dendrogram is separated into two main clusters. The unweighted pair group’s mean average (UPGMA) dendrogram similarity of springs analysis places the species into two main clusters.

![Fig 5](image2.png)

**Fig 5:** Two clustering groups in UPGMA analysis for the three springs based on binary data (presence–absence).
Regarding the results of the Bray–Curtis similarity analysis, the first cluster existed in the fast-flowing Deştin Spring (a rheocrene spring). The second cluster existed in the slow-flowing Pınarbaşı Spring (rheocrene) and Dipsiz Spring (limnocrene). The Bray–Curtis similarity index results showed that the ostracoda fauna of Pınarbaşı Spring were most similar to Dipsiz Spring (76.9%) and least similar to Deştin Spring (66.6%). The Ostracoda fauna of Deştin Spring were similar to Dipsiz Spring (66.6%), similarly Pınarbaşı Spring.

The similarity of ostracod species was demonstrated in binary (presence–absence of ostracod species) data using the Bray–Curtis similarity index and the UPGMA method (Fig. 6). Based on species occurrence, UPGMA was able to cluster 16 species into two groups with two species (I. gibba, P. zenkeri) in the first group and six (Pseudocandona marchica, Heterocypris salina, Heterocypris incongruens, Herpetocypris chevreuxi, Psychrodromus olivaceus, and Candona neglecta) in the second.

The results of this study reveal that the waters of the three springs have a relatively uniform temperature regime, varying only a few degrees throughout the year (see Table 1). Eight ostracod species (C. neglecta, H. incongruens, H. salina, H. chevreuxi, P. olivaceus, P. zenkeri, I. gibba, P. marchica) were found in all three springs. Of these, a continuous occurrence of C. neglecta was found in all three springs. C. neglecta was common in all three springs. Maximum, minimum, and mean values of environmental variables measured from the three springs are shown in Table 4.

### Table 4: The maximum, minimum and mean values of environmental variables in the three springs.

<table>
<thead>
<tr>
<th></th>
<th>T(w)</th>
<th>pH</th>
<th>Eh</th>
<th>Sal</th>
<th>EC</th>
<th>DO</th>
<th>%S</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Pınarbaşı Spring</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Max.</td>
<td>17.7</td>
<td>7.35</td>
<td>-28</td>
<td>0.1</td>
<td>699</td>
<td>6.56</td>
<td>75.2</td>
</tr>
<tr>
<td>Min.</td>
<td>14.9</td>
<td>7.13</td>
<td>-32</td>
<td>0</td>
<td>220</td>
<td>5.11</td>
<td>54.2</td>
</tr>
<tr>
<td>Mean</td>
<td>16.35</td>
<td>7.22</td>
<td>-29.3</td>
<td>0.04</td>
<td>402.4</td>
<td>5.91</td>
<td>61.5</td>
</tr>
<tr>
<td><strong>Dipsiz Spring</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Max.</td>
<td>18.8</td>
<td>7.35</td>
<td>-21</td>
<td>0.1</td>
<td>680</td>
<td>5.82</td>
<td>68.3</td>
</tr>
<tr>
<td>Min.</td>
<td>13.7</td>
<td>6.91</td>
<td>-24</td>
<td>0.1</td>
<td>423</td>
<td>4.98</td>
<td>50.4</td>
</tr>
<tr>
<td>Mean</td>
<td>15.75</td>
<td>7.11</td>
<td>-22.7</td>
<td>0.1</td>
<td>531.4</td>
<td>5.505</td>
<td>60.1</td>
</tr>
<tr>
<td><strong>Deştin Spring</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Max.</td>
<td>16.7</td>
<td>7.45</td>
<td>-27</td>
<td>0</td>
<td>432</td>
<td>5.86</td>
<td>55.9</td>
</tr>
<tr>
<td>Min.</td>
<td>13.7</td>
<td>7.11</td>
<td>-30</td>
<td>0</td>
<td>389</td>
<td>5.18</td>
<td>48.2</td>
</tr>
<tr>
<td>Mean</td>
<td>15.30</td>
<td>7.24</td>
<td>-27.8</td>
<td>0</td>
<td>412.5</td>
<td>5.50</td>
<td>51.6</td>
</tr>
</tbody>
</table>

Dissolved oxygen content is an important parameter in water quality assessment, and measured values in water reflect the physical and biological processes that prevail. The dissolved oxygen content in natural water varies with salinity, temperature, the water’s ionic composition, atmospheric pressure, and the photosynthetic activity of algae and macrophytes. The maximum dissolved oxygen values of Pınarbaşı Spring were recorded in July 2008 (6.56 mg/L) and the minimum in January 2008 (5.11 mg/L). The maximum dissolved oxygen values of Dipsiz Spring were recorded in March 2008 (5.82 mg/L), the minimum in August 2008 (4.98 mg/L). The maximum dissolved oxygen values of Deştin Spring were recorded in August 2008 (5.86 mg/L), the minimum in December 2008 (5.18 mg/L). Dipsiz, Deştin, and Pınarbaşı springs are freshwater, alkaline, cold-water, and oxygen-rich springs according to the assessment of the maximum, minimum, and mean values of environmental variables measured in the three springs (see Table 1 and 3). Ostracod species determined in the present study have been reported many times from the same types of springs by many other researchers.

In the present study, the recorded dissolved oxygen, salinity, electrical conductivity, temperature, and pH values are sufficient and tolerable for all ostracod species found in the three springs.
4. Discussion
C. neglecta, H. salina, H. incongruens, P. marchica, P. zenkeri, H. chevreuxi, P. olivaceus and I. gibba (synonymous of Ilyocypris biplicata) have been reported of the 63 major wetlands and their various habitats (such as fresh or brackish water lakes, spring-fed wetlands and coastal wetlands) of Turkey, respectively, in from of them 44, 40, 32, 23, 22, 17, 15 and 8 [27]. Once again, the results of this present study indicated that cosmopolitan and eurytopic ostracod species have a higher tolerance to Turkish wetlands than other geographical regions.

C. neglecta is one of the most common bottom-dependent species in the aquatic habitats of Turkey [25-28, 41, 42]. C. neglecta occurs in a variety of freshwater environments but is common in springs [27]. It has a wide tolerance to pH levels and dissolved oxygen concentrations [38]. Also, it has been reported from slightly saline inland and coastal water [43], and in a high altitude (1903 m) cirque Lake Kartal [40] and coastal oligomesosaline Sarsu Lagoon [41]. C. neglecta is a non-swimming, crawling, and digging species, so it is not surprising to find this species in the three springs in Yatağan District. Altınsaçlı’s findings [27] show that C. neglecta has a high level of tolerance to changes of physicochemical parameters in a variety of water bodies in Turkey. It was reported in the sandy substrate of Karagöl volcanic crater lake (Dikili, Izmir) by Altınsaçlı and Altınsaçlı [44] and in the sinkhole lake, Lake Meyil (in Konya) [27].

I. gibba (Ramdohr, 1808) has been synonymized with Ilyocypris biplicata (Koch, 1838) [39]. We determined the non-noded form (Ilyocypris forma biplicata) of I. gibba. This cosmopolitan species has a wide range of occurrence in a variety of water bodies such as ponds, streams, paddy fields, and lakes [35]. I. gibba is one of the most common species in the aquatic habitats of Turkey [25-27, 41, 42] and is one of the ostracod species which prefers springs [35]. This swimmer species is often found on muddy or sandy bottoms of habitats [31, 45]. I. gibba is a good swimmer, and is a crawling and digging species.

P. zenkeri is an inhabitant of slow-flowing streams and springs like Dipsiz Spring. P. zenkeri is almost cosmopolitan: It has been found in springs in Italy [40]. For example, P. zenkeri is mostly found in rather cold, running water [15]. It has been reported in the major wetlands and their connected springs in Turkey [24, 26, 27, 28]. P. zenkeri is a bottom-dependent and non-swimmer ostracod species due to reduced swimming setae [35].

Also, it is a crawling and digging species. H. chevreuxi is found in the littoral zones of lakes and slow-running streams in Europe, around the Mediterranean in Turkey, and in Iran, Asia, South Africa, and South America [35]. It is almost a cosmopolitan species. It has been reported in major wetlands and their various habitats [26-29]. H. chevreuxi is a bottom-dependent species and known as to be an active swimmer.

H. incongruens is a well-known cosmopolitan and euryplastic species. It has been reported of the 32 major wetlands and their connected springs of Turkey [24, 28, 41, 42]. It can be found in all kinds of fresh and brackish waters. It has been reported from an oligosaline (0.8-1 %) habitat of Lake Bafa by Altınsaçlı [10]. Meisch [35] described this ostracod as an indicator of pollution in freshwater habitats. H. incongruens is good swimmer and is bottom-dependent, crawling, and digging species.

H. salina is a cosmopolitan, euryplastic, and halobiont species, which has been recorded in pure freshwater springs [11, 20, 24, 28, 47] and brackish waters [11, 25, 27, 48], habitats of Turkey. Although known as a halophilic species, it alone is not a reliable brackish water indicator. H. salina is an ostracod species known to good swimmer species. A bisexual population of this species was found in the discharge water of the Dikili muddy thermal spa in Dikili District (Izmir) (coordinates: 39° 03’ 35.1” N, 26° 55’ 17.9” E; 38.5-39.2 °C; pH: 6.77; dissolved oxygen: 5.6 mg l-1; redox potential: -32 mV; electrical conductivity: 2.55 mS/cm; substrate type: muddy-sand) in 2008 and in a natural pond formed by flowing water from a thermal spa in Sarayköy District, Denizli Coordinates: 37° 55’ 14.2” N, 28° 49’ 43.3” E; water temperature: 38.6-39 °C; pH: 6.46; dissolved oxygen 8.35 mg l-1; redox potential: -36 mV; electrical conductivity: 2.55 mS/cm; substrate type: muddy-sand) in 2008 (unpublished data of Altınsaçlı). It was also reported in the sandy substrate of Karagöl volcanic Crater Lake [47] such as C. neglecta, and also reported in Acıgöl and Meke Maar Lakes (Karapınar, Konya) [27].

P. olivaceus mostly prefers flowing water from springs, springs ponds and spring brooks that are fed by springs [35]. P. olivaceus is bottom-dependent due to lack (or reduced) of swimming setae on second antenna [35]. P. olivaceus can show cosmopolitan characteristics with a wide range of tolerances, but it can still display selective behavior in some types of habitat and physicochemical conditions. It is one of the most common ostracod species in the aquatic habitats of Turkey [10, 24-28].

P. marchica is mostly found in springs and spring brooks [35]. It may also be present in temporary water bodies, although preferring permanent habitats [55]. A non-swimming and bottom-dependent species, it has been reported in the major wetlands and their connected springs in Turkey [24-27].

C. neglecta and C. opalatmica are the most commonly found species in Palearctic springs [49]. C. neglecta, H. incongruens, H. salina, P. zenkeri and I. gibba were found on the sandy-mud substrate of springs feeding Lake Kayı and Lake Balıklı [12]. These two lakes are spring-fed lakes. The geographical distribution and tolerance limits for environmental variables of ostracod species determined in that study can also be applied to our findings. C. neglecta, I. gibba, H. incongruens, and H. salina are cosmopolitan species; P. zenkeri, H. chevreuxi, and P. olivaceus are almost cosmopolitan, while P. marchica is widespread in different habitats of the Holarctic region. Cosmopolitan freshwater ostracod species have wide geographical distribution as they are adapted to a wide range of environmental conditions compared with non-cosmopolitan species [35]. Being highly tolerant to environmental variables suggests those species’ survival chances are increased. Common and cosmopolitan species occupy a wide range of habitats with various environmental factors characteristic to every species.

A high resistance to significant changes in abiotic and biotic variables is the most common feature of all the aforementioned species. Therefore, the existence of various ostracod species living in the harsh conditions of the three studied springs is to be expected.

C. neglecta, H. incongruens, H. salina, H. chevreuxi, P. olivaceus, and P. zenkeri were determined in the sandy-gravel substrate of Pınarbaşı Spring. C. neglecta, H. incongruens, H. salina, H. chevreuxi, P. olivaceus, P. zenkeri, I. gibba, and P. marchica in the sandy-mud substrate of Dipsiz Spring. C. neglecta, H. chevreuxi, P. olivaceus, and P. marchica were determined in the sandy-gravel substrate of Deştin Spring. Springs are known to be poor habitats with respect to species diversity. Ostracod species richness in limnocrenic springs is significantly greater than in rheocrenes or helocrenes [49]. Deştin (four species) and Pınarbaşı (five species) rheocrenic springs had poor species diversity compared with Dipsiz
Spring, a limnocrene spring (eight species). High water flowrate, the existence of nutrient-poor sandy-gravel substrates, and macrophyte species scarcity would seem to be reasons for low species diversity in the Deştin and Pınarbaşı rheocrene springs. In many other studies performed in Turkish wetlands [e.g. 11, 20, 27, 47, 48], higher number of individuals have been reported for all ostracod species than were determined in the present study, which may be because the ostracods living in the three springs exist in nutrient-poor habitats, sandy-gravel substrates, and high water flowrate habitats.

C. neglecta, H. incongruens, H. salina, and C. chevreuxi are eurytopic taxa. Their occurrence has been reported in nutrient-rich, eutrophic, and polluted waters in Turkey [26, 27]. Four cosmopolitan species (H. incongruens, I. gibba, H. salina, C. neglecta) displayed a high environmental tolerance to at least six different environmental variables. Results suggest that cosmopolitan species have the advantage of a strong ability to disperse in a variety of aquatic habitats within the study area. The presence of cosmopolitan and tolerant ostracod species in the three springs shows the strong dispersibility of ostracods. The same observations have repeatedly been recorded in many studies. C. neglecta, H. incongruens, H. salina, P. marchica, I. gibba, and P. zenkeri were reported in springs feeding Lake Acıgöl [29].

As a conclusion, we can observe that bottom-dependent, cosmopolitan species and almost cosmopolitan species have higher tolerance to different levels of environmental variables. The ostracod species that were observed in three springs, with their almost stable physicochemical parameters, are often able to use their limbs to hold on strongly to the substrate against the water flow, whether they are non-swimmers or good swimmers. The results of the present study show that the faunal composition of ostracods was related to water flow and benthic substrate characteristics. One of the most important conclusions in this study is that the faunal composition of ostracods varies in different types of springs depending on factors such as substrata type, the presence of different microhabitats, water discharge, and flowrate.

The loss of Mediterranean springs as a result of human activity will result in the loss of biodiversity within Turkey. We have seen during in other many studies conducted by us that many natural water springs have been destroyed in rural/pastoral areas of Turkey. There are meaningful relationships between the protection of natural springs and socio-economic and cultural factors. Conservation and restoration projects should be created and prioritized for the freshwater springs of Pınarbaşı, Dipsiz, and Deştin in order to reduce the anthropogenic impact on these springs.

5. Acknowledgements
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6. References
33. Bronshtein ZS. Fresh-water Ostracoda, Fauna of the USSR Crustacean, Russian Translation Series 64, Oconian Press, New Delhi, 1947, 2(1).