

# The Recent non-marine ostracods of Tunisia: an updated checklist with remarks on their regional distribution patterns and ecological preferences

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## ABSTRACT

Different lines of investigation have recently contributed to increasing the available knowledge about the invertebrates inhabiting inland waters of north Africa, but a comprehensive synopsis on Tunisian Ostracoda is missing to date. An updated checklist of Recent non-marine ostracods from Tunisia and data on their distribution is thus offered here, representing the most extensive survey on this crustacean group ever carried out in inland waters throughout the country. One-hundred-five sites covering various climate zones, from Mediterranean to desert areas, were sampled between 2002 and 2012. Most of the considered water bodies were temporary or ephemeral habitats, but a few permanent sites were sampled as well. Overall, 18 genera and 32 taxa of putative species rank were collected in the frame of this survey, among which nine species and five genera were new to Tunisian fauna. As a result of this study and based on previous investigations, nine families (Candonidae, Cyprididae, Cytheroidea, Darwinulidae, Ilyocyprididae, Leptocytheridae, Limnocytheridae, Loxoconchidae, Paradoxostomatidae), 29 genera and at least 45 species of non-marine ostracods are currently known for Tunisia, which thus prove to host the most diverse ostracod fauna among north African countries. The number of species occurring in a single sample varied from 1 to 4. The *Eucypris virens* complex was the most widespread taxon (58 records), followed by *Heterocypris barbara* (30 records), *Heterocypris incongruens* (22 records), and *Sarscypridopsis aculeata* (16 records). For some ostracod species, clear distributional gradients associated with different climatic conditions were observed. The affinities with adjacent Maghrebian ostracod faunas are discussed. This study confirms the crucial role played by marginal aquatic habitats for the conservation of biodiversity, in particular in arid and semi-arid regions.

## INTRODUCTION

Ostracods are a class of small bivalved crustaceans occurring in almost all aquatic ecosystems (Smith *et al.*, 2015). In inland surface waters, both characterised by temporary and permanent hydroperiods, they abound in the benthic and periphytic habitats. According to Meisch

*et al.* (2019), there are presently 2330 subjective species of non-marine ostracods in 270 genera.

Until a few years ago, information on non-marine ostracods of the Maghreb was largely based on the contributions by Gurney (1909), Gauthier (1928a, 1928b, 1928c), Klie (1943) and, to a lesser extent, on scattered data in the literature. Only recently, new studies have contributed towards increasing the knowledge on their distribution in this area and with the description of new species (among them, Danielopol *et al.*, 1990; Marmonier *et al.*, 2005; Schmit *et al.*, 2013a). Zaibi *et al.* (2013) reviewed the existing literature on the occurrence of non-marine ostracods in Tunisia and provided new data from a survey carried out in 15 sites from the central and southern part of the country. Overall, 30 non-marine species were recorded by these authors, including *Vestalenula* sp. B (Danielopol, 1980), and additional three taxa were identified at supra-specific level. Later on, Scharf *et al.* (2014) reported the non-native *Candonopsis novaezelandiae*, a species previously unknown in Tunisia, from five sites located in the northern part of the country.

In addition to the above mentioned studies on ostracods, other lines of investigation have recently contributed to increasing available knowledge about invertebrates of Tunisian inland waters, such as aquatic beetles (Touaylia *et al.*, 2011), erpobdellid leeches (Ahmed *et al.*, 2013),

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branchiopods and copepods (Turki and Turki 2010; Marrone *et al.*, 2016; Stoch *et al.*, 2016), freshwater brachyurans (Marrone *et al.*, 2020), and epigeal amphipods (Ayati *et al.*, 2019).

The high diversity of Tunisian landscape and waterbodies (Morgan, 1982; Stoch *et al.*, 2016), the strong gradient observed in its climatic conditions and its geographic position between the eastern and western basins of the Mediterranean, make Tunisia notably interesting as a study area, in particular for biogeographic researches on aquatic fauna.

Here we present an updated checklist of Recent ostracod fauna from Tunisia and data on taxa distribution, which are based on the most extensive survey on this crustacean group ever carried out in inland waters throughout the country.

## METHODS

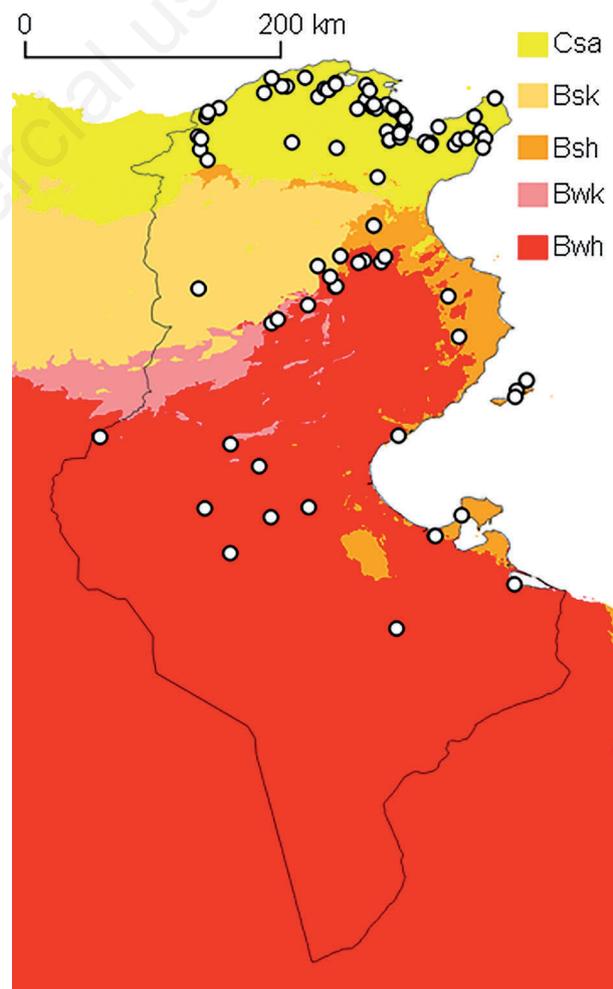
Sampling sites were selected in order to encompass all the climatic areas and the most representative types of natural inland aquatic habitats of Tunisia (Fig. 1; Tab. 1). Some examples of sampling sites investigated in this study are presented in Figs. S1 and S2.

Altogether, 106 ostracod samples were collected using a handnet with a mesh size of 200  $\mu\text{m}$  from 103 sites between 2002 and 2012. *Ex situ* rehydration of sediment collected from dry temporary habitats, the so-called “Sars’ method” as described in Marrone *et al.* (2019), allowed to study the ostracod assemblages from further two sites (Tab. 1). Geographical coordinates were recorded with GPS. Each sampling site was identified by an alphanumeric code consisting of a letter followed by a number, and then assigned to one of the following Köppen-Geiger climate zones according to Beck *et al.* (2018): Csa (temperate, dry summer, hot summer); Bsk (arid, steppe, cold); Bsh (arid, steppe, hot); Bwk (arid, desert, cold); Bwh (arid, desert, hot). When possible, the following environmental features were recorded: habitat type, estimated hydroperiod, water temperature, water conductivity at 20°C, water turbidity, and macrophyte coverage. Water temperature and electric conductivity were measured *in situ* using portable digital meters. Arbitrary values ranging from 0 to 4 were considered to represent water turbidity (0 = crystal clear water; 4 = extremely turbid water or argillotrophic system) and macrophyte coverage (0 = total absence of vegetation; 4 = absence of open water). The maps were generated using QGIS v. 3.4.15 (QGIS Development Team, 2018).

Ostracods were sorted under a binocular microscope and then fixed in 90% ethanol. The taxonomic identification of the collected material was based on adult specimens. Both soft parts and valves were checked for the identification at the lowest possible level, using

Danielopol *et al.* (1990), Meisch (2000), Rasouli *et al.* (2016) and Mazzini *et al.* (2014). Taxonomic nomenclature follows Meisch *et al.* (2019). Ostracod specimens are housed at the Department of Chemistry, Life Sciences and Environmental Sustainability, University of Parma, Italy. All species authorships are listed in Tab. 2.

Sample-based rarefaction curves (Gotelli and Colwell, 2001) were computed using EstimateS v. 9.1.0 software (Colwell, 2013) in order to evaluate if sampling effort was exhaustive enough to be representative of the Tunisian inland water ostracod fauna. The non-parametric species richness estimators ICE (Incidence Coverage-based Estimator) and Jack1 (first order Jackknife-based estimator) were calculated. Following the recommendation by Colwell (2013), the Bias-corrected formula of Chao2 estimator of species richness was excluded since the coefficient of variation of the incidence distribution was higher than 0.5, and in this case the estimator became imprecise. For



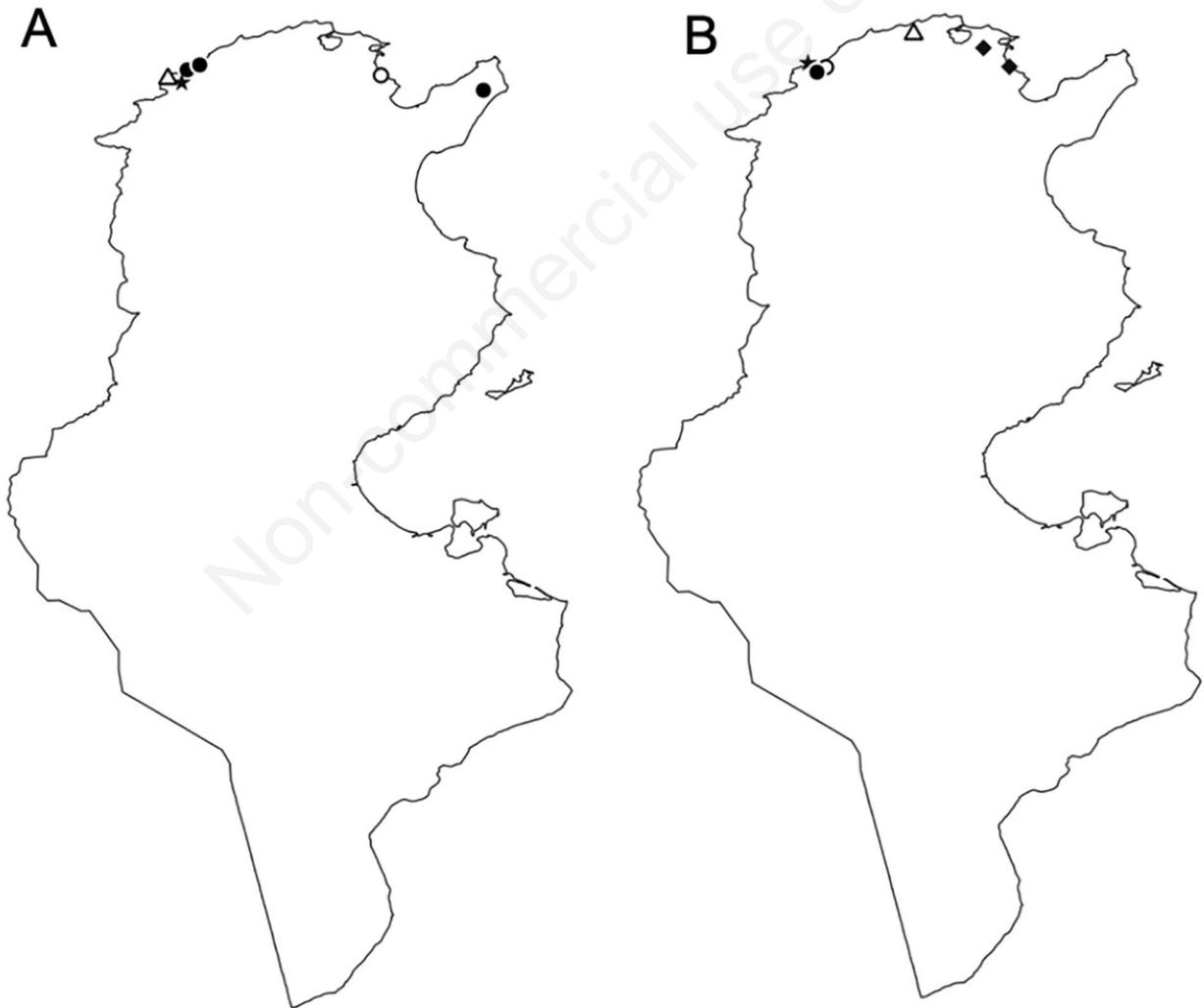
**Fig. 1.** Distribution of sampling sites in different Köppen-Geiger present-day (1980-2016) climate zones.

this reason, the ICE estimator was preferred over the classical Chao2 estimator, giving higher values, as recommended by Colwell (2013). The rarefaction curve of the mean values of “uniques” (*i.e.*, species present only in a single sample) was also calculated.

## RESULTS

A total of 32 taxa belonging to 18 genera in 4 families (Candonidae, Ilyocyprididae, Cyprididae and Limnocytheridae) were identified. Nine species (*Candona* cf. *muelleri*, *Cyclocypris laevis*, *Cypria ophthalmica*, *Eucypris mareotica*, *Heterocypris reptans*, *Isocypris beauchampi*, *Leucocythere* cf. *algeriensis*, *Potamocypris smaragdina*, *Potamocypris variegata*) and five genera (*Candona*, *Cy-*

*clocypris*, *Cypria*, *Isocypris*, *Leucocythere*) were new to Tunisia (Tab. 2). The number of species reported in a single sample varied from 1 (30 sites) to 4 (9 sites). Figs. 2-5 show the distribution of ostracods found in this study. The *Eucypris virens* complex was the most widespread taxon (in 58 sites, Fig. 4A), being very common in both Mediterranean and arid areas. It was characterised by a noteworthy morphological variability in the carapace shape, with three distinct morphotypes here annotated as “typical”, “elongate” and “subquadrate” forms, sometimes co-occurring in the same population. The “subquadrate” form was the most common (in 41 sites), followed by the “typical” (31 sites) and the “elongate” (15 sites) forms. The latter was distributed only in the northern part of the country (Fig. 4A). Other species that were commonly identified included *Het-*

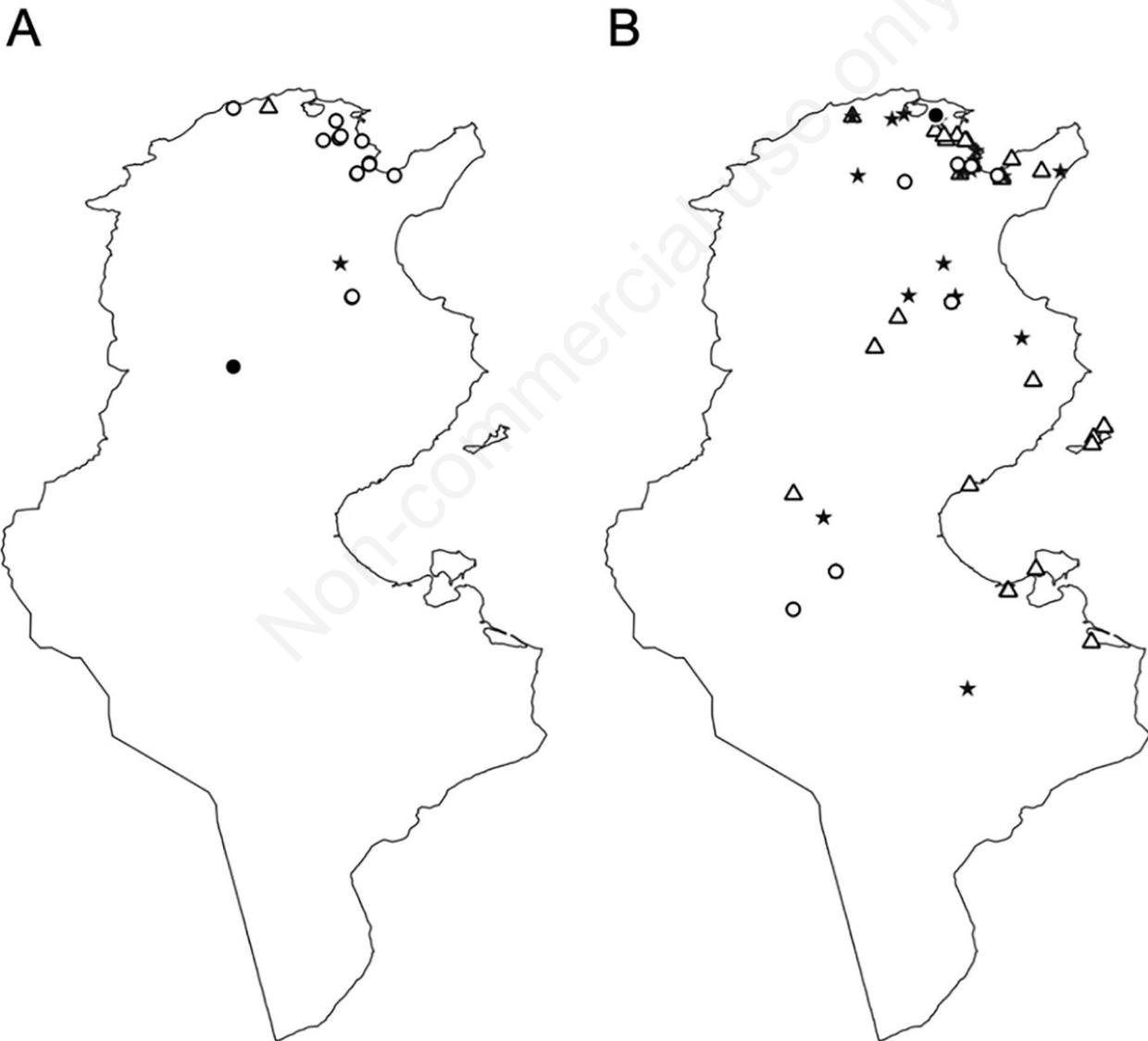


**Fig. 2.** A) distribution map of *Bradleystrandesia* sp. (Δ), *Bradleycypris obliqua* (★), *Cypris bispinosa* (●) and *Cypris pubera* (○) and B) *Cypridopsis* cf. *elongata* (Δ), *Cypridopsis hartwigi* (★), *Cypridopsis vidua* (●), *Cypridopsis* sp. (○) and *Plesiocypridopsis newtoni* (◆).

*erocypris barbara* (30 sites), *H. incongruens* (22 sites), and *Sarscypridopsis aculeata* (16 sites) (Figs. 3A and 3B). On the other hand, 13 taxa were represented in one site only (Tab. 2). More diverse ostracod assemblages mostly correspond to habitats with a higher vegetation coverage. *Heterocypris salina* and *S. aculeata* showed a marked preference for low-turbidity systems.

Most of the taxa occurred in the more temperate parts of the country, where temperature and precipitation are more suitable for the formation and persistence of more predictable aquatic habitats, and 19 were exclusively present under a Csa climate. *Potamocypris smaragdina* and *Potamocypris variegata* were recorded only in the steppic

and desert areas, respectively in Bsh and Bwk climatic zones (Fig. 3A). Other ostracods are adapted to cope with the physically harsh conditions that characterize the southern and inner part of Tunisia, among them *Heterocypris barbara*, *Heterocypris incongruens*, *Heterocypris salina*, *Eucypris mareotica*, *Eucypris virens* complex, *Cypridopsis vidua* and the only records of *Cypria ophthalmica* and *Ilyocypris bradyi*, all of them found in Bwh climatic zone (Figs. 2B, 3B, 4A, 5A and 5B). Five taxa (*Eucypris virens* complex, *Heterocypris barbara*, *Heterocypris incongruens*, *Ilyocypris getica* and *Tonnacypris lutaria*), in addition to Csa, were present in at least two other climatic zones (Figs. 4 A,B and 5B).

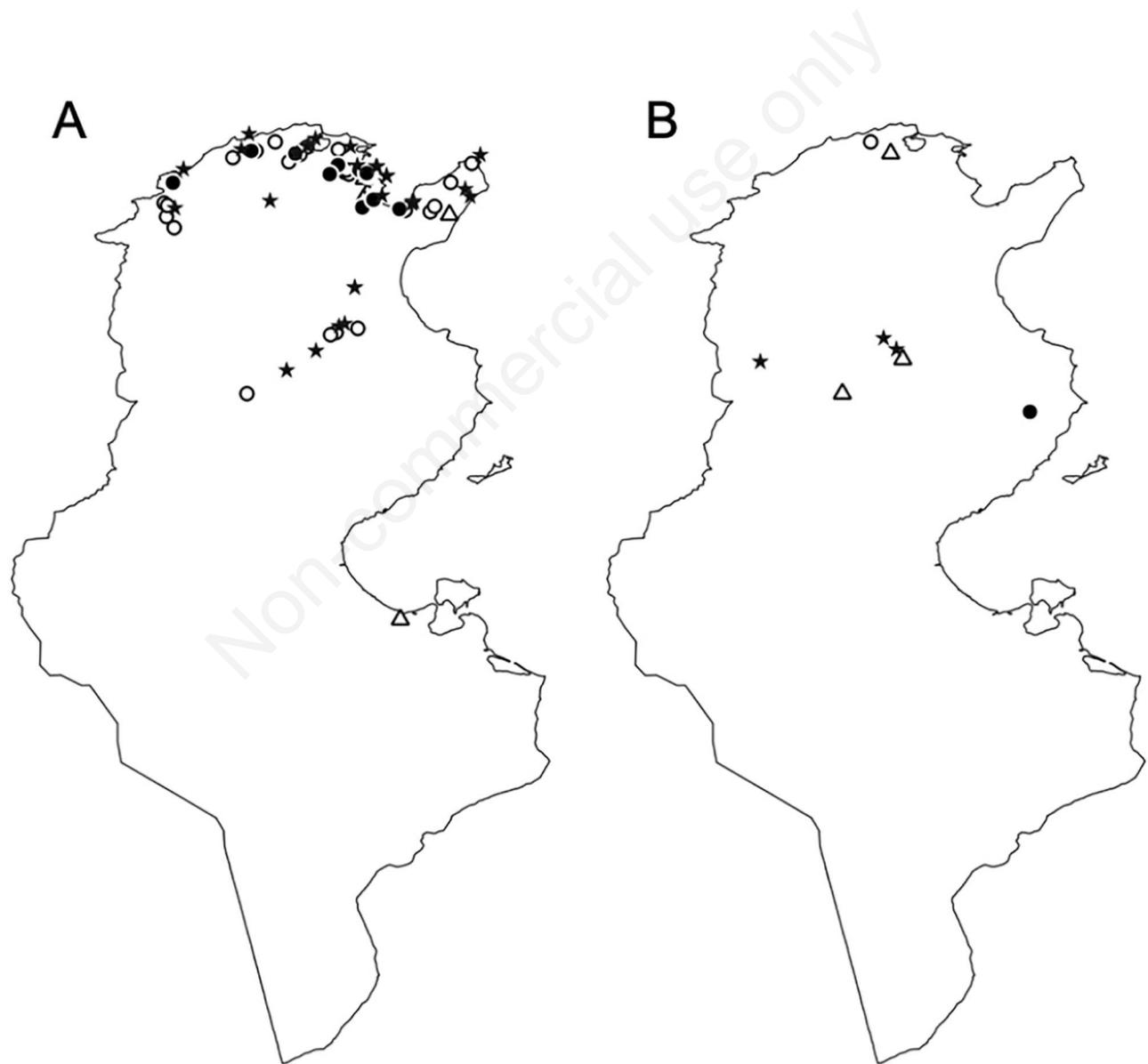


**Fig. 3.** A) distribution map of *Potamocypris arcuata* ( $\Delta$ ), *Potamocypris smaragdina* ( $\star$ ), *Potamocypris variegata* ( $\bullet$ ) and *Sarscypridopsis aculeata* ( $\circ$ ) and B) *Heterocypris barbara* ( $\Delta$ ), *Heterocypris incongruens* ( $\star$ ), *Heterocypris reptans* ( $\bullet$ ) and *Heterocypris salina* ( $\circ$ ).

Except for *Ilyocypris bradyi*, which was collected from permanent pools along river margins (F209), the other 10 taxa found in permanent sites were also present in temporary habitats. The sampled sites showed a wide range of salinity, from freshwater to hypersaline conditions (Tab. 1). Taking into account only the species found in at least four sites, those which solely occurred at low conductivity ( $<1 \text{ mS cm}^{-1}$ ) were *Ilyocypris getica* and *Tonnacypris lutaria*; other species seemed to prefer low to medium conductivity, as *Trajancypris clavata* and the “typical” morphotype of the *Eucypris virens* complex, or medium to high conductivity, as *Sarscypridopsis aculeata*. *Eucypris mareotica* and *Heterocypris salina* were

exclusively found in sites with conductivity  $>4 \text{ mS cm}^{-1}$ . The ostracods present in a wide range of salinity conditions were the “elongate” and “subquadrate” morphotypes of the *Eucypris virens* complex (between  $0.3$  and  $11.5 \text{ mS cm}^{-1}$  and between  $0.2$  and  $11.5 \text{ mS cm}^{-1}$ , respectively), *Heterocypris barbara* ( $0.4$ – $53.6 \text{ mS cm}^{-1}$ ) and *Heterocypris incongruens* ( $0.2$ – $5.6 \text{ mS cm}^{-1}$ ).

Rarefaction and estimation curves are shown in Fig. 6. The rarefaction curve was increasing, and no plateau could be observed. The ICE curve showed a reduction of its slope above 75 samples, while Jack1 showed a continuous increase. Uniques (species present in a single site) means tended to stabilize their trends at about 14 species (Fig. 6).

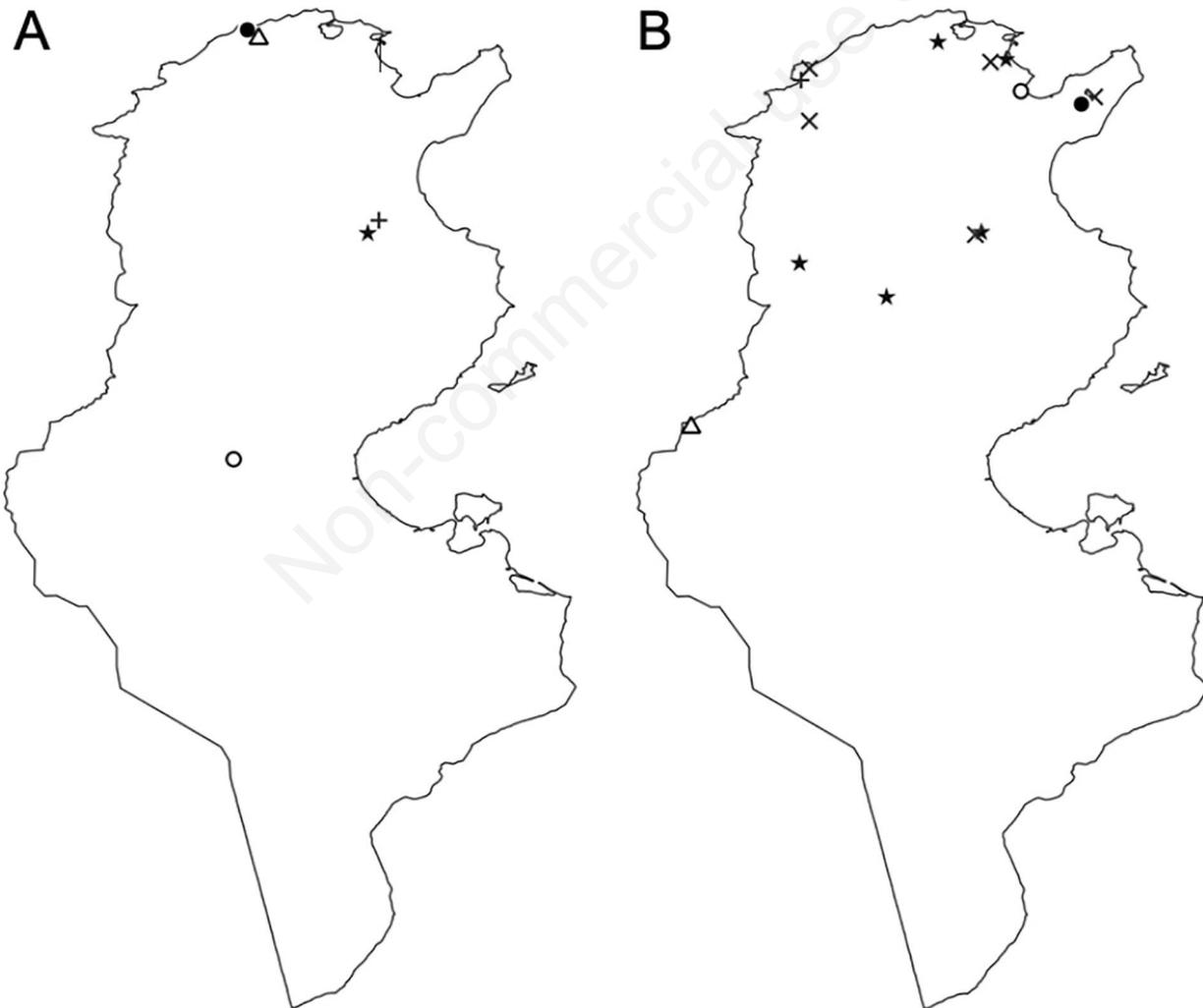


**Fig. 4.** A) distribution map of *Eucypris mareotica* ( $\Delta$ ) and *Eucypris virens* complex: “typical” form ( $\star$ ), “elongate” form ( $\bullet$ ), “subquadrate” form ( $\circ$ ) and B) *Tonnacypris lutaria* ( $\Delta$ ), *Trajancypris clavata* ( $\star$ ), *Herpetocypris* sp. ( $\bullet$ ) and *Isocypris beauchampi* ( $\circ$ ).

## DISCUSSION

The water bodies considered in this survey are mainly temporary or even ephemeral, with marked fluctuations in their environmental conditions (see Supplementary Material). The fauna detected is therefore made up of generally euryhaline, widely tolerant taxa that are capable of producing resting stages and tuning their life cycles according to the duration of the wet phase. Notwithstanding the constraints imposed by these severe conditions, the diversity of non-marine ostracod fauna of Tunisia, as revealed from data collected in this study and in previous ones, is rather high, consisting of nine families, 29 genera and at least 45 species (Tab. 2). Certainly the species diversity is still underestimated because in some cases the identification at species level was not achieved due to the preservation state

of the collected material, its scarcity or the absence of adult stages. In addition, in some ostracod genera, as in *Eucypris*, there are ill-defined (morpho)species (Meisch, 2000), consequent to the high phenotypic plasticity observed in valve shape and size (Baltanás *et al.*, 2002). *Eucypris virens* was the most frequently encountered ostracod in this study. Bode *et al.* (2010) indicated the existence of a species complex with more than 40 cryptic taxa, suggesting a revision of the single species status of *Eucypris virens*. More recently, Koenders *et al.* (2017) demonstrated that genetic species in the *Eucypris virens* complex cannot be recognized morphologically by valve shape. For these reasons, here we reported only the presence of different morphotypes into this species complex, to which no taxonomic significance must be attached; future investigation on genetic and morphological variation among these forms will hope-



**Fig. 5.** A) distribution map of *Candona cf. muelleri* (Δ), *Neglecandona neglecta* (★), *Cyclocypris laevis* (●), *Cyprina ophtalmica* (○) and *Leucocythere cf. algeriensis* (+) and B) *Ilyocypris bradyi* (Δ), *Ilyocypris getica* (★), *Ilyocypris cf. getica* (●), *Ilyocypris gibba* (○) *Ilyocypris cf. gibba* (+) and *Ilyocypris sp.* (X).

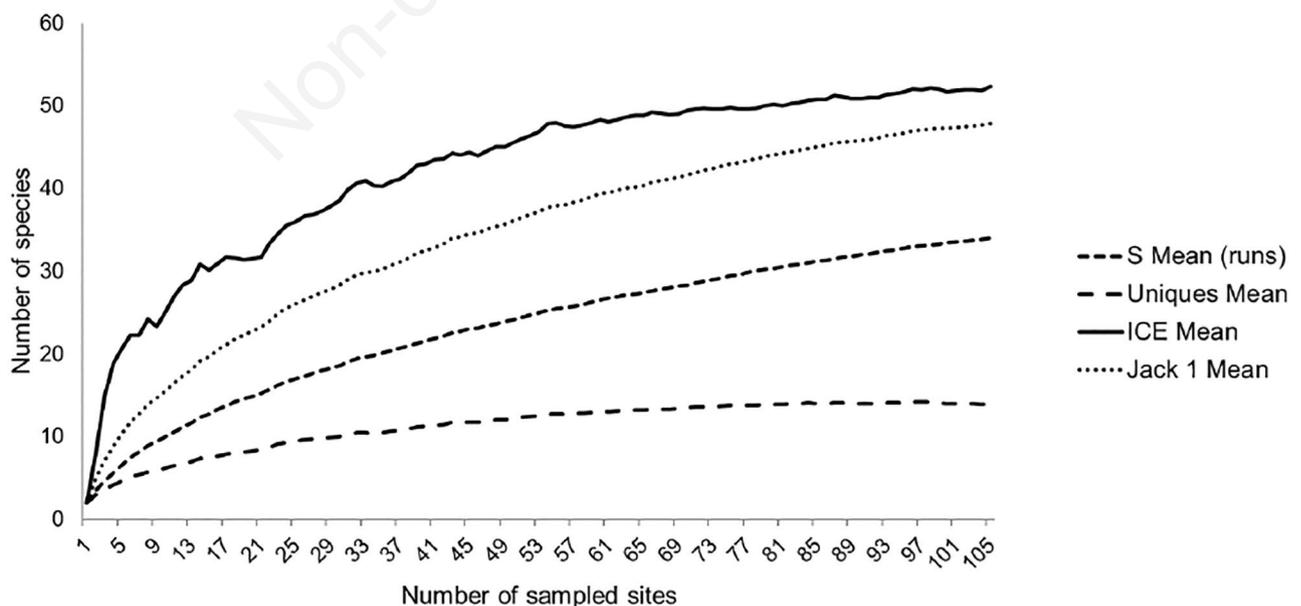
fully shed light on their taxonomic position. For some ostracod species identified in this study, distributional gradients associated with different climatic conditions were observed. The presence of a large part of the ostracod diversity in the Csa climate zone may be due to less harsh environmental conditions, but it should also be taken into account that most of the samples were collected there. As for the occurrence of *Potamocypris smaragdina* and *Potamocypris variegata* in a single climatic zone (Bsh and Bwk, respectively), the finding of these species in a single site each prevents to draw sound conclusions about their habitat preferences. Gauthier (1928c) had already reported a progressive rarefaction of the ostracod fauna diversity in Tunisia passing from the rainy area (average precipitation  $>500 \text{ mm y}^{-1}$ ), to the sub-steppic zone (between 300- and  $500\text{-mm y}^{-1}$ ) and finally to the steppic zone ( $<300 \text{ mm y}^{-1}$ ). The species present in all three areas were *Eucypris virens*, *Ilyocypris getica* and *Tonnacypris lutaria*.

The relatively low number (26) of samples in which ostracods were present and the poor sampling coverage in the driest part of Tunisia in the Gauthier's study (1928c) probably account for the limited concordance with our data on ostracod distribution in relation to climatic conditions. The 15 sampling sites selected by Zaibi *et al.* (2013) for a survey conducted in November 2010 did not include the rainiest part of the country, and lotic environments were mainly sampled, making a comparison with our data difficult. On the other hand, the results in Zaibi *et al.* (2013) show that Tunisian running waters host a rich ostracod diversity and species that are conceivably absent,

or much rarer, in standing waters, such as *Darwinula stevensoni* and *Psychrodromus tunisicus*.

The only non-native ostracod species found in Tunisia is *Candonocypris novaezelandiae* (Zaibi *et al.*, 2013), first described for New Zealand and subsequently reported from different biogeographic areas (Scharf *et al.*, 2014). *Isocypris beauchampi*, a species of Afrotropical origin first reported for Tunisia in this study, has been also found in Canada, South America and several European countries (Meisch, 2000). According to the latter author, although *Isocypris beauchampi* is unrecorded from subsaharian Africa where the genus *Isocypris* is represented by several species, it has probably been introduced in Europe from this same area by migrating birds. Therefore its occurrence in Northern Africa could be seen as the natural expansion of the species' range northward. Of particular biogeographic interest is the occurrence of two ostracod taxa. *Leucocythere cf. algeriensis*, recorded from a single site (F071, a temporary marsh) in the study area; *Leucocythere algeriensis* is currently known for its type locality only in southern Algeria (Danielopol *et al.*, 1990). *Ilyocypris* sp. from F198 shows affinities with *Ilyocypris biplicata anomala* described by Gauthier (1938) from North Africa. Although other related, possibly conspecific, specimens have been found in Italy (Mazzini *et al.*, 2014), Greece (unpublished data), and in several Spanish localities (Escrivà *et al.*, 2009; 2010; Schmit *et al.*, 2013b; Martínez-García *et al.*, 2019), its taxonomic position is still somewhat uncertain.

The sample-based rarefaction curve of mean species richness based on the collected data did not reach a



**Fig. 6.** Species accumulation curves (S Mean) and performance of estimators of ostracod species richness (ICE Mean, Jack 1 Mean) and of uniques (Uniques Mean). Number of sampled sites is on the x-axis, cumulative species richness is on the y-axis.

plateau, thus confirming that the species richness recorded is below the real ostracod diversity occurring in Tunisian inland waters. The trend of unique means is stable above 14 species; in this context, increasing sampling effort did not allow to increase species richness per site, maybe indicating a low sampling efficiency for single sites as a possible cause of the low overall non-exhaustiveness. The ICE and Jack1 means show an expected overall species richness between 48 and 52 species in the study area; these values are slightly higher than the cumulative species richness obtained by integrating all the ostracod occurrence data published to date for the country (Tab. 2), *i.e.*, at least 42 species. Such results further stress the need for carrying out further sampling surveys that take into account the heterogeneity of Tunisian surface waters both at a regional scale and at a local (single site) scale.

To date, except for Algeria and Tunisia, few studies have been published on non-marine ostracods from Maghreb. The scant available information from other parts of north-western Africa derives from occasional surveys and often from dated literature. For example, Ghigi (1932), Masi (1932) and Ramdani (1982) collected material and described new ostracod species from Morocco, and Klie (1943) described new species from Morocco and Mauritania. The only meaningful comparison that can be carried out is between Tunisian and North Algerian (Danielopol *et al.*, 1990; Ghaouaci *et al.*, 2017) ostracod faunas, being both relatively well known and resulting from commensurate sampling effort in a similar latitudinal range. Based on currently available data, the number of recorded genera is slightly higher in Tunisia than in Algeria (29 vs 25 genera), of which 21 are shared by the two countries; *Martenscypridopsis*, *Notodromas*, *Physocypria* and *Prionocypris* were found only in Algeria, whilst *Bradleystrandesia*, *Candona*, *Cytherois*, *Cytheromorpha*, *Darwinula*, *Leptocythere*, *Psychrodromus* and *Vestalenula* exclusively occurred in Tunisia. Six families were recorded in both countries (Candonidae, Cyprididae, Cytherideidae, Ilyocyprididae, Limnocytheridae, Loxoconchidae), Notodromadidae only in Algeria, and Darwinulidae, Leptocytheridae, Paradoxosomatidae only in Tunisia.

Tunisia is separated from Sicily and surrounding small islands, *i.e.* the southernmost part of Italy, only by a narrow sea strait (the distance between the coastlines of Tunisia and mainland Sicily is about 150 km, and those from Tunisia and Pantelleria and Pelagie Islands about 70 and 110 km, respectively). The non-marine ostracod faunas of these two areas show many similarities, but also some differences in their generic composition: 22 genera are in common, eight genera are exclusively found in Tunisia (*Bradleystrandesia*, *Bradleycypris*, *Candona*, *Cytherois*, *Cytheromorpha*, *Leptocythere*, *Leucocythere* and *Loxoconcha*) and seven only in Sicily (*Fabaeformiscandona*, *Mixtacandona*, *Notodromas*, *Physocypria*, *Prionocypris*,

*Pseudocandona* and *Tyrrhenocythere*) (Pieri *et al.*, 2020; Mazzini *et al.*, 2017). In both areas, typically temperate ostracods, possibly except for few isolated records (*e.g.*, *Neglecandona neglecta* in M034 in Tunisia), are absent even in the most humid and coolest parts of the investigated regions. A likely reason for this can be found in the scarcity of deep lakes and, in general, of permanent lentic waters. A similar pattern, as well as the close zoogeographic affinities with adjacent faunas of the Maghreb, were observed for Tunisian “large branchiopods” (Marrone *et al.*, 2016) and diaptomid copepods (Marrone *et al.*, 2017).

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## CONCLUSIONS

Although likely not exhaustive, this study is the most comprehensive survey ever conducted on non-marine ostracods of Tunisia. It contributes to update and significantly increase the knowledge on the diversity and distribution of this ecologically important invertebrate group in Tunisia and, more broadly, in the Maghreb. The obtained results show the presence of one of the most diversified ostracod fauna among North African countries. The number of taxa reported here for Tunisia is certainly underestimated, since in our study most of the sites were visited only once, and relatively few permanent or lotic aquatic habitat were sampled. In addition, a greater taxonomic detail is desirable, also using molecular techniques for those taxa whose identification is not conclusive when based on a morphological approach only. We hope that the newly collected data presented here will provide a reference for further comparative faunal studies aimed at investigating the distribution, affinities and origins of the circum-Mediterranean and north Africa inland water ostracod faunas.

Our results confirm the key role of temporary habitats and, more broadly, of marginal aquatic systems for biodiversity conservation in arid and semi-arid circum-Mediterranean areas. It is therefore essential to implement conservation measures for these systems that are seriously threatened by different types of impact, such as urban sprawl, the transformation of natural areas into cultivated land, and the effects of climate change (Zacharias and Zamparas, 2010).

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**Tab. 1.** List and main characteristics of sampling sites considered in this study.

Code	Date	Latitude	Longitude	CZ	Habitat type	H	Cond	Temp	Turb	MC
F001	28/12/2004	36.815549	10.293178	Csa	Roadside ditch	T	3500	7.3	4	0
F002	28/12/2004	36.813608	10.290118	Csa	Roadside ditch	T	>>3990	7.9	0	1
F003	28/12/2004	36.886894	10.299812	Csa	Flooded cultivated field	T	650	11.0	0	2
F004	28/12/2004	36.894066	10.291864	Csa	Waterbody in flat depression with salt crust	T	>>3990	10.0	0	2
F006	28/12/2004	36.882489	10.300410	Csa	Marsh	T	1880	13.1	0	3
F008	29/12/2004	37.031263	9.995808	Csa	Marsh	T	1210	12.0	0	2
F009	29/12/2004	37.148530	9.999483	Csa	Pond	T	3050	12.7	0	2
F011	03/02/2006	37.165999	9.760281	Csa	Pond	T	770	18.9	2	1
F016b	02/03/2006	34.821888	11.257612	Bsh	Pool	T	16850	19.3	0	0
F017	02/01/2005	34.823366	11.258134	Bsh	Marsh	T	3300	14.0	0	0
F021	03/01/2005	33.822781	9.548813	Bwh	Pool in a streambed	T	>>3990	17.1	0	0
F022	03/01/2005	33.744458	9.251739	Bwh	Marsh with slowly flowing water	T	>>3990	n.d.	0	3
F024	03/01/2005	33.461331	8.932915	Bwh	Spring	?	n.d.	n.d.	n.d.	n.d.
F032	05/01/2005 05/03/2006	35.415363	9.542656	Bwh	Marsh	T	430 522	11.0 16.8	2 4	1 0
F033	06/01/2005	35.788975	10.143312	Bwh	Pond	T	>>3990	10.3	0	2
F034	06/01/2005	35.753299	10.116522	Bwh	Roadside ditch	T	>>3990	14.0	0	2
F036	06/01/2005	36.422897	10.089013	Csa	Pond	T	n.a.	n.a.	n.a.	n.a.
F037	28/01/2006	36.954036	10.231609	Csa	Pond	T	n.d.	n.d.	n.d.	n.d.
F038	15.01.2006	36.817887	10.567251	Csa	n.d.	T	n.d.	n.d.	n.d.	n.d.
F040	23/01/2006	36.955043	10.227447	Csa	Roadside ditch	T	1197	11.3	0	2
F043	23/01/2006	36.926615	10.269296	Csa	Waterbody in flat depression with salt crust	T	47000	11.0	0	0
F047	28/01/2006	36.995056	10.158451	Csa	Pond	T	2080	9.3	3	0
F049	01/02/2006	36.783188	10.163131	Csa	Waterbody in flat depression with salt crust	T	16800	14.5	2	0
F050	01/02/2006	36.818229	10.293716	Csa	Waterbody in flat depression with salt crust	T	35200	15.9	0	1
F052	01/02/2006	36.816994	10.294604	Csa	Waterbody in flat depression with salt crust	T	32600	16.5	3	0
F053	03/02/2006	37.106561	10.025851	Csa	Marsh	P	2390	13.3	0	4
F059	03/02/2006	36.976907	10.048230	Csa	Marsh	T	7500	16.4	2	2
F060	07/02/2006	36.711302	10.180207	Csa	Marsh	T	5560	11.0	2	0
F061	15/02/2006	36.716664	10.184006	Csa	Marsh	T	2870	10.7	0	3
F063	09/02/2006	36.735240	10.264861	Csa	Roadside ditch	T	188	17.7	2	2
F064	09/02/2006	36.690177	10.493709	Csa	Canal	T	3710	15.2	0	2
F065	09/02/2006	36.690975	10.504716	Csa	Marsh	T	385	15.6	2	0
F066	09/02/2006	36.690729	10.504991	Csa	Roadside ditch	T	547	17.0	0	2
F067	09/02/2006	36.699492	10.461404	Csa	Marsh	T	11540	16.1	0	3
F071	13/02/2006	35.796014	10.146367	Bsh	Marsh	T	2480	11.0	1	1
F072	13/02/2006	35.765742	9.982058	Bwh	Pool in a streambed	T	614	11.7	3	0
F073	13/02/2006	36.041103	10.058864	Bsh	Pool	T	347	11.2	0	1
F074	13/02/2006	36.040990	10.058329	Bsh	Pool	T	230	11.2	0	0
F076	15/02/2006	36.791206	10.274467	Csa	Pool	T	1380	12.3	0	2
F077	15/02/2006	36.786548	10.275925	Csa	Marsh	T	9000	14.3	0	3
F078	15/02/2006	36.781577	10.273724	Csa	Pool	T	1374	14.2	0	2
F079	15/02/2006	36.769772	10.263266	Csa	Saltern	T	47800	12.4	1	0
F080	15/02/2006	36.769772	10.263266	Csa	Saltern	T	31500	14.1	1	0
F081	15/02/2006	36.769453	10.264426	Csa	Pool	T	7700	16.7	0	3
F084	15/02/2006 24/02/2006 07/02/2007	36.956236	10.220210	Csa	Marsh	T	1250 n.d. 813	15.0 n.d. 19.9	3 n.d. 3	1 n.d. 2

*To be continued on next page*

**Tab. 1.** Continued from previous page.

Code	Date	Latitude	Longitude	CZ	Habitat type	H	Cond	Temp	Turb	MC
F085	15/02/2006	36.970442	10.214045	Csa	Pool	T	6020	16.0	0	0
F088	25/02/2006	36.674855	10.492831	Csa	Flooded field	T	5290	13.5	3	1
F091	25/02/2006	36.677916	10.695688	Csa	Flooded field	T	588	13.3	4	0
F092	25/02/2006	36.718888	10.727949	Csa	Marsh	T	1380	15.7	1	2
F093	25/02/2006	36.728837	10.790902	Csa	Pool in a streambed	T	221	13.4	4	0
F094	25/02/2006	36.729347	10.790545	Csa	Pool in a streambed	T	n.d.	n.d.	4	0
F096	25/02/2006	36.784799	10.894614	Csa	Marsh	T	n.d.	n.d.	4	0
F101	26/02/2006	37.043707	11.009341	Csa	Pool	T	415	16.5	4	0
F103	26/02/2006	36.900072	10.849900	Csa	Marsh	T	939	13.8	0	3
F104	26/02/2006	36.728885	10.931882	Csa	Marsh	T	2890	19.5	2	2
F105	26/02/2006	36.654575	10.914250	Csa	Waterbody in flat depression with salt crust	T	39500	13.5	4	0
F106	28/02/2006	36.963133	10.078471	Csa	Roadside ditch	T	350	12.2	4	0
F107	28/02/2006	36.994050	10.061590	Csa	Pond	P	3580	13.5	0	3
F108	28/02/2006	37.111015	9.707563	Csa	Pool	T	808	17.7	4	0
F109	28/02/2006	36.962526	9.933141	Csa	Flooded cultivated field	T	2070	15.3	2	2
F110	28/02/2006	36.961469	9.930017	Csa	Pond	P	6060	13.2	0	1
F112	01/03/2006	35.166092	10.727158	Bsh	Waterbody in flat depression with salt crust	T	38000	19.5	0	0
F114	02/03/2006	34.740341	11.180175	Bsh	Waterbody in flat depression with salt crust	T	59500	17.1	0	0
F115	02/03/2006	34.692374	11.168808	Bsh	Waterbody in flat depression with salt crust	T	29500	21.8	3	0
F119	02/03/2006	34.386720	10.252606	Bsh	Waterbody in flat depression with salt crust	T	55000	16.5	0	0
F122	03/03/2006	33.598119	10.538237	Bwh	Pool in a streambed	T	34500	17.7	0	0
F123	03/03/2006	33.596193	10.542966	Bwh	Waterbody in flat depression with salt crust	T	53600	17.6	0	0
F124	03/03/2006	33.760047	10.748290	Bsh	Well	T	467	14.4	2	0
F129	04/03/2006	34.146010	9.160040	Bwh	Well	P	5620	19.8	0	0
F131	05/03/2006	34.319596	8.935240	Bwh	Pond	T	7240	13.8	0	0
F135	05/03/2006	35.272362	9.257520	Bwk	Reservoir	P	n.d.	n.d.	3	0
F136	05/03/2006	35.303194	9.306290	Bwk	Pool	T	420	19.0	2	2
F138	05/03/2006	35.559912	9.756026	Bwh	Reservoir	P	2360	17.5	0	0
F139	05/03/2006	35.562475	9.764359	Bwh	Pool	T	309	15.8	2	2
F146	07/03/2006	35.801877	9.796893	Bsh	Stream	P	907	16.8	0	4
F147	07/03/2006	35.748336	9.939861	Bwh	Marsh	T	1410	10.2	3	2
F153	08/03/2006	35.638288	9.716648	Bsk	Reservoir	T	216	10.2	2	0
F154	08/03/2006	35.722778	9.619487	Bsk	Reservoir	T	1241	9.4	1	1
F160	09/03/2006	36.641052	8.697033	Csa	Roadside ditch	T	506	10.9	1	2
F164	09/03/2006	36.743350	8.679090	Csa	Pool	T	230	16.2	3	2
F166	30/03/2006	36.725424	8.704644	Csa	Pond	T	280	20.2	0	0
F168	09/03/2006	36.898059	8.745653	Csa	Pond	T	274	17.7	3	3
F170	10/03/2006	36.931136	8.755627	Csa	Marsh	T	568	12.8	0	4
F171	10/03/2006	36.931245	8.756221	Csa	Marsh	T	n.d.	n.d.	0	2
F185	10/03/2006	37.055436	9.623718	Csa	Pool	T	1910	15.9	3	0
F195	30/03/2006	36.652830	9.767422	Csa	Waterbody in flat depression with salt crust	T	23700	23.6	0	1
F196	30/03/2006	36.696022	9.417019	Csa	Roadside ditch	T	479	24.2	1	1
F198	30/03/2006	36.557274	8.755233	Csa	Roadside ditch	T	480	26.8	3	2
F201	03/03/2006	33.215326	11.162801	Bwh	Waterbody in flat depression with salt crust	T	53600	17.6	0	0
F203	21/04/2006	32.867673	10.237606	Bwh	Concrete reservoir	T	1125	25.9	4	0
F204	22/04/2006	33.813802	8.733384	Bwh	Pool	T	9510	16.1	0	1
F206	22/04/2006	35.544959	8.684431	Bsk	Reservoir	P	391	15.7	2	2

*To be continued on next page*

**Tab. 1.** Continued from previous page.

Code	Date	Latitude	Longitude	CZ	Habitat type	H	Cond	Temp	Turb	MC
F208	28/12/2004	36.883036	10.302282	Csa	Pool	T	n.d.	n.d.	2	1
F209	26/07/2002	34.376482	7.912122	Bwh	Lateral pools along river margins	P	n.d.	n.d.	1	2
F210	31/12/2007	37.119058	9.673126	Csa	Pool	T	133	15.0	2	1
F231	01/01/2008	37.110325	9.697821	Csa	Pool	T	400	14.8	2	2
F232	01/01/2008	37.206117	9.520817	Csa	Pool	T	270	16.2	4	2
F233	31/12/2007	37.138077	9.375803	Csa	Roadside ditch	T	297	14.0	2	2
F234	31/12/2007	37.138865	9.338640	Csa	Marsh	T	348	16.8	4	1
F235	31/12/2007	36.967102	8.846171	Csa	Marsh	T	960	12.5	0	4
F248	02/01/2012	37.086655	9.199651	Csa	Marsh	T	360	13.1	4	1
F249	02/01/2012	36.936295	8.761124	Csa	Marsh	T	286	12.0	0	3
M029	29/06/2004	37.154742	9.761400	Csa	Roadside ditch	T	n.d.	n.d.	n.d.	n.d.
M034	SRL	37.201350	9.258189	Csa	Pool	T	n.a.	n.a.	n.a.	n.a.
M053	SRL	35.484819	10.643233	Bsh	Pool in a streambed	T	n.a.	n.a.	n.a.	n.a.

CZ, climate zone as in Fig. 1; H, hydroperiod (T = temporary, P = permanent); Cond, electric conductivity at 20°C ( $\mu\text{S cm}^{-1}$ ); Temp, water temperature (°C); Turb, turbidity (see text); MC, macrophyte coverage (see text); SRL, sediment re-hydrated in the laboratory; n.d., not determined; n.a., not available.

**Tab. 2.** Updated checklist of Recent non-marine ostracod taxa from Tunisia based on Zaibi *et al.* (2013) and references therein, Scharf *et al.* (2014), and present study. For the taxa found in the present study (marked with an asterisk), the sites in which they occurred are also reported (codes as in Tab. 1). New species and genera for the Tunisian fauna are reported in bold. *Cythere* sp. listed in Zaibi *et al.* (2013) was omitted because it is regarded here as a marine taxon.

Class Ostracoda Latreille, 1802
Subclass Podocopa G.O. Sars, 1866
Order Podocopida G.O. Sars, 1866
Suborder Cypridocopina Baird, 1845
Superfamily Cypridoidea Baird, 1845
Family Cyprididae Baird, 1845
Subfamily Cypricerinae McKenzie, 1971
Tribe Bradleystrandesini
Genus <i>Bradleystrandesia</i> Broodbakker, 1983
* <i>Bradleystrandesia</i> sp.
F171
Tribe Cypricerini McKenzie, 1971
Genus <i>Bradleycypris</i> McKenzie, 1984
* <i>Bradleycypris obliqua</i> (Brady, 1868)
F235
Subfamily Cypridinae Baird, 1845
Genus <i>Cypris</i> O.F. Müller, 1776
* <i>Cypris bispinosa</i> Lucas, 1849
F036, F096, F170, F171, F235
* <i>Cypris pubera</i> O.F. Müller, 1776
F084
Subfamily Cypridopsinae Kaufmann, 1900
Genus <i>Cypridopsis</i> Brady, 1867
* <i>Cypridopsis</i> cf. <i>elongata</i> (Kaufmann, 1900)
F232
* <i>Cypridopsis hartwigi</i> G. W. Müller, 1900
F103, F170
* <i>Cypridopsis vidua</i> (O.F. Müller, 1776)
F138, F170
* <i>Cypridopsis</i> sp.
F231, F235
Genus <i>Plesiocypridopsis</i> Rome, 1965
* <i>Plesiocypridopsis newtoni</i> (Brady and Robertson, 1870)
F047, F053, F085
Genus <i>Potamocypris</i> Brady, 1870
* <i>Potamocypris arcuata</i> (Sars, 1903)
F104, F232

To be continued on next page

**Tab. 2.** Continued from previous page.

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	* <i>Potamocypris smaragdina</i> (Vávra, 1891)	F073
	* <i>Potamocypris variegata</i> (Brady & Norman, 1889)	F135
	Genus <i>Sarscypridopsis</i> McKenzie, 1977	
	* <i>Sarscypridopsis aculeata</i> (Costa, 1847)	F004, F033, F053, F059, F060, F061, F067, F071, F076, F077, F078, F084, F107, F110, M034
Subfamily	Cyprinotinae Bronshtein, 1947	
	Genus <i>Heterocypris</i> Claus, 1892	
	* <i>Heterocypris barbara</i> (Gauthier and Brehm, 1928)	F004, F008, F016b, F017, F032, F032, F037, F038, F047, F060, F064, F077, F078, F081, F084, F088, F094, F106, F107, F112, F114, F115, F119, F122, F123, F124, F131, F153, F201, F233
	<i>Heterocypris exigua</i> (Gauthier and Brehm, 1928)	
	* <i>Heterocypris incongruens</i> (Ramdohr, 1808)	F001, F002, F022, F033, F034, F053, F061, F063, F066, F071, F074, F078, F104, F129, F146, F196, F203, F208, F210, F233, M029, M053
	* <i>Heterocypris reptans</i> (Kaufmann, 1900)	F009
	* <i>Heterocypris salina</i> (Brady, 1868)	F021, F022, F024, F034, F049, F067, F081, F195
Subfamily	Eucypridinae Bronshtein, 1947	
	Genus <i>Eucypris</i> Vávra, 1891	
	* <i>Eucypris mareotica</i> (Fischer, 1855)	F004, F043, F050, F052, F079, F080, F105, F123
	* <i>Eucypris virens</i> complex (Jurine, 1820)	F001, F003, F004, F006, F008, F009, F011, F032, F032, F037, F040, F047, F053, F059, F060, F061, F063, F064, F065, F066, F067, F071, F072, F073, F074, F076, F077, F078, F081, F084, F084, F085, F088, F091, F092, F096, F101, F103, F104, F106, F107, F108, F109, F110, F136, F139, F147, F160, F164, F166, F168, F171, F185, F196, F198, F210, F231, F232, F233, F234, F248, F249, M034
	Genus <i>Tonnacypris</i> Diebel and Pietrzeniuk, 1975	
	* <i>Tonnacypris lutaria</i> (Koch, 1838)	F074, F136, F139, F210
	Genus <i>Trajancypris</i> Martens, 1989	
	* <i>Trajancypris clavata</i> (Baird, 1838)	F135, F153, F154, F206
Subfamily	Herpetocypridinae Kaufmann, 1900	
	Tribe Herpetocypridini Kaufmann, 1900	
	Genus <i>Candonocypris</i> Sars, 1894	
	<i>Candonocypris novaezelandiae</i> (Baird, 1843)	
	Genus <i>Herpetocypris</i> Brady and Norman, 1889	
	<i>Herpetocypris chevreuxi</i> (Sars, 1896)	
	<i>Herpetocypris</i> sp.	F112
	Tribe Psychrodromini Martens, 2001	
	Genus <i>Psychrodromus</i> Danielopol and McKenzie, 1977	
	<i>Psychrodromus tunisicus</i> Zaibi <i>et al.</i> , 2013	
Subfamily	Isocypridinae Hartmann and Puri, 1974	
	Genus <i>Isocypris</i> G.W. Müller, 1908	
	* <i>Isocypris beauchampi</i> (Paris, 1920)	F231, F232
Family	Candonidae Kaufmann, 1900	
	Subfamily Candoninae Kaufmann, 1900	
	Tribe Candonini Kaufmann, 1900	
	Genus <i>Candona</i> Baird, 1845	
	* <i>Candona</i> cf. <i>muelleri</i> Hartwig, 1899	F234
	<i>Candona</i> spp.	
	Genus <i>Neglecandona</i> Krstić, 2006	
	* <i>Neglecandona neglecta</i> (Sars, 1887)	M034
Subfamily	Cycloocypridinae Kaufmann, 1900	
	Genus <i>Cycloocypris</i> Brady and Norman, 1889	
	* <i>Cycloocypris laevis</i> (O.F. Müller, 1776)	F234
	Genus <i>Cypria</i> Zenker, 1854	
	* <i>Cypria ophthalmica</i> (Jurine, 1820)	F124, F129

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**Tab. 2.** Continued from previous page.

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Family Ilyocyprididae Kaufmann, 1900
Subfamily Ilyocypridinae Kaufmann, 1900
Genus <i>Ilyocypris</i> Brady and Norman, 1889
* <i>Ilyocypris bradyi</i> Sars, 1890
F209
* <i>Ilyocypris getica</i> Masi, 1906
F032, F047, F072, F092, F136, F206, F210
* <i>Ilyocypris</i> cf. <i>getica</i> Masi, 1906
F091
* <i>Ilyocypris gibba</i> (Ramdohr, 1808)
F021, F081
* <i>Ilyocypris</i> cf. <i>gibba</i> (Ramdohr, 1808)
F168
* <i>Ilyocypris</i> spp.
F008, F059, F093, F147, F171, F198
Superfamily Darwinuloidea Brady and Robertson, 1885
Family Darwinulidae Brady and Robertson, 1885
Genus <i>Darwinula</i> Brady and Robertson, 1885
<i>Darwinula stevensoni</i> (Brady and Robertson, 1870)
Genus <i>Vestalenula</i> Rossetti and Martens, 1998
<i>Vestalenula</i> sp. B (Danielopol, 1980)
Superfamily Cytheroidea Baird, 1850
Family Cytherideidae Sars, 1925
Subfamily Cytherideinae
Genus <i>Cyprideis</i> Jones, 1857
<i>Cyprideis torosa</i> (Jones, 1850)
<i>Cyprideis</i> sp.
Family Leptocytheridae Sars, 1925
Genus <i>Leptocythere</i> Sars, 1925
<i>Leptocythere castanea</i> (Sars, 1866)
Family Limnocytheridae Sars, 1925
Subfamily Limnocytherinae Sars, 1925
Tribe Leucocytherini Danielopol <i>et al.</i> , 1990
Genus <i>Leucocythere</i> Kaufmann, 1900
* <i>Leucocythere</i> cf. <i>algeriensis</i> Martens, 1990
F071
Tribe Limnocytherini Klie, 1938
Genus <i>Limnocythere</i> Brady, 1867
<i>Limnocythere</i> sp.
Family Loxoconchidae Sars, 1925
Genus <i>Cytheromorpha</i> Hirschmann, 1909
<i>Cytheromorpha fuscata</i> (Brady, 1869)
Genus <i>Loxoconcha</i> Sars, 1866
<i>Loxoconcha elliptica</i> Brady, 1868
Family Paradoxostomatidae Brady & Norman, 1989
Genus <i>Cytherois</i> G.W. Müller, 1884
<i>Cytherois fischeri</i> (Sars, 1866)

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