

Diversity analyses of nonmarine ostracods (Crustacea, Ostracoda) in streams and lakes in Turkey

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Abstract: In order to compare species compositions of ostracods, 25 streams and 15 lakes were sampled in the spring, summer, and autumn seasons of 2018. A total of 26 ostracod species were found in lakes (18 spp.) and streams (12 spp.). The Shannon index (H') and evenness values of streams were higher than in lakes in all seasons. The highest H' values for all combined (lakes + streams) and lake data were reported in the autumn season, and in spring the highest values were in streams. According to the β -diversity (β) index values, the variability of ostracod species composition in lakes was higher than in streams, and its value was highest in spring (0.40) and lowest in summer (0.34) among all seasons for combined data. Pairwise comparison of spring and autumn displayed higher β -diversity values than other comparisons, while its value was 0.41 between lakes and streams. According to canonical correspondence analysis results, elevation had a significant ($P = 0.006$) effect on distribution of species. All results suggested the importance of seasonality for evaluating the biodiversity of a region rather than the number of sampling sites, and the autumn season seems to be richer than other seasons in terms of species diversity.

Keywords: Beta diversity, elevation, seasonality, ostracods, composition, distribution

1. Introduction

Species diversity is defined as the number of both rare and common species in a community, and that includes species richness (number of species) and evenness (equitability of individuals among species) (Hamilton, 2005). Although species diversity displays fluctuation over a spatial scale, it is important for the conservation of biodiversity (Engen et al., 2008). For this reason measuring diversity, and determining factors affecting it, plays a crucial role in developing biodiversity protection policy. To describe the richness and evenness of species in a community, statistical approaches (e.g., diversity indices) are commonly used (Magurran, 1988). Levels of diversity or total diversity in an area [γ] are characterized by alpha (α) and beta (β) diversity indices (Whittaker, 1960). Local average species richness is termed as α -diversity (Magurran, 2004), while β -diversity can be estimated as the dissimilarities of species composition among sites (e.g., habitats) (Anderson et al., 2006). Environmental differences and geographical distance between sites were commonly used for the calculation of β -diversity index (Clarke et al., 2008). Accordingly, β -diversity is frequently used to compare the heterogeneity of habitats. The effect of habitat heterogeneity on species diversity and richness is a known fact among

ecologists (MacArthur and MacArthur, 1961). While a biotic variability (e.g., pH, temperature, etc.) in a habitat is termed habitat heterogeneity (Hortal et al., 2009), species diversity indices (α and β) allow us to specify the variables controlling and influencing distribution and composition of species.

The class Ostracoda is a commonly found crustacean group in all types of aquatic bodies because of their ecological plasticity (Rodriguez-Lazaro and Ruiz-Muñoz, 2012). Physico-chemical variables of aquatic bodies affected by climatic and regional factors have important roles in shaping ostracod species composition in any type of habitat (Horne, 2007). Among individual species, the level of tolerance of these variables determines their existence, and individual species of ostracods exhibit species-specific responses to the ecological variability in aquatic bodies (Külköylüoğlu, 2013). Hereby, they are commonly used to evaluate the quality of habitats both past and present. The determination of effective regional and local factors where ostracod species are found provides noteworthy information to paleontologists who use them as proxies for estimating the paleoenvironmental evolution of water bodies (e.g., lakes and streams) (Carbonel et al., 1988). This is because ostracods grow by moulting up to 9 stages to

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reach adulthood, and the chemical contents of valves are taken from ambient water during each moulting stage (De Deckker et al., 1988). Geochemistry and ornamentation of the carapaces of ostracods light the way for assessing variability in the physico-chemical properties of the water bodies where they formed.

Species diversity in a region or habitat is affected by physico-chemical changes due to seasonal changes and the precipitation regime. For that reason, seasonal sampling reveals the species diversity of region/s more clearly and allows us to observe the change in species composition between seasons. This work aims to compare ostracod species composition between streams and lakes sampled in three seasons (spring, summer, and autumn) and to evaluate the influence of abiotic variables on the distribution of ostracod species.

2. Materials and methods

2.1. Study area

The study area is located on the border of nine provinces in Turkey (39°–41°N, 34°–40°E) (Figure 1). Nine types of Köppen–Geiger climate classification systems are observed on the border of the sampled provinces: (1) cold semiarid (Yozgat); (2) Mediterranean-influenced hot summer, humid continental (Sivas); (3) subarctic (Rize), (4) Mediterranean-influenced warm summer, humid continental (all provinces except Rize); (5) warm summer Mediterranean (all provinces except Rize and

Gümüşhane); (6) warm summer humid continental (Samsun, Rize, Gümüşhane, and Çorum); (7) hot-summer Mediterranean (Samsun, Tokat, Çorum, and Yozgat); (8) humid-subtropical (Samsun, Ordu, Giresun, Rize, and Çorum); and (9) temperate oceanic climates (all provinces except Sivas, Tokat, and Yozgat) (Climate-Data.org, 2020). Lakes and streams under the influence of different climatic conditions and regional factors provide the opportunity to observe how Ostracoda diversity can change through different seasons. Thus, the area in Figure 1 was chosen for the present study. Nine of the streams (9S, 12S, 15S, 20S, 21S, 23S, 25S, 27S, and 40S) are second (2nd) order streams according to the Strahler stream ordering method (Strahler, 1957), while others are first (1st) order streams. The first order streams represent the smallest unbranched tributaries, while the merge of two 1st order streams forms a 2nd order stream.

2.2. Sampling

The study was performed in 15 natural lakes except 39L (dam) and 25 natural streams (Figure 1). Sampling was carried out in the spring (May–June), summer (July), and autumn (September) of 2018 (Appendix). Descending water levels in the littoral region of lakes sampled over three seasons were observed in summer and autumn compared to spring (Appendix).

Some abiotic variables [e.g., dissolved oxygen concentration (DO, mg/L)], water temperature (T_w , °C), electrical conductivity (EC, $\mu\text{S}/\text{cm}$), pH, and salinity (‰)

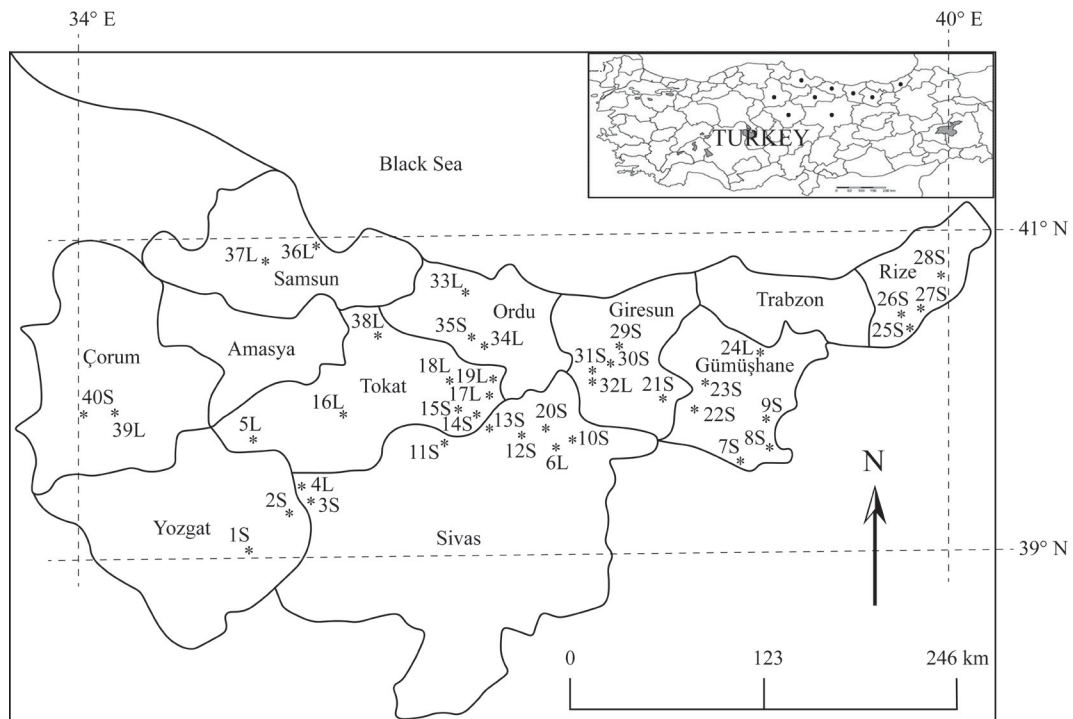


Figure 1. Location of sampling sites. The letters L and S indicate lake and stream, respectively.

were measured using the YSI Professional Plus multimeter before sampling. A GARMIN Etrex Vista H global positioning system (Garmin Ltd., Kansas, USA) was used to record the geographical data (elevation and coordinates; Appendix).

A standard nonmarine ostracod collection method was used to obtain samples from the littoral regions of lakes and the riparian zones of streams up to 1 m in depth using a standard-sized hand net (200 µm mesh). The samples were taken directly from the body of water and ca. 2–4 cm of sediment surface. Sediment samples including ostracods were stabilized with 70% ethanol in 250 ML plastic bottles in situ.

2.3. Laboratory analyses

Samples were washed under pressurized tap water and filtered through four standard-sized sieves (0.5, 1.0, 1.5, and 2.0 mm mesh) to eliminate coarse material. Ostracod specimens were sorted from the sediment under a stereo microscope (Olympus ACH 1X), and adult individuals with complete soft body parts were dissected in lacto-phenol solution to prepare a permanent slide for taxonomic identification. Following the taxonomic keys of Meisch (2000), species identification was done under a light microscope (Olympus BX-51). For the identification of species the morphology of carapaces and valves was also used.

2.4. Statistical analyses

Homogeneity of variance and normality of data were tested by Levene's test and Kolmogorov–Smirnov ($n > 50$) or Shapiro–Wilk ($n < 50$) tests, respectively. After assumptions were provided, parametric one-way ANOVA and independent-samples t-tests were used to analyse whether significant differences were present in the mean values of environmental variables among seasons (spring, summer, and autumn) and between habitats (streams and lakes), respectively (IBM-SPSS Statistics version 21). When the assumptions were not met, nonparametric Kruskal–Wallis and Mann–Whitney U-tests were performed along with the application of post-hoc analysis to determine likely significance. Possible correlations among environmental variables were tested by Pearson correlation analysis (IBM-SPSS Statistics version 21). To quantitatively measure species diversity the Shannon index (H'), evenness (Pielou's evenness), and Harrison beta diversity (β) index values for each habitat and the combined data of streams and lakes (all data), lakes, and streams in different seasons were calculated using PAST 3.26 software (Hammer et al., 2001). Beta diversity index values were also estimated for pairwise comparison of data to observe the dissimilarity of species composition. Shannon index and evenness values between habitat types and among seasons were performed using 9999 random permutation matrices (PAST 3.26). The gradient length

of the first two axes of detrended correspondence analysis (4.76) indicated the suitability of data for the application of unimodal canonical correspondence analyses (CCA) to explore the relationships between abiotic variables and ostracod species in the present study. The significance of variables on the distribution of ostracod species was tested by the Monte Carlo permutation test (999 permutations). CANOCO 4.5 software (ter Braak, 1986) was used to perform CCA, and species observed at least three times were used in CCA. In all analyses, adult individuals with undamaged carapaces and complete soft body parts were used.

3. Results

Minimum, maximum, mean, and standard deviation values of environmental variables for the combined data of both streams and lakes (all data), lakes, and streams indifferent seasons are given in Table 1. The mean values of all abiotic variables except dissolved oxygen showed significant differences ($P < 0.05$) between streams and lakes. Meaningful dissimilarities in the mean values of water temperature between spring and summer, pH between spring and autumn, and dissolved oxygen between summer and autumn ($P < 0.05$) were observed. Significant correlation percentages among abiotic variables at a 0.01 cut off level were presented in Figure 2.

A total of 26 ostracod species were recorded in all seasons combined (Table 2). Of these, 14 species belonging to eight genera (*Cypris*, *Eucypris*, *Herpetocypris*, *Psychrodromus*, *Stenocypris*, *Heterocypris*, *Cypridopsis*, and *Potamocypris*) are members of family Cyprididae, while families Candonidae (*Neglecandona angulata*, *Neglecandona neglecta*, *Paracandona euplectella*, *Pseudocandona albicans*, and *Physocypris kraepelini*) and Ilyocyprididae (*Ilyocypris* spp.) are represented by an equal number of species (5 spp.). The remaining two species belong to Limnocytheridae (*Limnocythere inopinata*) and Cytheridaidea (*Cyprideis torosa*) families. The abundance of species in habitats and among seasons, and the sites where they were found, are given in Table 2.

In all seasons combined, the number of species in lakes (18) was higher than in streams (12). The number of species in lakes and streams in spring (12 spp.) and autumn (8 spp.) were equal, while 9 and 5 species were recorded in streams and lakes, respectively, in summer. All data (streams + lakes) in the spring had the highest number of species (22 spp.) and individuals (274 ind.), when compared with summer (13 spp., 101 ind.) and autumn (15 spp., 112 ind.). Stream samplings in summer and autumn bore a higher number of individuals than lake samplings, except in the spring season (Table 2 and Figure 3). The Shannon index values of lakes in all seasons and separate seasons were lower than streams. In all

Table 1. Minimum (min), maximum (max), mean and standard deviations (std) of abiotic variables of combined data of both lakes and streams (all data), lakes' and streams' data in all three seasons (spring, summer and autumn).

Variables		All data	Lakes' data	Streams' data	Spring all data	Summer all data	Autumn all data	Spring lakes' data	Spring streams' data	Summer lakes' data	Summer streams' data	Autumn lakes' data	Autumn streams' data
Elevation	Min	0	0	980	0	70	70	0	998	70	980	70	980
	Max	2757	2757	2083	2757	2748	2748	2757	2083	2748	2042	2748	2048
	Mean	1455.9	1256.2	1563.42	1413	1480.2	1489.1	1173.7	1556.6	1318.3	1557.3	1317.9	1579.2
	Std	556.8	753.7	379.91	571.1	547.7	562.3	758.3	370.6	784.5	392.0	784.9	398.6
Water temperature	Min	5.1	5.1	7.1	5.1	10.4	12.2	5.1	7.1	11.1	10.4	13.2	12.2
	Max	27.4	27.4	27.2	24.7	27.4	26.7	24.7	19.9	27.4	27.2	26.7	21.1
	Mean	16.9	19.8	15.2	15.2	18.9	17.1	18.4	13.3	21.9	17.3	19.9	15.6
	Std	5.0	5.4	4.0	5.4	5.1	3.7	6.5	3.5	4.8	4.7	3.8	2.7
pH	Min	6.10	6.16	6.10	6.56	7.24	6.10	7.01	6.56	8.12	7.24	6.16	6.10
	Max	9.48	9.48	8.92	9.48	8.92	9.12	9.48	8.54	8.87	8.92	9.12	8.06
	Mean	7.95	8.22	7.80	7.99	8.28	7.54	8.17	7.88	8.41	8.22	8.12	7.24
	Std	0.69	0.65	0.68	0.63	0.43	0.81	0.69	0.58	0.24	0.49	0.86	0.61
Electrical conductivity	Min	17.81	55.5	17.81	17.81	26.6	39.2	55.5	17.81	58.8	26.6	59.1	39.2
	Max	3900	3900	646	3900	685	702	3900	646	685	637	702	440
	Mean	294.9	473.4	198.9	362.1	250.7	249.6	640.8	194.9	346.2	205.2	349.3	197.1
	Std	420.9	646.5	159.2	621.6	197.8	189.3	946.8	164.9	223.6	171.6	229.3	144.9
Dissolved oxygen	Min	2.87	2.87	6.66	2.87	6.66	6.66	2.87	7.45	7.10	6.66	6.66	7.62
	Max	10.84	10.84	9.57	10.81	8.97	10.84	10.81	9.53	8.90	8.97	10.84	9.57
	Mean	8.31	8.28	8.32	8.39	7.88	8.65	8.18	8.51	7.96	7.85	8.74	8.60
	Std	1.08	1.60	0.66	1.39	0.65	0.81	2.18	0.58	0.65	0.67	1.25	0.48
Salinity	Min	0.01	0.02	0.01	0.01	0.01	0.02	0.02	0.01	0.03	0.01	0.03	0.02
	Max	2.06	2.06	0.32	2.06	0.33	0.34	2.06	0.32	0.33	0.22	0.34	0.21
	Mean	0.14	0.23	0.09	0.18	0.11	0.12	0.32	0.09	0.16	0.09	0.17	0.10
	Std	0.22	0.34	0.07	0.33	0.09	0.09	0.50	0.08	0.10	0.07	0.11	0.07

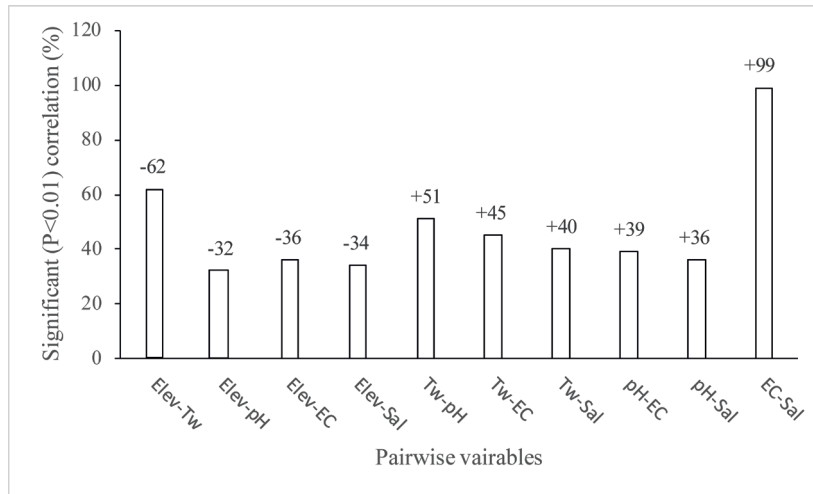


Figure 2. Pearson correlation results among elevation (Elev), water temperature (Tw), electrical conductivity (EC), salinity (Sal) and pH. The numbers above bars show the correlation coefficients between environmental variables.

seasons with combined data, the maximum value of the Shannon index was found in autumn ($H' = 2.203$), while index values were relatively similar in spring ($H' = 2.009$) and summer ($H' = 2.017$) seasons. The evenness value of all data in spring (0.6499) was smaller than in summer (0.7863) in contrast to the Shannon results, whereas almost all other evenness results indicated compatibility with the Shannon index findings (Figure 3). Significant differences ($P < 0.05$) for the Shannon index and evenness values between binary groups (among habitats and data in different seasons) were shown in Figure 3. For instance, the Shannon index and evenness values between streams and lakes were significantly distinct from each other.

Results of the β -diversity index evidenced higher variation in ostracod composition in lakes, compared to streams. The β -diversity index values were ordered from high to low as follows: spring (0.40) > autumn (0.38) > summer (0.34) for combined data in all seasons (Figure 4a). Binary β -diversity index values between all data and the same habitat type in different seasons displayed high species composition dissimilarities between spring and autumn, and a high β -diversity index (0.41) value was reported between streams and lakes in the autumn season (Figure 4b).

The first two axes of CCA elucidated 78.90% of relationships between species and environmental variables with a 7.4 cumulative percentage variance of species data. Among the five environmental variables used, elevation [Elev, λ (variance each variable explains) = 0.31, $P = 0.006$, $F = 2.37$] played an important role in the distribution of ostracod species in the present study, while water temperature (Tw, $\lambda = 0.18$, $P = 0.169$, $F = 1.42$), electrical

conductivity (EC, $\lambda = 0.15$, $P = 0.292$, $F = 1.18$), dissolved oxygen (DO, $\lambda = 0.15$, $P = 0.317$, $F = 1.11$), and pH ($\lambda = 0.02$, $P = 0.932$, $F = 0.17$) were subsidiary factors affecting the distribution of species. Ordination of ostracod species according to the effects of environmental variables on the CCA diagram was given in Figure 5. For example, *Eucypris virens* and *Limnocythere inopinata* demonstrated close relationships with elevation and electrical conductivity, respectively. *Ilyocypris inermis* was at the center of the diagram, while *Psychrodromus olivaceus* and *Potamocypis fallax* showed close associations with pH (Figure 5).

4. Discussion

4.1. Gamma diversity and seasonality

The ratio (0.65) of gamma diversity of ostracods ($\gamma = 26$ spp.) per sampled sites ($st = 40$) in all seasons combined herein was relatively higher than ratios in the following studies with wide sampling over a short period of time in Turkey; 0.20 in Ordu Province ($st = 133$) (Külköylüoğlu et al., 2012a), 0.51 in Van Province ($st = 57$) (Külköylüoğlu et al., 2012b), and 0.30 in Artvin ($st = 62$) (Külköylüoğlu et al., 2020). In addition, Van der Meeren et al. (2010) reported a low γ/st ratio (0.44) from 106 sampling sites in the summer seasons of 2004 and 2005 in Western Mongolia. Iglukowska and Namiotko (2012) recorded a γ/st ratio equal to 0.83, because sampling including different months/seasons from 2006 to 2008 in freshwater habitats of subarctic and temperate Europe. As seen in the abovementioned examples, studies including more than one sampling time or those that were done in different seasons resulted in higher gamma diversity than studies done in one season regardless of the number of sampling sites. However, this

Table 2. Species list and the abundance of species in different seasons (spring, summer and autumn) and habitats (streams and lakes), and the minimum (min) and maximum (max) values of environmental variables (Elev: elevation, Tw: water temperature, EC: electrical conductivity, DO: dissolved oxygen, and Sal: salinity) of sites where species were found. Abbreviations: *Na* (*Neglecandona angulata*), *Nn* (*Neglecandona neglecta*), *Ct* (*Cyprideis torosa*), *Cv* (*Cypridopsis vidua*), *Cp* (*Cypris pubera*), *Ev* (*Eucypris virens*), *Hint* (*Herpetocypris intermedia*), *Hi* (*Heterocypris incongruens*), *Hs* (*Heterocypris salina*), *Ib* (*Ilyocypris bradyi*), *Ige* (*Ilyocypris getica*), *Igi* (*Ilyocypris gibba*), *Ii* (*Ilyocypris inermis*), *Im* (*Ilyocypris monstrifica*), *Li* (*Limnocythere inopinata*), *Pe* (*Paracandona euplectella*), *Pa* (*Potamocypris arcuata*), *Pf* (*Potamocypris fallax*), *Pfu* (*Potamocypris fulva*), *Pp* (*Potamocypris pallida*), *Ps* (*Potamocypris similis*), *Pv* (*Potamocypris villosa*), *Psa* (*Pseudocandona albicans*), *Po* (*Psychrodromus olivaceus*), *Pk* (*Pyhsocypris kraepelini*), *Sf* (*Stenocypris fischeri*), *S* (stream), *L* (lake), *Spal* (combined data of both streams and lakes (all data) in spring), *Sumal* (all data in summer), *Autal* (all data in autumn), *SpS* (streams' data in spring), *SpL* (lakes' data in spring), *SumS* (streams' data in summer), *SumL* (lakes' data in summer), *AutS* (streams' data in autumn), *AutL* (lakes' data in autumn) and *Nst* (number of sampled sites).

Code	Habitats and Seasons											Elev (m)		Tw (°C)		pH		EC (µS/cm)		DO (mg/L)		Sal (‰)		Sampling sites
	S	L	Spal	Sumal	Autal	SpS	SpL	SumS	SumL	AutS	AutL	min	max	min	max	min	max	min	max	min	max	min	max	
Na	3		2	1		2		1				1270	2059	9.8	14.7	6.98	8.44	26.3	284	7.42	8.75	0.01	0.13	14S, 27S
Nn	56	5	24	14	23	24		14		18	5	998	2748	10.4	24	6.16	8.92	26.6	646	6.73	9.53	0.01	0.32	1S, 3S, 7S, 9S, 11S, 13S, 15S, 20S, 22S, 23S, 24L, 25S, 27S, 31S, 32L, 35S, 40S
Ct		3	3				3					0	0	23.6	23.6	9.48	9.48	1123	1123	9.74	9.74	0.56	0.56	36L
Cv		21	13	3	5		13		3		5	875	1800	15.1	27.4	7.14	9.12	185	702	2.87	10.7	0.09	0.34	4L, 6L, 19L, 37L, 38L
Cp		120	120				120					1800	1800	15.1	15.1	7.14	7.14	209	209	2.87	2.87	0.1	0.1	6L
Ev	1	4	2		3	1	1				3	1179	2748	13.2	15.1	7.14	8.91	59.1	329	2.87	8.54	0.03	0.15	6L, 11S, 32L
Hint		1			1						1	1210	1210	20.3	20.3	7.80	7.80	182	182	8.94	8.94	0.09	0.09	34L
Hi	3		1	1	1	1		1		1		1015	1089	16.5	25.6	7.94	8.55	320	637	6.85	8.72	0.03	0.15	15S, 40S
Hs		1	1				1					1043	1043	23.8	23.8	9.04	9.04	3900	3900	9.11	9.11	2.06	2.06	5L
Ib	26		7	13	6	7		13		6		1351	1625	10.1	27.2	7.2	8.44	277	457	6.66	9.18	0.13	0.22	1S, 3S, 9S, 13S
Ige		2	2				2					1043	1043	23.8	23.8	9.04	9.04	3900	3900	9.11	9.11	2.06	2.06	5L
Igi		4	3	1			3		1			1373	1373	24.7	24.7	8.62	8.62	696	696	8.30	8.30	0.34	0.34	4L
Ii	40		26	6	8	26		6		8		980	1980	10.7	21.1	6.77	8.57	40.5	401	7.28	9.57	0.02	0.19	7S, 8S, 10S, 11S, 14S, 20S, 26S, 35S
Im		3	3				3					541	541	23.9	23.9	7.75	7.75	652	652	4.94	4.94	0.32	0.32	16L
Li		10	1	5	4		1		5		4	900	1210	19.8	24.1	8.12	9.04	182.5	3900	7.33	9.11	0.09	2.06	5L, 34L, 39L
Pe		1	1				1					0	0	23.6	23.6	9.48	9.48	1123	1123	9.74	9.74	0.56	0.56	36L
Pa		1			1						1	900	900	19.8	19.8	8.40	8.40	602	602	8.09	8.09	0.29	0.29	39L
Pf	84	1	36	22	27	36		21	1	27		998	2059	10.7	27.2	6.3	8.77	67.5	457	6.66	9.57	0.03	0.22	1S, 2S, 7S, 8S, 11S, 13S, 14S, 15S, 21S, 27S, 35S, 38S
Pfu	1		1			1						1401	1401	15.9	15.9	8.42	8.42	342	342	8.98	8.98	0.16	0.16	12S
Pp	25		8	9	8	8		9		8		1980	1994	8.5	14.3	6.77	7.24	33.5	99.5	7.94	8.65	0.01	0.05	26S
Ps	5		2	1	2	2		1		2		1089	1843	9.7	17.3	7.03	8.02	39.5	440	8.34	8.98	0.02	0.21	29S, 31S, 40S
Pv		13			13						13	2544	2544	14.4	14.4	6.16	6.16	70.2	70.2	8.14	8.14	0.03	0.03	24L
Psa	2	1	3			2	1					1046	1808	12.6	20.9	8.21	8.37	153	594	6.51	8.11	0.07	0.29	10S, 17L
Po	39		7	24	8	7		24		8		1080	1980	12.2	27.2	6.56	8.57	38	457	6.66	9.21	0.02	0.22	1S, 7S, 11S, 13S, 14S, 20S, 23S, 26S, 28S, 30S
Pk		3		1	2				1		2	70	1168	20.7	24.1	8.14	8.87	188.1	339	8.9	10.84	0.09	0.18	18L, 33L, 38L
Sf		8	8				8					541	1800	15.1	23.9	7.14	7.75	209	652	2.87	4.94	0.1	0.32	6L, 16L
Nst	25	15	29	19	23	21	8	14	5	16	7													

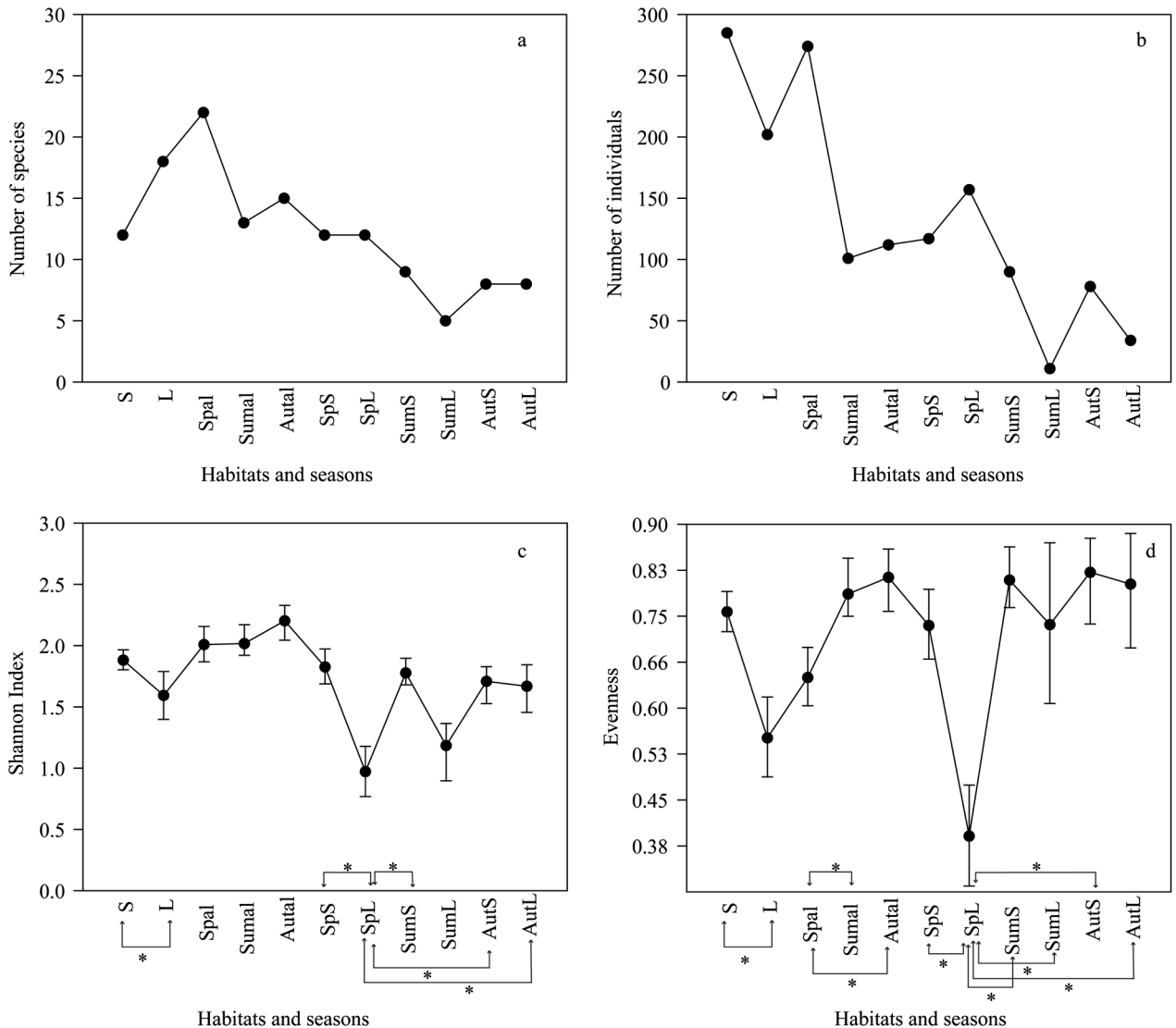


Figure 3. Number of species (a) and individuals (b), and values and standard error of Shannon index (c) and evenness (d) values in streams (S), lakes (L), all data in spring (Spal), summer (Sumal) and autumn (Autal), and streams' data (SpS) and lakes' data (SpL) in spring, and streams' data (SumS) and lakes' data (SumL) in summer, and streams' data (AutS) and lakes' data (AutL) in autumn. * indicates significant difference at 0.05 cut off level.

does not mean that the number of sampling sites is not very important. A recent study carried out in a tropical flood plain also showed the effect of seasonality and number of sites on the diversity of ostracods (Pereira et al., 2017). The authors noted the presence of 17 and 23 species, respectively, in the 6 lakes sampled in both dry and rainy seasons, while they collected 29 species in 17 other lakes sampled only in the rainy season. They explained the presence of 6 additional species in the 17 lakes (compared to 6 lakes sampled in both dry and rainy seasons) by the high number of sampling sites. Klkylođlu et al. (2016); however, proposed that increasing the number of sampling

sites is not a prerequisite for increasing the number of species. This is the case in the present study since a clear relationship was not observed between sampling sites and number of species in the spring (29 sites, 22 spp.), summer (19 sites, 15 spp.), and autumn (23 sites, 13 spp.) (Table 2). Therefore, further studies are needed to explore this topic. Unlike the number of sampling sites, the clear effect of seasonality on the number of species is apparent.

4.2. Species diversity and environmental variables

An evaluation based on the index value is more reliable than species richness since some species are found as sporadic or dominant species. Five species in spring and

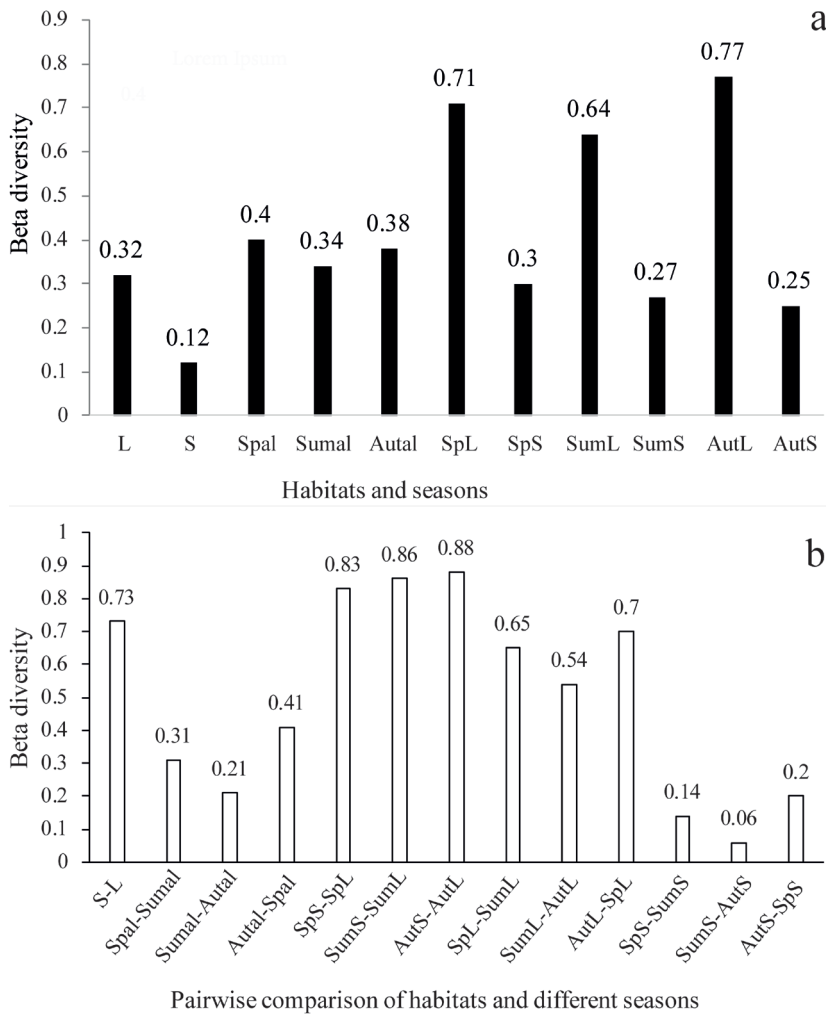


Figure 4. Individual (a) and pairwise (b) Beta diversity index values of lakes, streams, all data and individual habitat types in different seasons. Abbreviations of habitats and seasons are same with Figure 3.

summer and 3 species in autumn can be characterized as sporadic, since they were represented only by 1 individual in the present study (Table 2). In all data, it seems that the lowest mean pH value (7.54) and the highest dissolved oxygen value (8.65 mg/L) drove the highest diversity in autumn (see Table 1). Although both variables were not effective variables in the present study, after the application of CCA, their importance for ostracod communities was shown in studies conducted both inside and outside Turkey (e.g., Iglukowska and Namiotko, 2012; Uçak et al., 2014). The significant differences in mean values of pH between spring and autumn and dissolved oxygen between summer and autumn also indicate the importance of both variables in the high diversity value in autumn. In addition, the Shannon index values of spring ($H' = 2.09$) and summer ($H' = 2.017$) were very close to

each other, and similarly, with the exception of water temperature, their mean environmental variable values did not display significant distinctness. Despite the prevalence of water temperature as an important variable driving the distribution of ostracods in the literature (Yavuzatmaca, 2019), it had no important effect on the diversity index values between spring and summer in the present study.

It seems that the elimination of five lakes with a mean electrical conductivity value equal to 1238.4 $\mu\text{S}/\text{cm}$ (209–3900 $\mu\text{S}/\text{cm}$) had a positive effect on the Shannon index and evenness values of lakes from spring to autumn (Figure 3). Along with the decrease in electrical conductivity, the highest mean dissolved oxygen value was also an important variable in the occurrence of high evenness and Shannon index values through autumn in lakes (Table 1 and Figure 3). Unlike lakes, no significant

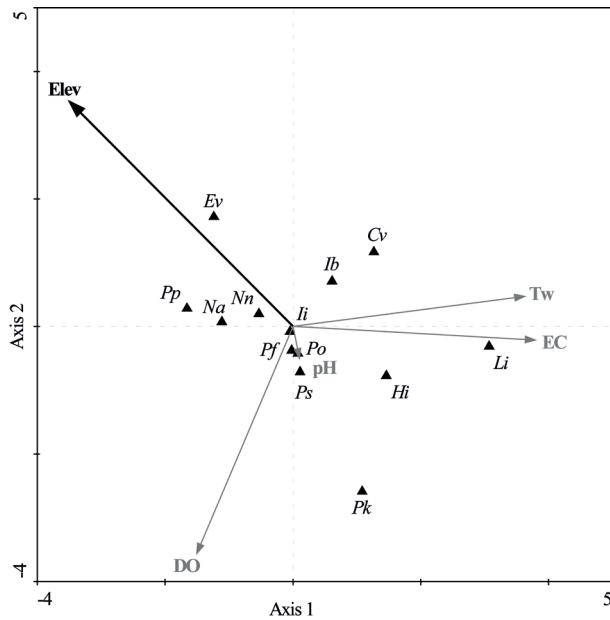


Figure 5. Ordination of 13 species occurred more than three times according to the effect of elevation (Elev), water temperature (Tw), electrical conductivity (EC), pH and dissolved oxygen (DO) on their distribution. Black arrow indicates variable with a significant effect when grey arrows show variables with insignificant effects. For species codes see Table 1.

differences were observed in diversity index values among seasons for streams (Figure 3).

The negative linear relationship between air temperature and elevation is a known situation (McCain and Grytnes, 2010), and elevational increases cause declining air temperature. Any changes in air temperature can cause fluctuations in water temperature, as water and air temperatures display a linear relationship (Schindler et al., 1990; Preud'homme and Stefan, 1992; Yavuzatmaca et al., 2018). In other words, elevation has an important effect on the critical local factor, temperature, which regulates not only species richness and abundance in aquatic and terrestrial environments but also the physico-chemical parameters of aquatic bodies (Rogora et al., 2008; Külköylüoğlu et al., 2012c; Yavuzatmaca, 2019). This is also the case in the present study, inasmuch as elevation indicated a strong negative correlation with water temperature ($P < 0.01$) when water temperature exhibited positively significant correlations with other variables (e.g., pH and electrical conductivity) (Figure 2). As in the current work, previous studies conducted along and near the border of the present study area [Çankırı (Külköylüoğlu et al., 2016), Kırşehir (Külköylüoğlu et al., 2019), Artvin (Külköylüoğlu et al., 2020), and Giresun (Çapraz et al., 2020 under review)] also declared the importance of elevation on the distribution of ostracods,

with the exception studies in Erzincan (Akdemir and Külköylüoğlu, 2014), Ordu (Külköylüoğlu et al., 2012a), Kahramanmaraş (Külköylüoğlu et al., 2012c), and Sinop (Yavuzatmaca et al., 2017). Elevation was also an important variable that had a role in the composition of ostracods in Friuli Venezia Giulia (Northeast Italy) (Pieri et al., 2009), Western Mongolia (Van der Meeren et al., 2010), and Kenya (Rumes et al., 2016). The abovementioned literature and the results of the present study revealed that elevation is a secondary factor that cannot be ignored when talking about the distribution and composition of ostracods. Therefore, species which have the ability to tolerate changes resulting from regional factors (elevation herein) in aquatic bodies may also commonly occur in different regions. For example, eurybiont species in the present study (e.g., *N. neglecta*, *I. inermis*, *P. olivaceus*, and *P. fallax*) closely ordinated through the center of the CCA diagram (Figure 5) which means that these species have wider tolerance of the variables used herein, and as a result, are commonly found (see Table 2). Such species may increase the alpha and gamma diversity values, while decreasing the β -diversity value.

4.3. Species diversity between streams and lakes

Although the number of sampled streams (25) is ca 1.67 times the number of lakes (15), a higher number of species was reported in lakes (18 spp.) than in streams (12 spp.) (Table 2). This result supports the suggestions of Vannote et al. (1980) and Minshall et al. (1985) who pinpointed the occurrence of high biodiversity at intermediate-sized streams. In other words, they emphasized an increase in species richness with growth in the size of streams. As 64% of streams (16) herein are 1st order streams this could elucidate low species numbers since stream size increases from headwater (or 1st order) to streams of other orders (2nd, 3rd, etc.) (Vander Vorste et al., 2017). The increase in diversity from headwater to wider or deeper channels was correlated with temperature, food availability, and flow regime. In the present study, the mean water temperature values of lakes (19.8 °C) and streams (15.2 °C) were significantly different ($P < 0.05$), but mean dissolved oxygen values were not (Table 1). Along with the system (headwater streams), water temperature can also be shown to cause low species numbers in streams when compared to lakes. The high species numbers in lakes might be due to the stable environmental conditions and the presence of vegetation at the littoral regions of lakes, as stated by Iglukowska and Namiotko (2012). The presence of specialized vegetation and different kinds of sediments in the littoral region provides a wide range of habitats (or niches) for species. This may be the primary reason for the high species numbers in lakes in the present study. On the other hand, the permanent water flow in the headwater streams acts as a homogenizing factor in the narrow

channel. This decreases niche opportunities (sediment or vegetation availability), and therefore, negatively affects species richness. Although the highest species richness occurs in lakes in the present study, the Shannon index value of streams ($H' = 1.881$) was higher than lakes ($H' = 1.594$) when abundance and species richness were combined. This is because the even distribution of individuals among species in streams (evenness = 0.7571) was higher than in lakes (evenness = 0.5514). This result may be related to the resilience of the streams surveyed in the present study which are 1st and 2nd order streams with permanent water flows. However, water level fluctuations were noticed in the littoral zones of lakes during the study. These kinds of fluctuations allow some species comprised of more individuals to become dominant while others, represented by a few individuals, become rare species (Table 2). Therefore, the disappearance probability of species in streams maybe much lower than in lakes.

4.4. Beta diversity

The low β -diversity value (0.12) of all streams data (Figure 4a) indicated the similarity of the streams herein. In addition, small fluctuations in β -diversity among seasons also hint at the stability of environmental conditions in streams across seasons (Figure 4a). The uniform environmental conditions of most headwater streams in the current study contribute to this low β -diversity value. In contrast to streams, the high β -diversity value (0.32) of lakes in the present study pinpointed their distinctness. The abovementioned heterogeneity in the littoral regions of lakes causes the differences among lakes, since each of these closed lakes have their own characteristic environmental conditions. The positive effect of environmental heterogeneity on β -diversity for macrobenthic assemblages was also noted by Anderson et al. (2006). Crist and Veech (2006) correlated high levels of β -diversity with habitat heterogeneity and dispersal limitation. Dispersal of ostracods over a long distance may be done passively by humans, birds, fishes, insects, wind (for resting eggs), etc. (McKenzie and Moroni, 1986; Green, 2016). Because ostracods do not actively distribute across long distances without these kinds of vectors, the unconnectedness of these aquatic bodies and the absence of vectors in the area of aquatic bodies resulted in a high β -diversity value. This is the case in most, but not all, the lakes in the present study since they are far from settlement areas and located at high altitudes. Among the seasons, β -diversity values of lakes exhibited a similar pattern to streams, although the values are higher than those of streams (Figure 4a). Four of the 26 species (*N. neglecta*, *E. virens*, *P. fallax*, and *P. olivaceus*) were common in both lakes and streams, and so a large β -diversity value (0.73)

was observed between them (Figure 4b). According to the pairwise comparison, high β -diversity values were seen between the spring and autumn for all, stream, and lake data. This is maybe a result of the seasonality of ostracods, because they can breed more than once per year and their life spans show variability across species (Delorme, 1991; Meisch, 2000). Therefore, the low β -diversity values between spring–summer and summer–autumn pairwise comparisons are understandable.

5. Conclusion

The importance of seasonality on ostracod species, rather than number of sampling sites, was emphasized in order to evaluate the biodiversity of a region. Instead of a single season or a large number of sampling sites taken in a short period of time, the sampling of an appropriate number of sites in different seasons can provide more accurate information about biodiversity. Considering all data (streams+lakes) and lakes, autumn is higher in terms of diversity. However, no significant difference was observed in terms of biodiversity index values among seasons for streams. The higher diversity index value of streams over lakes implies the resilience of streams when compared to lakes. In addition, the streams herein are very similar to each other as they are headwater streams with permanent water flows. Therefore, their β -diversity value was lower than that of lakes. It seems that α -diversity was largely affected by local factors (e.g., pH and electrical conductivity), while the β -diversity index was mostly impacted by the system (lotic and lentic habitats). The importance of the effects of regional factors, namely elevation, on ostracod species composition was underlined again. In all, the diversity of ostracods cannot be explained by a single parameter, because the composition and distribution of species are controlled by interconnected seasons, systems, and local and regional factors.

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Conflict of interest

The author declares no potential conflicts of interest with respect to research, authorship, and/or publication of this paper.

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Appendix. Site codes (Stcod), habitat types (Hty), province where sites located, sampling seasons (spring (May-June), summer (July) and autumn (September (Sep))), elevation (Elev, m a.s.l.) and coordinates of sampling sites in the present study. Due to transportation disruptions, 5 lakes and 3 rivers were sampled only in the spring season. NS means not sampled.

Stcod	Hty	Province	May-June	July	Sep	Elev	Coordinates
1S	Stream	Yozgat	+	+	+	1585	39°32'00.4"N, 35°53'06.8"E
2S	Stream	Yozgat	+	dry	dry	1320	39°42'39.5"N, 36°01'32.4"E
3S	Stream	Sivas	+	NS	NS	1351	39°44'38.7"N, 36°06'49.8"E
4L	Lake	Sivas	+	+	+	1373	39°47'05.6"N, 36°06'38.6"E
5L	Lake	Tokat	+	NS	NS	1043	39°59'46.3"N, 35°59'08.9"E
6L	Lake	Sivas	+	NS	NS	1800	40°00'16.5"N, 38°06'40.0"E
7S	Stream	Gümüşhane	+	+	+	1906	39°54'39.2"N, 39°32'47.6"E
8S	Stream	Gümüşhane	+	+	+	1701	39°58'26.2"N, 39°38'56.6"E
9S	Stream	Gümüşhane	+	+	dry	1625	40°09'52.8"N, 39°42'05.5"E
10S	Stream	Sivas	+	NS	NS	1808	40°00'23.4"N, 38°06'36.3"E
11S	Stream	Sivas	+	+	+	1168	40°13'56.6"N, 36°53'51.2"E
12S	Stream	Sivas	+	NS	NS	1401	40°08'05.7"N, 37°09'58.4"E
13S	Stream	Sivas	+	+	+	1573	40°12'44.6"N, 37°01'55.3"E
14S	Stream	Tokat	+	+	+	1270	40°15'05.2"N, 37°01'44.9"E
15S	Stream	Tokat	+	+	+	1015	40°17'13.6"N, 36°57'16.6"E
16L	Lake	Tokat	+	NS	NS	541	40°16'50.6"N, 36°09'17.5"E
17L	Lake	Tokat	+	+	+	1046	40°22'37.1"N, 37°27'54.8"E
18L	Lake	Tokat	+	+	+	952	40°26'45.4"N, 37°16'27.5"E
19L	Lake	Tokat	+	+	+	1195	40°27'09.9"N, 37°27'45.0"E
20S	Stream	Sivas	+	+	+	1787	40°06'33.3"N, 37°51'05.6"E
21S	Stream	Giresun	+	+	+	1885	40°21'37.0"N, 38°55'57.2"E
22S	Stream	Gümüşhane	+	+	+	2057	40°18'57.1"N, 39°03'10.3"E
23S	Stream	Gümüşhane	+	+	+	1988	40°26'19.2"N, 39°06'34.1"E
24L	Lake	Gümüşhane	+	+	+	2544	40°34'39.0"N, 39°41'26.4"E
25S	Stream	Rize	+	+	+	1982	40°36'26.9"N, 40°36'27.5"E
26S	Stream	Rize	+	+	+	1980	40°38'21.1"N, 40°38'52.2"E
27S	Stream	Rize	+	+	+	2059	40°43'55.2"N, 40°47'36.1"E
28S	Stream	Rize	+	+	+	1080	40°53'05.5"N, 40°55'45.4"E
29S	Stream	Giresun	+	+	+	1090	40°39'00.6"N, 38°28'43.2"E
30S	Stream	Giresun	+	+	+	1181	40°33'41.7"N, 38°18'33.6"E
31S	Stream	Giresun	+	+	+	1829	40°33'25.3"N, 38°12'45.6"E
32L	Lake	Giresun	+	+	+	2748	40°30'50.4"N, 38°11'30.4"E
33L	Lake	Ordu	+	+	+	70	40°58'24.9"N, 37°30'14.0"E
34L	Lake	Ordu	+	+	+	1210	40°37'42.5"N, 37°32'45.5"E
35S	Stream	Ordu	+	+	+	998	40°39'16.5"N, 37°26'40.2"E
36L	Lake	Samsun	+	NS	NS	0	41°16'54.6"N, 36°56'14.6"E
37L	Lake	Samsun	+	NS	NS	875	40°53'39.0"N, 36°02'10.0"E
38L	Lake	Tokat	+	+	+	1168	40°49'41.9"N, 36°36'05.2"E
39L	Dam	Çorum	+	+	+	900	40°23'42.8"N, 34°39'52.3"E
40S	Stream	Çorum	+	+	dry	1089	40°21'35.8"N, 34°34'14.1"E