



# Determination of environmental variables groups affecting the occurrence of non-marine ostracods (Crustacea) in the Eastern Mediterranean region of Turkey

Mehmet Yavuzatmaca<sup>1</sup>

Received: 27 February 2022 / Accepted: 16 August 2022 / Published online: 3 September 2022

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## Abstract

Abiotic factors play important roles on the habitat preferences and dispersal decisions of species. The objective of this study was to estimate the groups of abiotic variables best explaining the variation of ostracods species taken from 31 water bodies (27 streams and 4 lakes) sampled twice (October 2020 and April 2021) in the Eastern Mediterranean region of Turkey. In this study, a total of 34 ostracod taxa (24 recent and 10 sub-fossil) were reported, and *Ilyocypris bradyi* and *Prionocypris zenkeri* were the most common species occurred more than 10 times. Based upon to the variation partitioning analysis, dissolved oxygen+elevation+magnesium (DO+Elev+Mg<sup>2+</sup>) was the best model with 12.66% explanation power in the variations of ostracod species in the present study. The distance-based redundancy analysis elucidated 14.1% of the total variation in the species distribution matrix that was significantly affected by Mg<sup>2+</sup> and Elev. The effectiveness of variables on the occurrence of species was tested by Generalized Linear Models resulted in positive roles of Elev for *Psychrodromus olivaceus*, *Neglecondona neglecta* and *Pseudocandona albicans*, Ca<sup>2+</sup> for *Cyprideis torosa* and *P. albicans*, and DO for *P. olivaceus*, but negative roles of Tw for *N. neglecta* and *P. albicans*, Ca<sup>2+</sup> for *Herpetocypris helenae*, and DO for *N. neglecta*. A positively significant association was found between high Mg<sup>2+</sup> values and the abundance of *P. zenkeri*. Results suggest that effectiveness rates of environmental factors on the occurrence of species can change according to ecoregional differences when the variables are evaluated in the analyses together. Therefore, especially ecoregion-based ecological information of species should be determined for better inferences about the ecological preferences of species.

**Keywords** Ostracoda · Magnesium · Model · Variation partitioning · Effectiveness of variables

## Introduction

A high level of biodiversity, approximately 100,000 out of 1.8 million species, is found in freshwaters constituting about 0.8% of the Earth's surface area (Dudgeon et al. 2006). Freshwaters are one of the best ecosystems to study the metacommunity dynamics because they are the systems bearing high environmental heterogeneities in the sense of connectivity and spatial extension (Heino 2011). Through time and space, the structure of ecological communities is shaped by the association of biotic (e.g., competition,

predation) and abiotic (e.g., local environmental conditions) factors, historical and dispersal process (Leibold et al. 2004). Distribution of aquatic invertebrates is affected by environmental factors, and the determination of the responses of organisms to the environmental variables and climate is an important issue to estimate the ecological results of regional and global changes (Heino et al. 2009).

Of the aquatic invertebrates, ostracods are the small (0.3–5 mm long) bivalved crustaceans living in all aquatic bodies from fresh to marine waters, underground waters and even in semiterrestrial habitats (Moore 1961; Meisch 2000). A pair of chitinous valves consisting of low magnesium calcite documenting the host water's chemical and isotopic data or carapaces of ostracods allow the fossilization of them, and they have been recorded from Ordovician to recent (Holmes and Chivas 2002; Oakley et al. 2012; Siveter et al. 2014). They are commonly used to reconstruct past environmental

✉ Mehmet Yavuzatmaca  
yavuzatmaca46@gmail.com

<sup>1</sup> Department of Biology, Faculty of Arts and Science, Bolu Abant İzzet Baysal University, Gölköy, 14280 Bolu, Turkey

conditions (Griffiths and Holmes 2000). Their distribution and abundance are controlled by ecoregional biotic and abiotic factors, which is also the subject of this study. For example, water ion composition (Baltanás et al. 1990), nutrient levels (Danielopol et al. 1993), habitat type and nature of substrate type (Benzie 1989), elevation, water temperature, salinity, calcium, alkalinity, and nutrients (Van der Meeren et al. 2010), pH, lake area and magnesium (Viehberg 2006) and dissolved oxygen and moisture (Uçak et al. 2014) are some of the abiotic factors affecting the diversity of ostracod assemblages. The sensitivity of ostracods to a wide range of chemical compounds is much more than other invertebrates (e.g., Odonata) (Shuhaimi-Othman et al. 2011; Ruiz et al. 2013; César dos Santos Lima et al. 2019). They also show species-specific respond to organic pollutions and so they can be used as the indicator of habitat disturbance and water quality (Mezquita et al. 1999a).

Above mentioned abiotic factors affecting the distribution of ostracods allow one to consider that whether the ecoregional differences (Loucks 1962; Omernik 1987) change the effects of variables on the distribution of ostracods. Most recently, Çelekli et al. (2021) underlined the importance of ecoregion (differences in soil structure, land uses, climate, altitude, geology, and hydrology data) on the trophic weight and ecological preferences of benthic diatoms. This is also the case for ostracods. A large-scale survey research emphasized the importance of precipitation, temperature, and elevation on the distribution of ostracods in South America (de Oliveira da Conceição et al. 2019). A recent study conducted in another region pinpointed that the ostracods found in 77% of the 243 Patagonian freshwater bodies (in Argentina) with climatic heterogeneity are mainly controlled by the dissolved oxygen, water temperature, precipitation, and air temperature (Ramos et al. 2022). The authors also suggested that further studies need to be done in the different regions with respect to the spatial scale and environmental heterogeneity. In addition, Cusminsky et al. (2020) reported the associations of non-marine ostracods (28 taxa) collecting from 69 environments in Argentinian ecoregions with electrical conductivity, altitude, pH and water temperature. Therefore, the determination of the ecoregion-based abiotic factors affecting the species distribution will give more accurate result about the ecological preferences of species (e.g., ostracods).

Abiotic factors play important roles on the habitat preferences and dispersal decisions of species (Katz et al. 2017). Therefore, choosing of the correct abiotic variable/s to evaluate the distribution of species allow us to make strong and accurate inferences about the ecological preferences of species. In the present study, estimation of the efficient abiotic variable group/groups explaining the variation of ostracod species was aimed. To reach this aim, 31 water bodies (27 streams and 4 lakes) were sampled twice in the Eastern Mediterranean region of Turkey.

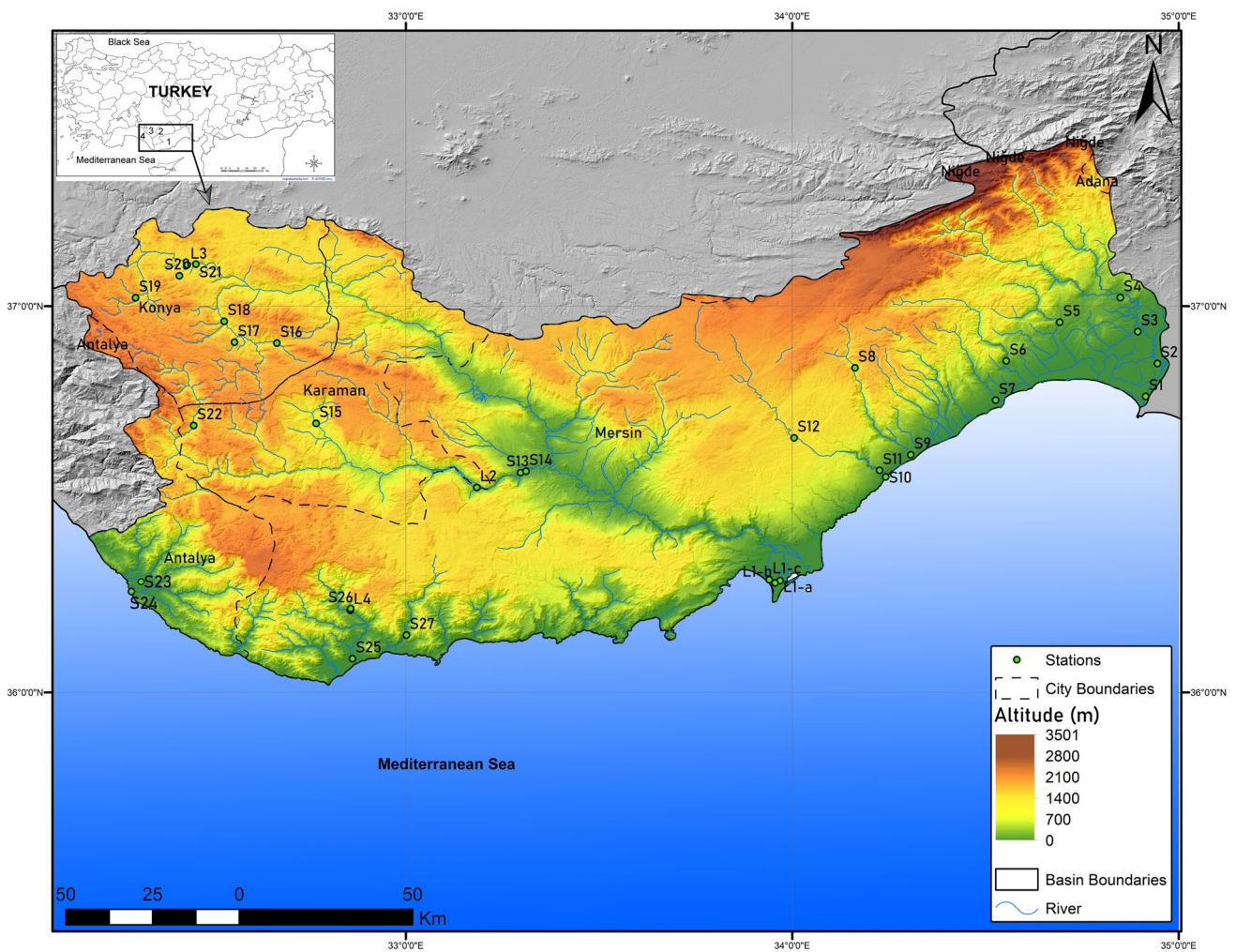
## Material methods

### Study area

The study was carried out within the borders of Antalya, Mersin, Karaman, and Konya provinces in the Eastern Mediterranean region of Turkey (does not cover the whole borders of these provinces). It is noteworthy to mention that this region is one of the hottest regions in Turkey (Fig. 1). In 2016, the monthly general temperature average of the region was reported as 17.78 °C, while the monthly lowest (−0.64 °C) and highest (30.57 °C) average temperatures were recorded in Konya (in December) and Mersin (in July), respectively. The annual average precipitation rate of the region is about 745 mm, and lower precipitation values are seen from west to east and inland. From the coastline to inland, terrestrialization is evident and the difference in annual average precipitation between the coastal and inland parts is about 500 mm (RTMFWM 2018). According to MGM (2022), the average data for the last 70–90 years show that the average temperature (Atemp) and precipitation (Apré) values of the provinces located on the coastline (Atemp = 19.2 °C and Apré = 615.5 mm for Mersin from 1940 to 2020, and Atemp = 18.8 °C and Apré = 1061.7 mm for Antalya from 1930 to 2020) were higher than the provinces located in the interior (Atemp = 11.7 °C and Apré = 329.2 mm for Konya from 1929 to 2020, and Atemp = 12 °C and Apré = 339.8 mm for Karaman from 1951 to 2020). The region generally has a karstic structure, and its geology consists of different aged limestone formations. Therefore, the soils are generally calcareous clay and loamy clay types with neutral or alkaline reactions, and the iron in the soil has caused the soil to turn red because of chemical reactions with the effect of climatic heat (RTMFWM 2018).

### Sampling and laboratory analyses

Samples were taken from 31 water bodies (4 lakes and 27 streams) twice, in October of 2020 and April of 2021 (Fig. 1 and Table 1). Before ostracod samplings, 100 ml of water samples were picked up from each sampling site and stored in a cooler container for the analyses of total phosphate (Tot-P), total nitrogen (Tot-N), magnesium ( $Mg^{2+}$ ), and calcium ( $Ca^{2+}$ ) according to APHA (1998). Total hardness (Tot-Hard) was calculated using the values of  $Ca^{2+}$  and  $Mg^{2+}$  for each site (Tot-Hard ( $mg\ L^{-1}\ CaCO_3$ ) = ( $Ca^{2+} \times 2.5$ ) + ( $Mg^{2+} \times 4.12$ )) (Boyd et al. 2016). Of the abiotic variables dissolved oxygen concentration (DO,  $mg\ L^{-1}$ ), water temperature (Tw, °C), electrical conductivity (EC,  $\mu S\ cm^{-1}$ ), pH, salinity (‰), and total



**Fig. 1** Locations of sampling sites in the study area. L and S represent lake and stream, respectively, while lowercase letters (a, b, and c) mean the multiple samplings in a site

dissolved solids (TDS,  $\text{mg L}^{-1}$ ) were measured by the aid of a YSI Professional Plus multimeter. Water samples taken from each site were read 10 times with a turbidimeter (WPA Turbidity Meter TU1100) to determine the average turbidity value of water. Geographical data (elevation and coordinates) was gained by a GARMIN Etrex Vista H global positioning system (Garmin Ltd., Kansas, USA).

Sediments including ostracod specimens were collected from the littoral regions of lakes (up to 1 m depth) and slow-flowing parts of the streams (up to 0.5 m in depth) with a hand net (200 mm mesh size). Subsequently, collected samples were fixed with 70% ethanol in 250 ml plastic bottles in situ. In the laboratory, all samples were washed under tap water through standard-sized sieves with 0.5, 1.0, 1.5 and 2.0 mm mesh sizes. Afterwards, ostracods were sorted from the sediment using fine needles under a stereo microscope (Olympus ACH 1X) and put into small glass vials with 70% ethanol for further research. Soft body parts of adult

specimens with complete carapaces were dissected in lactophenol solution for taxonomic description following Meisch (2000) and Karanovic (2012) under an Olympus BX-51 light microscope. Each sample was deposited in the Limnology Laboratory of Bolu Abant İzzet Baysal University, and can be available upon request.

**Statistical analyses**

Ecological Community Analysis II Software was used to test the multicollinearity among the environmental variables (Seaby and Henderson 2007). Accordingly, variables having an inflation factor larger than 10 indicating a possibility of multicollinearity were removed from the analyses. Of the abiotic variables, total phosphate and total nitrogen were not used in this analysis because their values were below detectable limits for many sites (Online Resource 1). Consequently, six abiotic variables (water temperature ( $T_w$ ),

**Table 1** Site codes (StC), habitat types, the province where sampling sites were found, sampling dates (October 2020 (Oc20)) and April 2021 (Ap21)), elevation (Elev, m asl.) and coordinates of sampling sites, and ostracod taxa encountered in each sampling site

StC	Habitat	Province	Oc20	Ap21	Elev	Coordinates	Taxa
L1-a	Lake	Mersin	+	+	1	36°17'22.88"N, 33°58'06.05"E	<b>Ct</b>
L1-b	Lake	Mersin	Ns	+	1	36°17'00.58"N, 33°57'18.87"E	<b>Ct</b> , Hts,
L1-c	Lake	Mersin	Ns	+	0	36°17'30.65"N, 33°56'25.78"E	Ct, Ls,
L2	Lake	Mersin	+	+	316	36°31'49.84"N, 33°11'01.28"E	Csp, Is, <b>Li</b>
L3	Lake	Konya	+	+	1152	37°06'24.27"N, 32°26'02.49"E	<b>Ib</b> , Ls, <b>Pz</b>
L4	Lake	Mersin	+	+	140	36°12'45.14"N, 32°51'22.32"E	Csp, <b>Hh</b> , Hsp, Is, <b>Sb</b>
S1	Stream	Mersin	+	+	3	36°45'58.64"N, 34°54' 52.51"E	Ct, <b>Psm</b>
S2	Stream	Mersin	+	+	6	36°51'05.40"N, 34°56'41.78"E	<b>Hi</b>
S3	Stream	Mersin	+	+	23	36°56'03.15"N, 34°53'38.49"E	<b>Hh</b> , Is, <b>Pz</b>
S4	Stream	Mersin	Dry	+	100	37°01'19.64"N, 34°50'56.07"E	Csp, <b>Ib</b>
S5	Stream	Mersin	+	+	131	36°57'29.65"N, 34°41'32.46"E	<b>Hs</b> , <b>Ib</b> , <b>Psm</b>
S6	Stream	Mersin	+	+	126	36°51'30.20"N, 34°33'13.67"E	<b>Hi</b> , <b>Ib</b> , <b>Pz</b>
S7	Stream	Mersin	+	+	28	36°45'23.95"N, 34°31'34.35"E	Ct, Cv, <b>Hin</b> , <b>Hi</b> , <b>Hs</b> , <b>Ib</b> , <b>Id</b> , <b>Pz</b> , <b>Ib</b>
S8	Stream	Mersin	+	+	1230	36°50'25.02"N, 34°9' 44.14"E	<b>Hi</b> , <b>Ib</b> , <b>Psp</b> , <b>Pz</b>
S9	Stream	Mersin	Dry	+	20	36°36'54.53"N, 34°18'24.34"E	<b>Pz</b>
S10	Stream	Mersin	+	+	8	36°33' 29.18"N, 34°14'30.27"E	Pa, <b>Po</b>
S11	Stream	Mersin	+	+	33	36°34'29.81"N, 34°13'33.12"E	<b>Hb</b> , <b>Ib</b> , <b>Pz</b> , Pssp
S12	Stream	Mersin	+	+	996	36°39'33.02"N, 34°00'17.12"E	<b>Ib</b> , <b>Pz</b> , Pssp
S13	Stream	Mersin	+	+	145	36°34'08.37"N, 33°17'47.82"E	Es, <b>Hr</b> , <b>Ib</b>
S14	Stream	Mersin	+	+	135	36°34'21.88"N, 33°18'38.75"E	<b>Hh</b> , Hts, <b>Ib</b> , Is, <b>Pz</b>
S15	Stream	Karaman	+	+	774	36°41'47.81"N, 32°46'03.37"E	<b>Hi</b> , <b>Hs</b> , <b>Ib</b> , <b>Nn</b> , <b>Pz</b> , <b>Pa</b> , <b>Po</b>
S16	Stream	Konya	Dry	+	1273	36°54'15.41"N, 32°39'58.37"E	Cs, <b>Po</b>
S17	Stream	Konya	+	+	1413	36°54'23.70"N, 32°33'23.00"E	<b>Ib</b> , <b>Nn</b> , <b>Pfu</b> , <b>Psp</b> , <b>Pz</b> , <b>Pa</b> , Pssp
S18	Stream	Konya	+	+	1046	36°57'38.30"N, 32°31'48.02"E	Is, <b>Li</b> , <b>Pv</b>
S19	Stream	Konya	+	+	1357	37°01'17.50"N, 32°18'01.95"E	Is, <b>Nn</b> , <b>Pf</b> , Pssp
S20	Stream	Konya	+	+	1353	37°04'42.52"N, 32°24'49.66"E	<b>Ep</b> , <b>Fb</b> , Is, <b>Nn</b> , <b>Pz</b> , <b>Pa</b> , <b>Po</b>
S21	Stream	Konya	+	+	1090	37°06'31.16"N, 32°27'23.71"E	<b>Ib</b> , <b>Pz</b>
S22	Stream	Karaman	+	+	1202	36°41'27.47"N, 32°27'03.00"E	<b>Po</b>
S23	Stream	Antalya	Dry	+	18	36°17'42.24"N, 32°18'24.02"E	<b>Hi</b> , Is
S24	Stream	Antalya	+	+	4	36°15'40.96"N, 32°17'22.73"E	Ct, Cv, <b>Hh</b> , <b>Hi</b> , <b>Hs</b>
S25	Stream	Mersin	+	+	6	36°05'15.56"N, 32°51'42.67"E	<b>Po</b>
S26	Stream	Mersin	+	+	226	36°13'01.48"N, 32°51'25.56"E	<b>Ii</b> , <b>Ps</b> , <b>Po</b> , Pssp
S27	Stream	Mersin	+	+	44	36°08'54.61"N, 33°00'05.16"E	Hts, <b>Ib</b> , Ss

Recent (living) taxa are in bold

Abbreviations: not sampled (Ns), *Candona* sp. (Cs), *Cyprideis torosa* (Ct), *Cypridopsis vidua* (Cv), *Cypridopsis* sp. (Csp), *Eucypris pigra* (Ep), *Eucypris* sp. (Es), *Fabaeformiscandona balatonica* (Fb), *Herpetocypris brevicaudata* (Hb), *H. helenae* (Hh), *H. intermedia* (Hin), *H. reptans* (Hr), *Herpetocypris* sp. (Hsp), *Heterocypris incongruens* (Hi), *H. salina* (Hs), *Heterocypris* sp. (Hts), *Ilyocypris bradyi* (Ib), *I. decipiens* (Id), *I. inermis* (Ii), *Ilyocypris* sp. (Is), *Limnocythere inopinata* (Li), *Limnocythere* sp. (Ls), *Neglecandona neglecta* (Nn), *Potamocypris fallax* (Pf), *P. fulva* (Pfu), *P. similis* (Ps), *P. smaragdina* (Psm), *P. variegata* (Pv), *Potamocypris* sp. (Psp), *Prionocypris zenkeri* (Pz), *Pseudocandona albicans* (Pa), *Psychrodromus olivaceus* (Po), *Psychrodromus* sp. (Pssp), *Stenocypris* sp. (Ss) and *Stenocypris bolieki* (Sb)

pH, dissolved oxygen concentration (DO), calcium (Ca<sup>2+</sup>), magnesium (Mg<sup>2+</sup>), and elevation (Elev)) did not display multicollinearity. Then after, the triple combination of these six environmental variables ( $C(6, 3) = 6!/(6-3)!*3! = 20$ ) were used in most of the analyses given below to estimate the best environmental variables group/groups explaining the variation in the ostracod species. Variation partitioning (VP) (Borcard et al. 1992) was applied to determine the

relative contribution of each predictor variable to elucidate the variation of ostracod species in the triple combination (or model) of abiotic variables using the adjusted  $R^2$  (Peres-Neto et al. 2006). The importance of each model was tested by aid of 999 random permutations. The sum of all the explained variations and residual variance may exceed "1" or "100%" when looking at the VP results because of the presence of the negative explained variances and certain relationships in

the data. Distribution of ostracod species among the abiotic variables used in VP analysis was displayed in ternary plots for better visualization. Generalized Linear Models (GLM) were performed for the presence-absence of ostracod species data using binomial family and logit link functions (Zuur et al. 2009) to see the effect of each predictor in the six abiotic variables and in the triple combination of them. Level of variation explained by the predictor variables in GLM was calculated with aid of the formula  $(\text{Null deviance} - \text{Residual deviance})/\text{Null deviance} \times 100$ , and this is termed as the *Pseudo-R*<sup>2</sup> throughout the manuscript. Null and Residual deviances represent how well response variable forecasting by a model only with intercept term and by a specific model with predictors, respectively. The significance of each model was tested by the Chi-square test (Zuur et al. 2009). The relationships between the abiotic variables (Tw, pH, DO, Ca<sup>2+</sup>, Mg<sup>2+</sup>, and Elev) and the ostracod species distribution matrix were examined by a distance-based linear model (DISTLM) / a distance-based redundancy analysis ordination (dbRDA) using Bray Curtis similarities and Akaike Information decision Criterion (AICc) in PRIMER v7 with PERMANOVA+ (Clarke and Gorley 2015). Species data were Hellinger transformed because of including many zeros (Peres-Neto et al. 2006; Legendre and Gallagher 2001) for Variation partitioning and DISTLM / dbRDA analyses, while log transformation was applied to environmental variables to get the near-normal distribution except pH in PAST

3.26 software (Hammer et al. 2001) for the last analysis. A weighted averaging regression was used to estimate the optimum (Opt) and tolerance (Tol) levels of species for explanatory variables using C2 Software (Juggins 2003). A non-parametric Spearman Rank Correlation analysis was used to test the meaningful correlations between species and environmental variables, and among environmental variables (IBM-SPSS Statistics Version 21). The packages Vegan 1.5–7 (Oksanen et al. 2020) and glmm (Knudson et al. 2018) in R Version 3.6.3 (R Core Team 2020) were used to perform Variation partitioning and Generalized Linear Model analyses, respectively. For ternary plots, ggtern package (Hamilton and Ferry 2018) in R Version 4.1.2 (R Core Team 2021) were utilized. Above mentioned packages in different versions of R-statistics were runned with aid of RStudio software v.1.4.1103 (R studio Team 2021). In all statistical analyses, adult individuals occurring at least two or more times with complete soft body parts and carapaces were used.

## Results

The descriptive statistics of abiotic variables measured in the present study are given in Table 2. A total of 34 ostracod taxa (24 recent and 10 sub-fossil) were found in the present study (Tables 1 and 3). High ostracod taxa diversity was

**Table 2** Descriptive statistics of abiotic variables measured in the present study

All data													
	Tw	pH	EC	Sal	DO	Tot-N	Tot-P	Turb	Tot-Hard	Ca <sup>2+</sup>	Mg <sup>2+</sup>	TDS	Elev
Min	10.50	7.40	2.93	0.09	5.90	0.14	0.01	0.47	8.80	1.19	0.66	122.00	0.00
Max	26.00	8.80	7940.00	4.50	10.30	3.50	1.98	165.00	1195.50	181.81	190.73	1164.00	1413.00
Mean	17.26	8.00	872.57	0.47	7.92	0.76	0.07	12.94	250.36	60.36	24.19	258.32	449.98
Std	4.18	0.37	1827.60	1.03	0.87	0.91	0.28	30.90	222.09	25.77	42.60	222.07	541.19
Data of sampling sites bearing all living species													
	Tw	pH	EC	Sal	DO	Tot-N	Tot-P	Turb	Tot-Hard	Ca <sup>2+</sup>	Mg <sup>2+</sup>	TDS	Elev
Min	10.50	7.48	2.93	0.09	5.90	0.14	0.01	0.47	87.58	27.69	3.11	132.00	1.00
Max	26.00	8.77	7930.00	4.44	10.30	3.50	1.98	165.00	1195.50	181.81	190.73	1164.00	1413.00
Mean	17.25	8.08	877.29	0.48	7.89	0.75	0.09	13.12	262.04	63.34	25.23	264.60	484.64
Std	4.31	0.39	1804.02	1.02	0.94	0.92	0.33	32.60	223.19	26.39	42.90	225.38	550.66
Data of sampling sites bearing the living species encountered two or more times													
	Tw	pH	EC	Sal	DO	Tot-N	Tot-P	Turb	Tot-Hard	Ca <sup>2+</sup>	Mg <sup>2+</sup>	TDS	Elev
Min	10.50	7.48	2.93	0.09	5.90	0.14	0.01	0.47	87.58	27.69	3.11	132.00	1.00
Max	26.00	8.77	7930.00	4.44	10.30	3.50	1.98	165.00	1195.50	181.81	190.73	1164.00	1413.00
Mean	17.36	8.08	901.11	0.49	7.85	0.75	0.09	13.58	265.93	63.71	25.94	267.00	469.07
Std	4.32	0.39	1842.89	1.03	0.94	0.92	0.34	33.29	227.63	26.96	43.76	230.39	543.42

Abbreviations: minimum (Min), maximum (Max), standard deviation (Std), water temperature (Tw, °C), electrical conductivity (EC, μS cm<sup>-1</sup>), salinity (Sal, ‰), dissolved oxygen (DO, mg L<sup>-1</sup>), total nitrogen (Tot-N, mg L<sup>-1</sup>), total phosphate (Tot-P, mg L<sup>-1</sup>), turbidity (Turb, NTU), total hardness (Tot-Hard, mg L<sup>-1</sup>CaCO<sub>3</sub>), calcium (Ca<sup>2+</sup>, mg L<sup>-1</sup>), magnesium (Mg<sup>2+</sup>, mg L<sup>-1</sup>), total dissolved solids (TDS, mg L<sup>-1</sup>) and elevation (Elev, m asl.)

**Table 3** Distribution of recent, juvenile, carapace, and valve numbers of ostracod taxa between sampling seasons

Taxa	Code	Recent		Juvenile		Carapace		Valve	
		Oct-2020	Apr-2021	Oct-2020	Apr-2021	Oct-2020	Apr-2021	Oct-2020	Apr-2021
<i>Candona</i> sp.	Cs						1		
<i>Cyprideis torosa</i> (Jones, 1850)	Ct	270	290	9	30	6	37	1	11
<i>Cypridopsis vidua</i> (O.F. Müller, 1776)	Cv	46				3		1	
<i>Cypridopsis</i> sp.	Csp					1	7		
<i>Eucypris pigra</i> (Fischer, 1851)	Ep		11						5
<i>Eucypris</i> sp.	Es								1
<i>Fabaeformiscandona balatonica</i> (Daday, 1894)	Fb	1							
<i>Herpetocypris brevicaudata</i> Kaufmann, 1900	Hb	3	3						
<i>Herpetocypris helenae</i> G.W. Müller, 1908	Hh	2	8				1	1	13
<i>Herpetocypris intermedia</i> Kaufmann, 1900	Hin		3						
<i>Herpetocypris reptans</i> (Baird, 1835)	Hr		19				1		
<i>Herpetocypris</i> sp.	Hsp						2		1
<i>Heterocypris incongruens</i> (Ramdohr, 1808)	Hi	4	7			2	1	3	13
<i>Heterocypris salina</i> (Brady, 1868)	Hs	151	167	20		15	1	13	3
<i>Heterocypris</i> sp.	Hts						2	4	
<i>Ilyocypris bradyi</i> Sars, 1890	Ib	19	191			7	21	13	58
<i>Ilyocypris decipiens</i> Masi, 1905	Id	6				33		4	
<i>Ilyocypris inermis</i> Kaufmann, 1900	Ii	17				1		1	
<i>Ilyocypris</i> sp.	Is					12		9	6
<i>Limnocythere inopinata</i> (Baird, 1843)	Li	21				11		2	
<i>Limnocythere</i> sp.	Ls							3	2
<i>Neglecandona neglecta</i> (Sars, 1887)	Nn	5	8	1	1	1	1	1	4
<i>Potamocypris fallax</i> Fox, 1967	Pf	1							
<i>Potamocypris fulva</i> (Brady, 1868)	Pfu		1				1		1
<i>Potamocypris similis</i> G.W. Müller, 1912	Ps	3							
<i>Potamocypris smaragdina</i> (Vávra, 1891)	Psm	3				2			
<i>Potamocypris variegata</i> (Brady & Norman, 1889)	Pv	2				1			
<i>Potamocypris</i> sp.	Psp					2			
<i>Prionocypris zenkeri</i> (Chyzer & Toth, 1858)	Pz	237	178	19	18	33	15	28	28
<i>Pseudocandona albicans</i> (Brady, 1864)	Pa	3	1					2	
<i>Psychrodromus olivaceus</i> (Brady & Norman, 1889)	Po	4	34		1		4	1	23
<i>Psychrodromus</i> sp.	Pssp					2	2	7	12
<i>Stenocypris</i> sp.	Ss						1		
<i>Stenocypris bolieki</i> Ferguson, 1962	Sb	157				2		5	
Total		955	921	49	50	134	98	99	181

found in the coastline sampling site S7 (9 taxa (6 recent), followed by the inland parts sampling sites as S15 (7 recent), S17 (7 taxa (5 recent)) and S20 (7 taxa (6 recent)) (Table 1 and Online Resource 2). S7 was the hottest (mean water temperature = 21.4 °C) site among the above-mentioned sites. The mean pH values of these sites were ranged from 7.9 to 8.2 which indicates the slightly alkaline conditions. S7 had the lowest mean of calcium ( $\text{Ca}^{2+} = 62.1 \text{ mg L}^{-1}$ ) and the highest mean of magnesium ( $\text{Mg}^{2+} = 46.8 \text{ mg L}^{-1}$ ), when

S17 was of the lowest mean  $\text{Mg}^{2+}$  ( $6.8 \text{ mg L}^{-1}$ ) and highest mean  $\text{Ca}^{2+}$  ( $77.1 \text{ mg L}^{-1}$ ) values, compared with S15 and S20. They were ranked from low to high as S17, S20, S15, and S7 in the sense of elevation (Online Resource 1).

Of the 24 recent species, ten were collected only in October of 2020 while four species (*Eucypris pigra* (Fischer, 1851), *Herpetocypris intermedia* Kaufmann 1900, *H. reptans* (Baird, 1835) and *Potamocypris fulva* (Brady, 1868)) in April of 2021, and the rest species were common among

both sampling periods (Table 3). *Ilyocypris bradyi* Sars, 1890 was the most common species with an occurrence frequency (Ocfr) of 17 times, followed by *Prionocypris zenkeri* (Chyzer & Toth, 1858) with a frequency of 12 times (Online Resource 2). In the sense of abundance, *I. bradyi* was the last one among first five species with 210 individuals (ind.) while the first four are *Cyprideis torosa* (Jones, 1850) (560 ind. from three Ocfr), *P. zenkeri* (415 ind. from 12 Ocfr), *Heterocypris salina* (Brady, 1868) (318 ind. from five Ocfr) and *Stenocypris bolieki* Ferguson, 1962 (157 ind. from two Ocfr) (see Table 3 and Online Resource 2).

The explained percentage fractions of variation in the ostracod species by abiotic variables in different models based on the triple combination of water temperature (Tw, °C), pH, dissolved oxygen concentration (DO, mg L<sup>-1</sup>), calcium (Ca<sup>2+</sup>, mg L<sup>-1</sup>), magnesium (Mg<sup>2+</sup>, mg L<sup>-1</sup>) and elevation (Elev, m asl.) were given in Fig. 2. While all models were statistically significant ( $p < 0.05$ ), models presented in Figs. 2r (Tw + Ca<sup>2+</sup> + pH), 2s (pH + DO + Ca<sup>2+</sup>), 2t (Tw + pH + DO) and 2u (Tw + pH + Elev) were not. Among the statistically important models, the highest explanation power was reported for DO + Elev + Mg<sup>2+</sup> with 12.66% (Fig. 2a) whereas Ca<sup>2+</sup> + Tw + DO was the lowest with 4.11% (Fig. 2o). Overall models, Mg<sup>2+</sup> was the single abiotic variable delineating the most fractions in the Tw + pH + Mg<sup>2+</sup> model with 6.77% (Fig. 2h) that is followed by Elev in the DO + Elev + pH model with 4.61% (Fig. 2m). Highest elucidation fractions in the double (3.20%) and triple (0.72%) intersections were observed between Ca<sup>2+</sup> and Mg<sup>2+</sup> in Ca<sup>2+</sup> + Mg<sup>2+</sup> + Elev model (Fig. 2e) and among Ca<sup>2+</sup> + DO + Mg<sup>2+</sup> (Fig. 2g), respectively. Relatively high unexplained fractions from 88.62% to 99.13% were found in all models (Fig. 2). The distributions of the species among the triple combinations of abiotic variables were displayed in the ternary plots in Fig. 2. These distributions showed changes from partially homogeneous (e.g., Fig. 2a, b) to highly clustered structures (e.g., Fig. 2o–t) when the explained variations in ostracod species going from high to low.

The ordination of ostracod species according to the effect of predictor variables on their distribution was displayed in Fig. 3 after the application of distance-based linear model (DISTLM) and distance-based redundancy analysis (dbRDA). The dbRDA elucidated only 14.1% of the total variation in the species distribution matrix. The first two axes of dbRDA explained 67.1% of the fitted variations that is meaning a high correlation between predictor environmental variables and species distribution matrix. According to the correlation's coefficients between predictor variables and axes, axis 1 was constructed by magnesium (Mg<sup>2+</sup>, 46.4%) and elevation (Elev, 78.1%) when axis 2 by calcium (Ca<sup>2+</sup>, 74.4%) and water temperature (Tw, 54.1%). The distribution matrix of ostracod species was significantly affected

by Mg<sup>2+</sup> ( $Pseudo-F = 2.92$ ,  $p = 0.004$ ) and Elev ( $Pseudo-F = 3.55$ ,  $p = 0.002$ ) but not by DO ( $Pseudo-F = 1.49$ ,  $p = 0.152$ ), Ca<sup>2+</sup> ( $Pseudo-F = 1.47$ ,  $p = 0.164$ ), Tw ( $Pseudo-F = 1.21$ ,  $p = 0.272$ ) and pH ( $Pseudo-F = 0.26$ ,  $p = 0.989$ ).

The Generalized Linear Model (GLM) revealed the effects of predictor variables on the presence-absence probability of only *P. olivaceus* (Brady & Norman, 1889), *C. torosa*, *H. helenae* G.W. Müller, 1908, *N. neglecta* (Sars, 1887) and *P. albicans* (Brady, 1864) (Table 4). Dissolved oxygen (DO) showed a positive efficacy on the occurrence probability of *P. olivaceus* in all triple combination groups of abiotic variables, and elevation (Elev) also displayed a similar effect for this species in Ca<sup>2+</sup> (calcium) + DO + Elev and pH + DO + Elev groups. Calcium indicated positive impact on the occurrence probability of *C. torosa* in all triple groups, but negative for *H. helenae* in Ca<sup>2+</sup> + pH + Mg<sup>2+</sup> (magnesium) group. The occurrence probability of *P. albicans* was positively affected by Ca<sup>2+</sup> in two groups. The GLM results showed the negative influence of DO and water temperature (Tw) on the occurrence probabilities of *N. neglecta* and *P. albicans*, while another regional factor, Elev, indicated a positive effect on the occurrence probabilities of both species (see Table 4).

Spearman Rank correlation analysis pointed out the negative and positive correlations of water temperature with elevation ( $r_s = -0.408$ ,  $p < 0.01$ ) and pH ( $r_s = 0.465$ ,  $p < 0.01$ ), respectively. Negatively important correlations were observed between dissolved oxygen and calcium ( $r_s = -0.393$ ,  $p < 0.01$ ), and between elevation and magnesium ( $r_s = -0.49$ ,  $p < 0.01$ ). *Prionocypris zenkeri*, as the only species showing an important association with one of the abiotic variables, exhibited a positively strong correlation with magnesium ( $r_s = 0.689$ ,  $p < 0.05$ ).

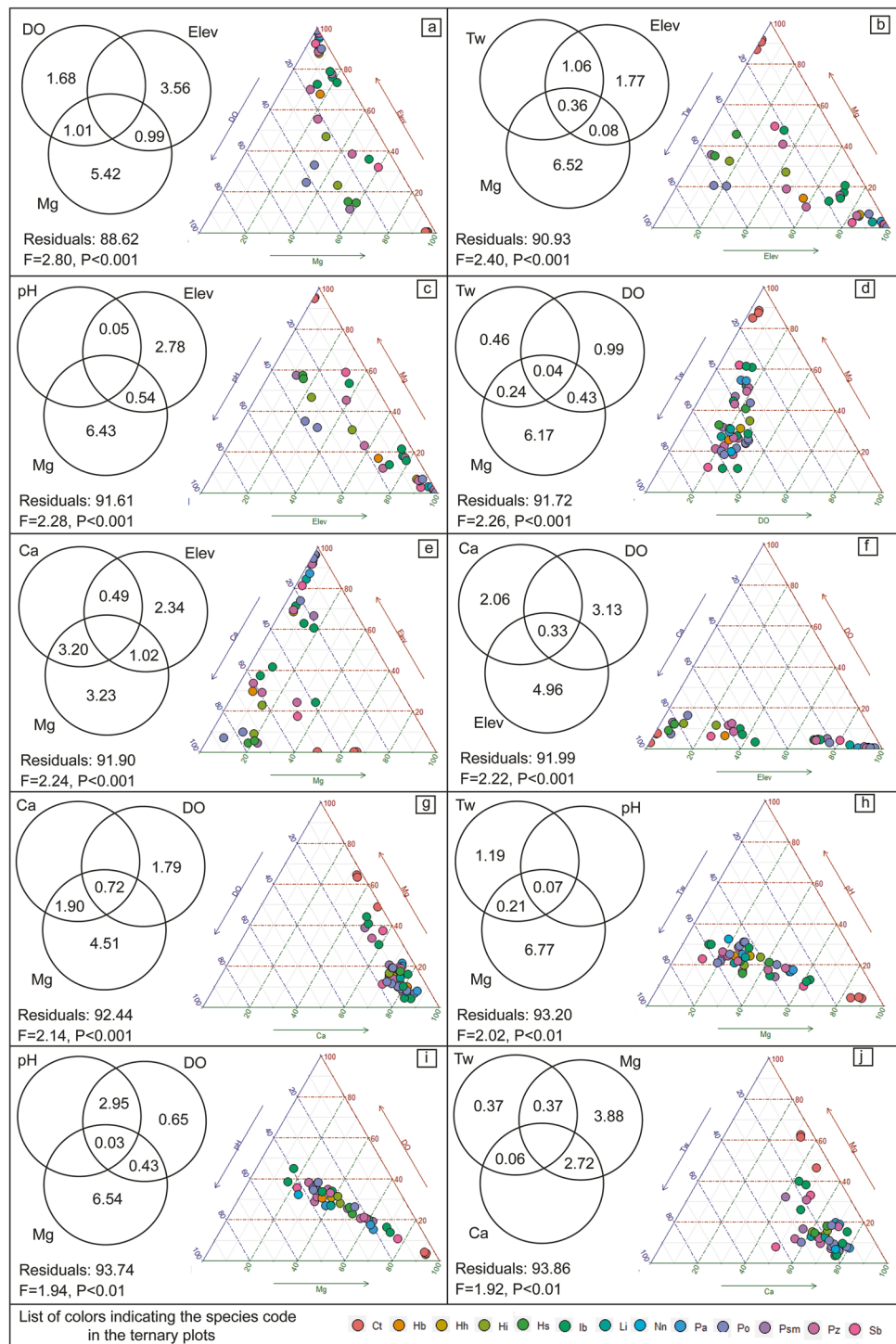
Optimum and tolerance levels of species encountered at least two times in the present study for water temperature, pH, calcium, magnesium, elevation and dissolved oxygen were dedicated in Table 5. Accordingly, *H. helenae* and *P. olivaceus* were of lowest and highest optimum levels for pH (7.76) and dissolved oxygen (8.73 mg L<sup>-1</sup>), respectively. *Cyprideis torosa* was the only species having highest optimum levels for both of calcium (129.66 mg L<sup>-1</sup>) and magnesium (180.35 mg L<sup>-1</sup>) (see Table 5).

## Discussion

### Seasonal species diversity

Relatively high species diversity was found in the present study. The ostracod taxa per sampling sites (or ratio = 1.1) herein was approximately 2.5 times higher than the ratio (0.43) obtained from the study including 117 sampling sites in the Mersin province in October 2015 (Dalgakiran et al. 2020). The low ratios were also reported in studies handling

**Fig. 2** Venn diagram indicating the fraction of variation explained by the individual variables and combination of them in each model constructed by the triple combination of water temperature (Tw, °C), pH, dissolved oxygen concentration (DO, mg L<sup>-1</sup>), calcium (Ca<sup>2+</sup>, mg L<sup>-1</sup>), magnesium (Mg<sup>2+</sup>, mg L<sup>-1</sup>) and elevation (Elev, m asl.) according to variation partitioning of ostracod communities, and ternary plots showing the distribution of ostracod species among the triple combination of abiotic variables. Residuals show the unexplained fractions in each model and species codes were given in Table 3. Lower case letters from a to u indicate the models and ternary plots constructed by the triple combination of used six environmental variables

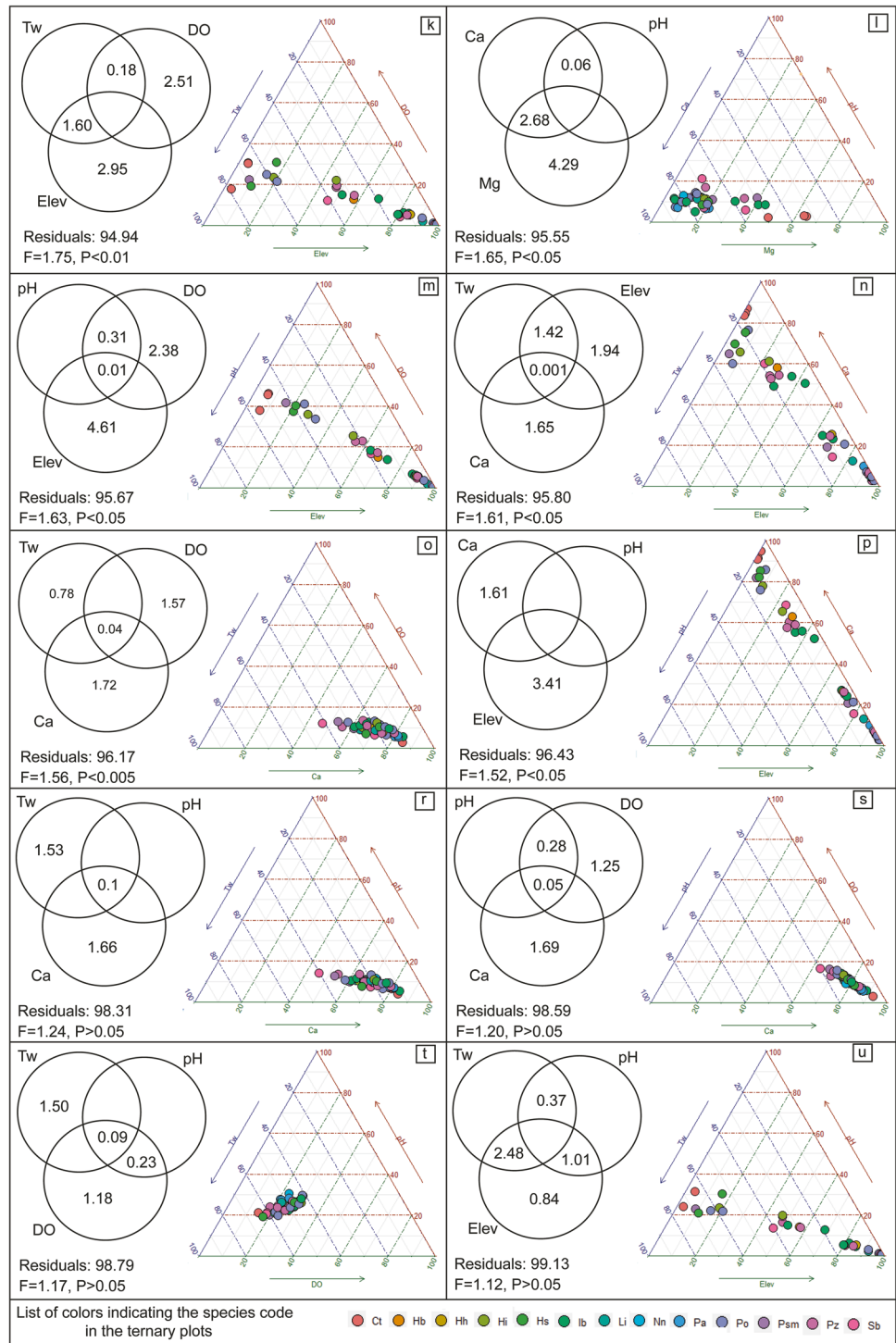


in the close or neighbor regions, e.g., a ratio of 0.44 getting from the 63 samplings in Osmaniye province in May 2015 (Külköylüoğlu et al. 2021), 0.3 ratio in Hatay province after 70 samplings in summer of 2012 (Akdemir and Külköylüoğlu 2021), ratios of 0.81, 0.68 and 0.66 recorded from the 26 sites in Konya closed, 22 sites in Antalya and 32 sites in West Mediterranean basins sampled in August

and October of 2017, respectively (Yavuzatmaca 2019) and a ratio of 0.41 acquired from the 68 sites sampled in July of 2014 in Muğla province (Akdemir et al. 2020). Of them, Yavuzatmaca (2019) announced the highest diversity of ostracods in October for Konya closed and West Mediterranean basins like the present study (22 taxa in April and 30 taxa in October, Table 3), while it was the highest in August



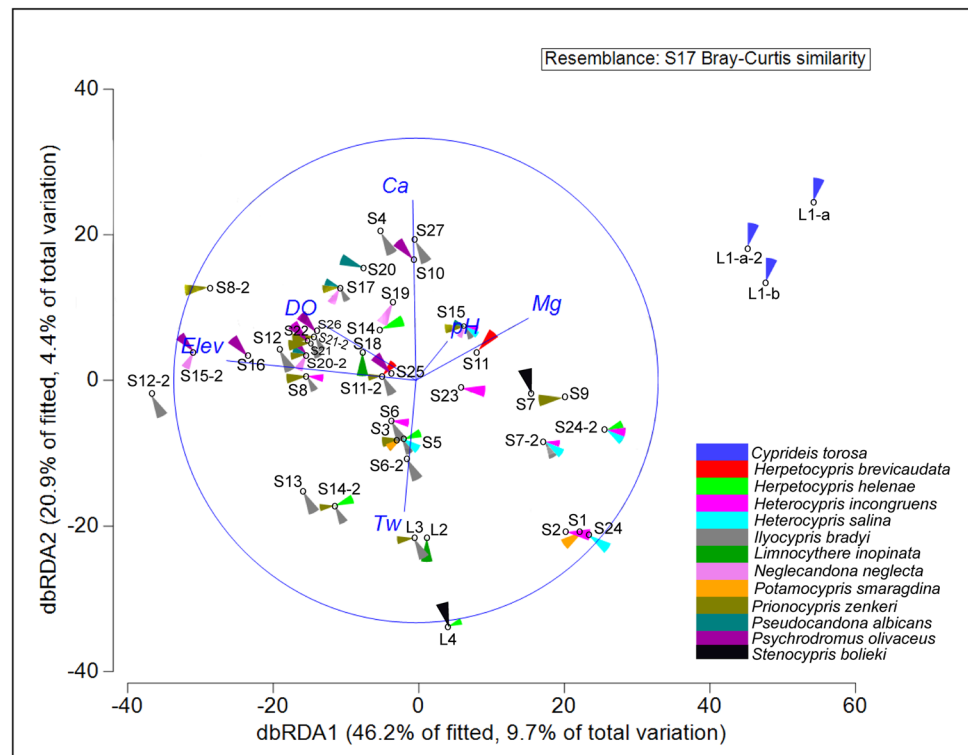
Fig. 2 (continued)



for Antalya River basin (13 species in August, eight species in October, and six species common). Most recently, the ratios 0.63 and 0.73 were noted from the 41 sites sampled in three seasons (spring, summer and autumn) of 2018 in the Eastern Black Sea and from the 40 sites sampled in three seasons of 2019 in the Eastern Anatolian regions of Turkey by Yavuzatmaca (2020a) and Yavuzatmaca (2021),

respectively. In both studies, the highest species diversity was recorded in autumn (Yavuzatmaca 2020a) and summer (Yavuzatmaca 2021) seasons. Like the above given studies including more than one habitat types from Turkey, lower ratios from the present study were found in the studies outside of Turkey, e.g., a ratio of 0.44 from 106 sampling sites sampled in summer of 2004 and 2005 in Western Mongolia

**Fig. 3** Distance-based redundancy analysis (dbRDA) of the ostracod species distribution matrix and environmental variables (arrows). Abbreviations of environmental variables were provided in Table 2, and the number (2) after the sampling site code (e.g., S24–2) means the sampling in the April of 2021. Distribution of species in each sampling site (circle) was showed using triangles with different colors



(Van der Meeren et al. 2010) and 0.83 ratio from 49 sites visited from 2006 to 2008 in different months and seasons in subarctic and temperate Europe (Iglukowska and Namiołko 2012). As stated before (Külköylüoğlu et al. 2016; Yavuzatmaca 2020a), species diversity does not increase with the number of sampling sites up to a critical level because seasonality appears play important role more than the number of sampling sites. In addition, although it seemed that the autumn season (from September to November in Turkey) can be rich in species diversity, higher diversity can also be found in different seasons even in regions close to the study area. Thereby, regional (or local) differences should be considered, and then regional biodiversity studies should be conducted by investigating which season or seasons can be better in terms of species richness.

### Species diversity and stream orders

Streams (S7, S15, S17 and S20) with high taxa diversity in the present study were at least 2nd order streams according to Strahler (1957) classification system, which supports the finding of Yavuzatmaca (2020a) as low ostracod species diversity in 1st order streams. The reason of it has been linked to the streams' size increasing from head (or 1st order) to following streams (2nd, 3rd order streams) (Vander Vorste et al. 2017) because an ascending trend is observed in the species diversity with that (Vannote et al. 1980). Examination of this issue has not been widely discussed

for ostracods in literature so studies revealing species distributions and characteristics according to streams' size are needed to provide ecologically important information for estimation of habitat characteristics (lotic in this sense) using ostracods.

### Explanation power of environmental variables on the species composition

The changing of rates of variation explained by each of the individual variables combined with other variables effective on the ostracod species composition can be seen in Fig. 2. Even if the percentage of explanation powers of models were low, the best model might be the dissolved oxygen+elevation+magnesium (DO+Elev+Mg<sup>2+</sup>) with 12.66% (Fig. 2a) followed by pH+DO+Mg<sup>2+</sup> with 10.6% (Fig. 2i). The importance of local factors (e.g., DO, Mg<sup>2+</sup>, Tw) and the secondary effect of Elev (as a regional factor) were commonly discussed in the literature for ostracods (e.g., Viehberg 2006; Uçak et al. 2014; Yavuzatmaca et al. 2018). However, changing of one variable in the models caused a decrease of important percentage in the explained variation, e.g., 2.38, 2.86 and 2.87% reduction were observed when the accompanying variable of Mg<sup>2+</sup> and Elev was calcium (Ca<sup>2+</sup>, Fig. 2e), pH (Fig. 2c) and Tw (Fig. 2b) instead of DO (Fig. 2a), respectively. The situation was the same when checking the whole models in Fig. 2. These results emphasized the importance of understanding

**Table 4** Generalized Linear Model results indicating the significant effects of each abiotic variable (Explanatory variables) in all abiotic variables and in the triple combinations of them (Group) on the occurrence probability of ostracod species (Model)

Group	Model	Explanatory variables	$\beta$ + SE	Z-value	p value	AIC	Pseudo-R <sup>2</sup>	Chi-square / p -value
Tw + pH + Ca <sup>2+</sup> + Mg <sup>2+</sup> + Elev + DO	Po	Intercept	-24.90 ± 19.45	-1.28	0.200	37.59	38.25	Chi = 14.617 p = 0.02
		DO	2.23 ± 1.01	2.21	0.027**			
Tw + pH + Ca <sup>2+</sup>	Ct	Intercept	-0.25 ± 25.36	-0.01	0.99	19.42	47.52	Chi = 10.341 p = 0.02
		Ca <sup>2+</sup>	0.07 ± 0.04	1.95	0.05*			
Tw + Ca <sup>2+</sup> + DO	Ct	Intercept	-10.29 ± 9.68	-1.06	0.29	19.47	47.30	Chi = 10.292 p = 0.02
		Ca <sup>2+</sup>	0.08 ± 0.04	1.97	0.048**			
pH + DO + Ca <sup>2+</sup>	Ct	Intercept	-4.17 ± 24.82	-0.17	0.87	19.40	47.58	Chi = 10.354 p = 0.02
		Ca <sup>2+</sup>	0.08 ± 0.04	1.96	0.049**			
Ca <sup>2+</sup> + pH + Mg <sup>2+</sup>	Hh	Intercept	28.09 ± 17.00	1.65	0.10	29.47	30.55	Chi = 9.445 p = 0.02
		Ca <sup>2+</sup>	-0.15 ± 0.08	-1.86	0.062*			
Tw + pH + DO	Nn	Intercept	-7.49 ± 17.39	-0.43	0.67	28.36	34.13	Chi = 10.551 p = 0.01
		Tw	-0.51 ± 0.27	-1.91	0.056*			
Tw + pH + Ca <sup>2+</sup>	Nn	Intercept	-30.36 ± 17.64	-1.72	0.09	29.29	31.12	Chi = 9.62 p = 0.02
		Tw	-0.62 ± 0.30	-2.06	0.039**			
Tw + pH + Mg <sup>2+</sup>	Nn	Intercept	-19.94 ± 14.06	-1.42	0.16	30.85	26.07	Chi = 8.06 p = 0.04
		Tw	-0.44 ± 0.23	-1.92	0.055*			
Tw + Mg <sup>2+</sup> + DO	Nn	Intercept	15.51 ± 7.23	2.15	0.032**	28.80	32.70	Chi = 10.109 p = 0.02
		DO	-1.52 ± 0.75	-2.02	0.043**			
Tw + Ca <sup>2+</sup> + DO	Nn	Intercept	15.59 ± 8.10	1.93	0.054*	30.14	28.37	Chi = 8.77 p = 0.03
		Tw	-0.36 ± 0.20	-1.76	0.079*			
		DO	-1.51 ± 0.79	-1.90	0.057*			
Tw + Ca <sup>2+</sup> + Elev	Nn	Intercept	-3.21 ± 4.20	-0.76	0.44	29.21	31.39	Chi = 9.702 p = 0.02
		Elev	0.003 ± 0.002	1.80	0.072*			
pH + Mg <sup>2+</sup> + Elev	Nn	Intercept	-8.29 ± 11.76	-0.71	0.48	29.95	28.98	Chi = 8.958 p = 0.03
		Elev	0.003 ± 0.002	1.95	0.05*			
Ca <sup>2+</sup> + Mg <sup>2+</sup> + Elev	Nn	Intercept	-5.87 ± 2.74	-2.14	0.03**	29.12	31.66	Chi = 9.788 p = 0.02
		Elev	0.003 ± 0.001	1.89	0.06*			
Ca <sup>2+</sup> + pH + Elev	Nn	Intercept	-11.79 ± 12.47	-0.94	0.35	29.46	30.57	Chi = 9.45 p = 0.02
		Elev	0.003 ± 0.001	2.14	0.03**			
Ca <sup>2+</sup> + DO + Elev	Nn	Intercept	-0.86 ± 7.26	-0.12	0.91	29.23	31.32	Chi = 9.682 p = 0.02
		Elev	0.003 ± 0.002	1.90	0.06*			
Tw + Mg <sup>2+</sup> + Elev	Nn	Intercept	-1.97 ± 3.92	-0.50	0.61	29.62	30.06	Chi = 9.293 p = 0.03
		Elev	0.003 ± 0.002	1.69	0.09*			
Tw + pH + DO	Pa	Intercept	28.06 ± 22.29	1.26	0.21	20.85	51.71	Chi = 13.763 p = 0.003
		DO	-2.64 ± 1.23	-2.14	0.03**			
Tw + pH + Ca <sup>2+</sup>	Pa	Intercept	-25.37 ± 18.32	-1.39	0.17	23.67	41.11	Chi = 10.941 p = 0.01
		Tw	-1.01 ± 0.51	-2.01	0.04**			
Tw + Elev + DO	Pa	Intercept	-0.40 ± 12.02	1.43	0.15	19.77	55.77	Chi = 14.844 p = 0.002
		DO	-2.17 ± 1.20	-1.81	0.07*			
Tw + Mg <sup>2+</sup> + DO	Pa	Intercept	25.30 ± 10.93	2.31	0.02**	20.87	51.63	Chi = 13.741 p = 0.003
		DO	-2.58 ± 1.15	-2.24	0.03**			
Tw + Ca <sup>2+</sup> + DO	Pa	Intercept	23.16 ± 10.84	2.14	0.03**	20.42	53.33	Chi = 14.194 p = 0.003
		DO	-2.36 ± 1.16	-2.03	0.04**			
Tw + Ca <sup>2+</sup> + Mg <sup>2+</sup>	Pa	Intercept	1.31 ± 4.17	0.31	0.75	26.12	31.93	Chi = 8.497 p = 0.04
		Ca <sup>2+</sup>	0.06 ± 0.03	1.88	0.06*			
Tw + Ca <sup>2+</sup> + Elev	Pa	Intercept	-6.40 ± 6.89	-0.93	0.35	20.42	53.35	Chi = 14.20 p = 0.003
		Ca <sup>2+</sup>	0.08 ± 0.04	1.72	0.09*			
Ca <sup>2+</sup> + DO + Mg <sup>2+</sup>	Pa	Intercept	10.26 ± 7.58	1.35	0.18	24.72	37.19	Chi = 9.898 p = 0.02
		DO	-1.99 ± 1.04	-1.92	0.06*			
Ca <sup>2+</sup> + pH + Elev	Pa	Intercept	-8.09 ± 15.70	-0.52	0.61	21.74	48.36	Chi = 12.872 p = 0.005
		Elev	0.006 ± 0.003	1.81	0.07*			
pH + DO + Elev	Pa	Intercept	31.93 ± 21.14	1.51	0.13	19.82	55.58	Chi = 14.793 p = 0.002
		DO	-2.40 ± 1.39	-1.73	0.08*			

**Table 4** (continued)

Group	Model	Explanatory variables	$\beta \pm SE$	Z-value	p value	AIC	Pseudo-R <sup>2</sup>	Chi-square / p -value
pH+DO+Mg <sup>2+</sup>	Pa	Intercept	35.80 ± 21.03	1.70	0.09	24.86	36.66	Chi=9.758 p=0.02
		DO	-2.75 ± 1.25	-2.20	0.03**			
pH+DO+Ca <sup>2+</sup>	Pa	Intercept	39.47 ± 23.34	1.69	0.09	26.04	32.20	Chi=8.571 p=0.04
		DO	-2.79 ± 1.35	-2.07	0.04**			
Tw+Elev+DO	Po	Intercept	-16.80 ± 6.51	-2.58	0.01	35.11	29.05	Chi=11.10 p=0.01
		DO	1.83 ± 0.72	2.56	0.01**			
Tw+Mg <sup>2+</sup> +DO	Po	Intercept	-8.09 ± 5.14	-1.58	0.12	35.67	27.58	Chi=10.538 p=0.01
		DO	1.21 ± 0.63	1.93	0.05*			
Tw+Ca <sup>2+</sup> +DO	Po	Intercept	-14.72 ± 7.29	-2.02	0.04**	38.16	21.06	Chi=8.048 p=0.04
		DO	1.74 ± 0.81	2.14	0.03**			
DO+Mg <sup>2+</sup> +Elev	Po	Intercept	-14.51 ± 6.53	-2.22	0.03**	34.00	31.95	Chi=12.209 p=0.007
		DO	1.58 ± 0.71	2.24	0.03**			
Ca <sup>2+</sup> +DO+Mg <sup>2+</sup>	Po	Intercept	-12.34 ± 6.12	-2.02	0.04**	35.14	28.97	Chi=11.068 p=0.01
		DO	1.26 ± 0.63	2.00	0.04**			
Ca <sup>2+</sup> +DO+Elev	Po	Intercept	-25.25 ± 9.55	-2.65	0.008***	33.46	33.37	Chi=12.75 p=0.005
		DO	2.39 ± 0.92	2.59	0.009***			
		Elev	0.003 ± 0.001	2.25	0.02**			
pH+DO+Elev	Po	Intercept	-15.74 ± 11.44	-1.38	0.17	35.23	28.73	Chi=10.976 p=0.01
		DO	1.80 ± 0.70	2.57	0.01**			
		Elev	0.002 ± 0.001	2.14	0.03**			
pH+DO+Mg <sup>2+</sup>	Po	Intercept	-7.15 ± 12.17	-0.59	0.56	36.49	25.44	Chi=9.721 p=0.02
		DO	1.09 ± 0.60	1.81	0.07*			

Significant levels \*0.1, \*\*0.05, \*\*\*0.01

Abbreviations: parameter estimates ( $\beta$ ), standard error (SE), Akaike Information Criterion (AIC), and percentage level of variation explained by the predictor variables (Group) in species presence-absence probability (*Pseudo-R*<sup>2</sup>). The significantly effective variables on the presence-absence of species in the statistically meaning group models at a cut-off level 0.05 (Chi-square (Chi) / p value) are only showed in the Table. Codes of abiotic variables (Group) and species (Model) are provided in Tables 2 and 3, respectively

the intertwined and complex relationships among abiotic variables to determine whether which variable increases or decreases the effectiveness of the accompanying variable/s. These findings are also enabled to estimate the best environmental variable groups elucidating the species composition variation.

Elevation displays a negative association with the rate of dissolved gases (e.g., DO, CO<sub>2</sub>) because of the effect of barometric pressure on their solubility (Goldman and Horne 1983). In addition, ionic salinity because of decreasing the available intermolecular space (Goldman and Horne 1983) and temperature (Wetzel 2001) have negative correlations with the solubility or occurrences of gases (e.g., DO) in waters. Similarly, a negatively significant correlation was reported between DO and Ca<sup>2+</sup> in the present study. Main rock structure in the studied area, limestone, is a mixture of calcite (CaCO<sub>3</sub>) and dolomite (MgCO<sub>3</sub>) but mostly CaCO<sub>3</sub> in nature that is dissolved by the disintegration of CO<sub>2</sub> in waters to form carbonic acid (Boyd et al. 2016). Then, this weak acid solubilizes limestone and resulted in an increase in the amount of ionized Ca<sup>2+</sup> and HCO<sub>3</sub><sup>1-</sup> in water (Wetzel 2001). Using of HCO<sub>3</sub><sup>1-</sup> for photosynthesis increase the CO<sub>3</sub><sup>2-</sup> and OH<sup>1-</sup> causing pH to rise. The presence of

Ca<sup>2+</sup> limits the increase of pH by precipitating CO<sub>3</sub><sup>2-</sup> as CaCO<sub>3</sub>. This implies the relationships among pH, DO and the availability of Ca<sup>2+</sup> in water. Considering the limestone dominancy in the studied area and the pH range (7.40–8.80 (Table 2)), the availability of Ca<sup>2+</sup> in water seems a non-limiting factor for the ostracods to calcify their valves. Although Mg<sup>2+</sup> is found in the limestone, its main source is dolomite rock, and Mg<sup>2+</sup> is indicated as a counterpart of Ca<sup>2+</sup> because of their similar chemistry (Goldman and Horne 1983). However, Mg<sup>2+</sup> compounds are more soluble than Ca<sup>2+</sup>. Therefore, important amount of MgCO<sub>3</sub> and magnesium hydroxide start to precipitate when the pH of waters increases to very high levels (>10) (Wetzel 2001).

After the information given above, the explanatory power of Mg<sup>2+</sup> mostly increased with water temperature (Fig. 2b–h) in the present study but the level of this power changed according to the variable added as the 3rd in the model. For instance, elevation is a factor indirectly affecting the concentration of Mg<sup>2+</sup> in waters, and a significantly negative relationship was also found between them in the present study ( $p < 0.01$ ). Also, the distribution of species found in waters at high elevations corresponds to low Mg<sup>2+</sup> content in the ternary plot when looking at Fig. 2b. The

**Table 5** Optimum (Opt) and tolerance (Tol) levels of ostracod species for different abiotic variables

Species	Count	Max	N <sub>2</sub>	Water temperature (°C)		pH		Calcium (mg L <sup>-1</sup> )		Magnesium (mg L <sup>-1</sup> )		Elevation (m asl.)		Dissolved oxygen (mg L <sup>-1</sup> )	
				Opt	Tol	Opt	Tol	Opt	Tol	Opt	Tol	Opt	Tol	Opt	Tol
<i>Heterocypris incongruens</i>	7	3	5.76	16.87	3.34	8.01	0.38	55.75	16.50	23.84	13.64	274.55	457.23	7.66	0.95
<i>Pseudocandona albicans</i>	4	1	4.00	13.78	1.82	8.08	0.07	78.89	25.62	16.25	9.90	1223.25	300.83	6.78	0.88
<i>Neglecandona neglecta</i>	5	4	3.93	14.25	2.48	8.11	0.23	65.04	25.43	8.54	5.78	1144.46	340.19	7.62	1.29
<i>Herpetocypris helenae</i>	5	5	3.13	17.40	3.30	7.76	0.46	47.55	8.34	9.56	2.79	100.00	66.45	8.37	0.61
<i>Psychrodromus olivaceus</i>	7	20	2.96	15.60	4.56	8.01	0.31	56.47	13.60	7.74	3.40	809.37	511.14	8.73	0.77
<i>Cyprideis torosa</i>	3	270	2.88	20.28	5.89	8.20	0.42	129.66	53.87	180.35	11.80	1.00	276.68	6.92	1.05
<i>Ilyocypris bradyi</i>	17	130	2.47	15.76	2.70	7.79	0.38	92.82	41.73	20.72	11.93	177.33	366.29	7.89	0.69
<i>Prionocypris zenkeri</i>	12	220	2.32	14.52	1.78	7.88	0.39	81.37	12.50	22.27	5.63	892.09	304.60	6.71	1.07
<i>Heterocypris salina</i>	5	165	2.16	21.53	5.21	8.01	0.24	55.40	16.79	29.76	17.31	30.96	130.97	7.64	0.77
<i>Herpetocypris brevicaudata</i>	2	3	1.92	16.70	0.35	8.07	0.16	63.23	7.54	6.76	1.89	33.00	276.68	8.02	0.85
<i>Potamocypris smaragdina</i>	2	2	1.80	24.00	0.21	8.53	0.52	41.21	9.84	24.20	10.18	88.33	90.51	8.63	0.78
<i>Limnocythere inopinata</i>	2	20	1.10	16.96	2.33	8.52	0.35	56.46	6.39	10.66	0.40	1011.24	413.72	7.04	0.18
<i>Stenocypris bolieki</i>	2	150	1.09	24.60	2.83	8.63	0.28	30.00	36.65	6.58	33.51	135.01	61.52	7.30	0.82

Abbreviations: numbers of species occurrence (Count), maximum numbers of individuals (Max) and Hill’s coefficient or measure of an effective number of occurrences (N<sub>2</sub>)

explanatory power of Mg<sup>2+</sup> was the highest alone (Fig. 2h) when the 3rd factor was pH but decreased partially when DO used (Fig. 2d). On the other hand, Ca<sup>2+</sup> (Fig. 2j) was the 3rd factor, power of Mg<sup>2+</sup> was almost halved, while the joint explanatory power rises. This may be due to the importance of Mg<sup>2+</sup> and Ca<sup>2+</sup> for the calcification of ostracod shells and their similar chemistry (see above). The increasing power of Mg<sup>2+</sup> with Tw may be explained by the correlation between Tw and ostracod shells’ Mg<sup>2+</sup> content (Palacios-Fest and Dettman 2001) because Tw has the management roles on the minor element composition of ostracod shells (see Dettman et al. 2002). Water temperature also affects the development, body size and life cycle of ostracods (Ruiz et al. 2013; Aguilar-Alberola and Mesquita-Joanes 2014) that may elucidate the reason why 2.87% of variations were less explained when using Tw instead of DO with Elev+Mg<sup>2+</sup> (Fig. 2a, b). The effects of water temperature on the ostracods can be more effective on the growth period and its molting stage, in other words, it is more effective on juveniles rather than adults. Considering the used adults in the last stage of molting in all analyses, it can be understood why dissolved oxygen-bearing metabolic importance for aerobic organisms displayed the higher explanation power than Tw in the present study. Furthermore, DO was also effective against pH, when used together with Ca<sup>2+</sup> and Mg<sup>2+</sup> because the variation explained by the model with DO was 8.92% (Fig. 2g), while the model with pH explained only 7.03% (Fig. 2i). An increase of the activity of Ca<sup>2+</sup> intersection Mg<sup>2+</sup> along with pH was observed since they elucidated 2.68% (Fig. 2l) and 1.90% (Fig. 2g) of variations when the accompanying

variable was pH and DO, respectively. The activity of Mg<sup>2+</sup> was not significantly changed when using the DO (Fig. 2g) and pH (Fig. 2i) with Ca<sup>2+</sup>. When the efficiency of Ca<sup>2+</sup> and DO was examined, the activity of DO was higher than that of Ca<sup>2+</sup> checking the models made with Elev (Fig. 2f) and Mg<sup>2+</sup> (Fig. 2g). Looking the models constructed with Ca<sup>2+</sup>, Tw and pH (Fig. 2r), and others (see Fig. 2), a ranking like Ca<sup>2+</sup>, Tw and pH can be seen according to their explanation powers on the variation of ostracod species. In all, the importance or explanation power of environmental variables on the ostracod species composition in the present study can be listed as Mg<sup>2+</sup>, Elev, DO, Ca<sup>2+</sup>, Tw and pH.

The answer to question “Why is Mg<sup>2+</sup> more effective than other variables?” lie in the geology of Turkey. This is because Turkey is located on a dense tectonic activity consisting of approximately 40% of carbonate and evaporitic rocks suitable for dissolution, and this ratio can also reach to a value of 60% when caves as the characteristics of ground karstification are considered (Nazik and Poyraz 2017). Therefore, it is expected that the dissolved Ca<sup>2+</sup> ratio in waters will be higher than Mg<sup>2+</sup>, and the results of the studies conducted in different regions of Turkey also strengthen this argument. Of them, Yavuzatmaca et al. (2017a) reported a higher mean Ca<sup>2+</sup> value (71.26 mg L<sup>-1</sup>) than Mg<sup>2+</sup> (15.25 mg L<sup>-1</sup>) in the Sinop province in the Black Sea region of Turkey, and similarly Külköylüoğlu et al. (2020) shared mean Ca<sup>2+</sup> = 46.9 mg L<sup>-1</sup> and Mg<sup>2+</sup> = 9.63 mg L<sup>-1</sup> values for the ten sampling sites in Artvin province located in the same region of Turkey. The similar results were also published in the Kütahya

province ( $\text{Ca}^{2+} = 69.6 \text{ mg L}^{-1}$ ,  $\text{Mg}^{2+} = 30.7 \text{ mg L}^{-1}$ ) in Aegean (Külköylüoğlu et al. 2018), Muğla province ( $\text{Ca}^{2+} = 55.17 \text{ mg L}^{-1}$ ,  $\text{Mg}^{2+} = 17.54 \text{ mg L}^{-1}$ ) in the Southwest (Akdemir et al. 2020), Malatya province ( $\text{Ca}^{2+} = 85.2 \text{ mg L}^{-1}$ ,  $\text{Mg}^{2+} = 31.3 \text{ mg L}^{-1}$ ) in East Anatolia (Batmaz et al. 2020) and in Mersin province ( $\text{Ca}^{2+} = 58.11 \text{ mg L}^{-1}$ ,  $\text{Mg}^{2+} = 11.53 \text{ mg L}^{-1}$ ) in Mediterranean (Dalgakıran et al. 2020) regions of Turkey. The  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$  ratios in water are of great importance for ostracods because they get the cation and anions (e.g.,  $\text{Ca}^{2+}$ ,  $\text{Mg}^{2+}$ ,  $\text{CO}_3^{2-}$ ) required to calcify their low magnesium calcite carapaces from the waters where they live (Turpen and Angell 1971). Accordingly,  $\text{Mg}^{2+}$  may be shown as a limiting factor for the ostracods in the current study when considering the rock formation of the studied area, and the mean  $\text{Ca}^{2+}$  ( $60.36 \text{ mg L}^{-1}$ ) and  $\text{Mg}^{2+}$  ( $24.19 \text{ mg L}^{-1}$ ) values in the present study (Table 2).

The results of the dbRDA also supported the importance of  $\text{Mg}^{2+}$  and Elev ( $p < 0.05$ ) among other variables for the distribution and abundance of ostracod species in the present study (Fig. 3). Similarly, the significant influence of  $\text{Mg}^{2+}$  (e.g., Viehberg 2006; Yavuzatmaca et al. 2017a) and Elev (e.g., Külköylüoğlu et al. 2019; Yavuzatmaca 2019, 2020b) on the occurrence and distribution of species were reported. Notwithstanding, this does not mean that other variables (DO,  $\text{Ca}^{2+}$ , Tw and pH) are not important for the distribution of ostracods. This is because their importance was emphasized for many times before in and out of Turkey (e.g., Van der Meer et al. 2010; Iglkowska and Namiotko 2012; Akdemir et al. 2020; Külköylüoğlu et al. 2020, 2021) and even in a study where the studied area overlapping with some of the studied area here in the present study (Dalgakıran et al. 2020). Most recently, Cusminsky et al. (2020) highlighted the effects of EC, elevation, and pH for the ostracod assemblages in Patagonian (Argentinian) ecoregions and stated that they are followed by  $\text{Mg}^{2+}$  and Tw. All these suggest that the effects of especially local factors on the distribution of ostracods may vary from region to region, in the sampling season or times, and even in the sampled habitat differences. Therefore, revealing the ecoregion-based effective factors should be the topic of future studies to efficiently use indicator species to estimate past and present environmental conditions.

### Environmental variables and individual species

GLM results showed the positive effect of  $\text{Ca}^{2+}$  on the presence of (Table 4) euryhaline widespread species, *C. torosa*, that is mostly occur in the brackish water of coastal areas (Meisch 2000). This is the case in the present study because the living and subfossil forms of species were encountered in the coastal sampling sites S1, S7, S24 and L1 (Fig. 1, Table 1, Online Resource 2). The positively

strong correlations of species with conductivity (Yavuzatmaca 2019), and  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$  (Akdemir et al. 2020) and the effect of  $\text{Mg}^{2+}$  level on its occurrence (Viehberg 2006) were announced. Recently, Gusakov et al. (2021) collected species from the polyhaline Chernavka River having high  $\text{Ca}^{2+}$  ( $0.92\text{--}1.44 \text{ g L}^{-1}$ ) and  $\text{Mg}^{2+}$  ( $0.68\text{--}0.89 \text{ g L}^{-1}$ ) levels of the Lake Elton Basin in the European territory of the Russian Federation, and they declared a very high upper limit of salinity tolerances ( $96\text{--}150 \text{ g L}^{-1}$ ) for *C. torosa*. The range of  $\text{Ca}^{2+}$  ( $88.41\text{--}181.81 \text{ mg L}^{-1}$ ) and  $\text{Mg}^{2+}$  ( $165.53\text{--}190.73 \text{ mg L}^{-1}$ ) of water where living form of species found (Online Resource 1) and the highest optimum levels for both  $\text{Ca}^{2+}$  ( $129.66 \text{ mg L}^{-1}$ ) and  $\text{Mg}^{2+}$  ( $180.35 \text{ mg L}^{-1}$ ) (Table 5) in the present study reinforced these previous statements about the *C. torosa*. The previously reported close relationship of the species with conductivity (see above) and the strong association between conductivity and  $\text{Ca}^{2+}$  (Iglkowska and Namiotko 2012) are considered, the answer to the question “why did the  $\text{Ca}^{2+}$  has a positive action on the occurrence of *C. torosa* in the present study?” has been given.

Mezquita et al. (1999b) pinpointed the occurrence of *H. helena* in  $\text{Mg}^{2+}$  enriched waters concerning  $\text{Ca}^{2+}$  and its preference for high dissolved oxygen and pH level. It was found in waters with high pH mean (8.04) but low  $\text{Ca}^{2+}$  ( $46.04 \text{ mg L}^{-1}$ ) and  $\text{Mg}^{2+}$  ( $10.05 \text{ mg L}^{-1}$ ) mean values (Online Resource 1), and it displayed low  $\text{Ca}^{2+}$  ( $47.55 \text{ mg L}^{-1}$ ) and  $\text{Mg}^{2+}$  ( $9.56 \text{ mg L}^{-1}$ ) optimum levels when compared with other species (Table 5) in the present study. Also, mean DO value ( $7.84 \text{ mg L}^{-1}$ ) of water where species present (Online Resource 1) showed conformity with the DO range ( $5.4\text{--}19.1 \text{ mg L}^{-1}$ ) (Uçak et al. 2014; Mezquita et al. 1999b) getting from the literature. All these findings support that *H. helena* prefers waters with low  $\text{Ca}^{2+}$  level because escalating of DO and pH cause the depletion of  $\text{Ca}^{2+}$  and so the  $\text{Mg}^{2+}$  and  $\text{Ca}^{2+}$  levels of water begin approach to each other as emphasized by Mezquita et al. (1999b). Thereby, the negatively significant effect  $\text{Ca}^{2+}$  on the occurrence of *H. helena* in the present study was a promising finding (Table 4) due to the geologic rock structure of the studied area.

*Neglecandona neglecta* is a well-known cosmopolitan species (Külköylüoğlu 2013). Tolerance of species to hypoxic condition (below  $3 \text{ mg L}^{-1}$  DO) (Meisch 2000), its presence in anoxic environment ( $= 0.32 \text{ mg L}^{-1}$  DO) (Külköylüoğlu 2009) and an important negative correlation of it with DO (Külköylüoğlu et al. 2014) were previously documented. After all, Yavuzatmaca (2020b) underlining statistically important indicator potential of *N. neglecta* for DO level equals  $9.10 \text{ mg L}^{-1}$  showed conformably the close relationship of the species with DO in streams (Yavuzatmaca 2021). Rieradevall and Roca (1995) indicated the contradictory effect of high Tw on the abundance of *N. neglecta*, while low Tw having positive outcome on its development

in Lake Banyoles, Spain. Yavuzatmaca (2020b) found the separation of a group of sampling sites possessing mean Tw value corresponds to 19.7 °C from other by *N. neglecta*, when the negatively significant relationships of species with Tw were presented in literature (e.g., Yılmaz and Külköylüoğlu 2006; Yavuzatmaca 2021). Both DO and Tw have negative relationships with Elev (see mentioned above) but *N. neglecta* formerly displayed positive correlation with Elev (Pieri et al. 2009; Yavuzatmaca 2019). In the present study, elevation revealed a positive result on the occurrences of *N. neglecta*, while Tw and DO negatively affect the occurrence of the species (Table 4). Also, *N. neglecta* had high (1144 m asl.) and low (14.25 °C) optimum level for Elev and Tw, respectively, after *P. albicans* (Table 5). Models constructed with Tw, DO, Elev, pH, Ca<sup>2+</sup> and Mg<sup>2+</sup> explained a range from 26.07% to 34.13% in the variation of the occurrence probability of *N. neglecta* (Table 4). The causation of these low percentage ratios may be the wide tolerance of the species to those variables (e.g., Tw, pH, DO, Elev). The validity of this view is consistent with the ranges of variables significantly affecting the occurrence probability of *N. neglecta* provided by Yavuzatmaca et al. (2017b), e.g., Tw (2.13–28.9 °C), DO (0.32–15.4 mg L<sup>-1</sup>) and Elev (0–3194 m asl.). Like *N. neglecta*, the cosmopolitan species (Külköylüoğlu et al. 2012a), *P. albicans*, had been found in the wide ranges of DO (0.75–15.8 mg L<sup>-1</sup>), Tw (2.9–29.2 °C) and Elev (61–2290 m asl.) (Yavuzatmaca et al. 2017b). Among the importantly effective variables, DO and Tw showed higher negative coefficients than variables (Elev and Ca<sup>2+</sup>) that had positive coefficients on the probability of *P. albicans* (Table 4), while its positive association with Tw (Yavuzatmaca 2021) but negative with Tw and Elev (Külköylüoğlu et al. 2012b) were shown. Although the percentages explained (31.93–55.77%) by the models in Table 4 were higher than *N. neglecta*, the activities of the variables were similar to *N. neglecta*. The range of Ca<sup>2+</sup> (41.18–98.35 mg L<sup>-1</sup>) (Online Resources 1 and 2) for waters where *P. albicans* gathered exhibited conformity with the range (10–160 mg L<sup>-1</sup>) given in Iglukowska and Namiotko (2012). The optimum (78.89 mg L<sup>-1</sup>) and tolerance (25.52 mg L<sup>-1</sup>) levels of Ca<sup>2+</sup> for species (Table 5) were higher than the optimum (66.1 mg L<sup>-1</sup>) but lower than tolerance (35.04 mg L<sup>-1</sup>) levels in Batmaz et al. (2020). Also, Van der Meeren et al. (2010) encountered species in waters with %Ca<sup>2+</sup> mean equals to 55.9 in Western Mongolia. All these suggest that species prefers a high Ca<sup>2+</sup> level that is also supported by the positive role of Ca<sup>2+</sup> on the occurrence probability of *P. albicans* in the present study (Table 4).

*Psychrodromus olivaceus* is another well-known cosmoecious species (Külköylüoğlu 2013) and it was found in a wide range of DO (1.74–20 mg L<sup>-1</sup>) and Elev (0.5–1700 m asl.) (Yavuzatmaca et al. 2017b). Even

though Külköylüoğlu et al. (2013) write up the negative correlation of species with DO, the indicative potential of *P. olivaceus* for two groups of sampling sites having high DO mean values (8.62 and 9.64 mg L<sup>-1</sup>) were estimated in Yavuzatmaca (2020b). A strong positive correlation of species with elevation ( $r_s = 0.88$ ) was demonstrated by Yavuzatmaca et al. (2018), and then after, Dalgakıran et al. (2020) find the changing of length and height of *P. olivaceus* across elevational ranges and reported the high tolerance of species to elevation. Highest optimum for DO (8.73 mg L<sup>-1</sup>) and tolerance for Elev (511.4 m asl.) (Table 5) and the positive effect of DO and Elev on the occurrence of species (Table 4) reinforce these previous findings. Although the species has wide tolerance to environmental variables, it can be said that DO has a positive effect on the presence of species. The cosmopolitan species, *P. zenkeri*, showed a very strong correlation with the Mg<sup>2+</sup> but it was encountered in a limited range of Mg<sup>2+</sup> (5.69–25.4 mg L<sup>-1</sup>) in the present study (Online Resources 1 and 2). The optimum (22.27 mg L<sup>-1</sup>) level of species for Mg<sup>2+</sup> in the present study (Table 5) was higher than the levels equal to 16.65 mg L<sup>-1</sup> and 12.29 mg L<sup>-1</sup> given in Batmaz et al. (2020) and Dalgakıran et al. (2020), respectively. For the abundance of species (Online Resources 1 and 2), it could be seen that the highest abundance of species are found in sites having high Mg<sup>2+</sup> concentrations as S15 (Mg<sup>2+</sup> = 25.4 mg L<sup>-1</sup>; 220 individuals (ind.)) and S21 (Mg<sup>2+</sup> = 20.1 mg L<sup>-1</sup>, 160 ind.) and both were sampled in October of 2020 but only 3 ind. were collected in S9 sampled in April of 2021 with 23.5 mg L<sup>-1</sup> Mg<sup>2+</sup> concentration (Online Resources 1 and 2). In terms of the species colonization, the low abundance in S9 with a high Mg<sup>2+</sup> value in the April sampling seems normal since the S9 station dried at all in October. These results show that high Mg<sup>2+</sup> values favor the abundance of *P. zenkeri*.

## Conclusion

A relatively high ostracod taxon diversity (34 taxa) was detected in four lakes and 27 streams located in the Eastern Mediterranean region of Turkey. The model constructed with DO+Elev+Mg<sup>2+</sup> was found as the best model to declare the variation in the species composition that is followed by pH+DO+Mg<sup>2+</sup>. Among environmental variables, Mg<sup>2+</sup> and Elev showed statistically important direct effects on species composition in the present study. The variation in activities of environmental variables on the species has been observed when comparing the results in the present study with the finding reported in different geographical regions even if the species (e.g., cosmopolitan species) have wide tolerance levels to ecological variables. This pinpoints the importance of ecoregion-based studies because they will be better to reveal

the environmental variable preferences of the species and the activities of these variables. Using of findings as presented in this study may result in more accurate data as compared to general ecological data in terms of estimation of current or past environmental conditions by using bioindicator species (e.g., ostracods). This deduction supports the statement of Willis and Whittaker (2002) as variables having important roles for the local and/or recent time species richness may not be such factors for the richness at regional spatial scale or longer time scale. Therefore, the increase in the number of studies to determine the region-based ecological preferences of species in the future will allow us to use species more effectively as bioindicators.

**Supplementary Information** The online version contains supplementary material available at <https://doi.org/10.1007/s11756-022-01208-2>.

**Acknowledgments** My special thanks go to Prof. Dr. Okan Küllköylüoğlu (Bolu Abant İzzet Baysal University, Turkey), Prof. Dr. Abuzer Çelekli (Gaziantep University, Turkey) and Mrs. Filiz Batmaz (Bolu Abant İzzet Baysal University, Turkey) for their comments on the first draft. I also thank to Mrs. Mary Theresa Dorothy Williams (North Carolina State University) for her help with English. I would like to thank Mr. Ömer Lekeşiz (Gaziantep University, Turkey) for his help to construct the map.

**Funding** This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

## Declarations

**Conflict of interest** The author declares that he has no conflict of interest.

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