# Water quality affects the structure of copepod assemblages along the Sfax southern coast (Tunisia, southern Mediterranean Sea)

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**Abstract.** The Sfax southern coast (Gulf of Gabes, Mediterranean Sea) has been under increased anthropogenic pressure for many years. In the present study we investigated the effects of this anthropisation on the spatial distribution of copepod assemblages in relation to the physicochemical features of seawater at 20 stations sampled on 19 March 2013. Copepods represented 73% of total zooplankton abundance. Small planktonic copepods (<1.45 mm), including pollution-tolerant species (e.g. *Oithona nana*, *Paracalanus parvus*, *Harpacticus littoralis* and *Tisbe battagliai*), proliferated exclusively in stations of ~0.5-m depth characterised by high coastal anthropogenic inputs. The largest copepod species were dominated by *Calanus helgolandicus* (1.45–2.5 mm) in the offshore zone in depths of ~3 m. Substantial numbers of *Oithona plumifera* (7.5%) were found at depths between 0.5 and 3 m. Copepod diversity was significantly higher in the southern zone, which is less affected by sewage, than in the northern zone, which was subjected to higher pressure (Shannon–Wiener index $H' = 1.5$–$2.5$ and $\leq 1.5$ bits individual$^{-1}$). A shift in the planktonic copepod community between the two zones was linked to deterioration of water quality, with higher phosphorus levels, turbidity and chemical oxygen demand (COD) in the northern zone.

**Additional keywords:** anthropogenic inputs, chemical oxygen demand–5-day biochemical oxygen demand, COD–$BOD_5$, diversity, zooplankton.

## Introduction

Plankton represents the base of the marine food web and thus plays a pivotal role in fisheries. Primary productivity and plankton growth are closely related to the physicochemical parameters of seawater (i.e. light, nutrients and oxygen; Shastri 2000; Gang *et al.* 2006; Khwaja *et al.* 2014). Zooplankton is an essential component of the aquatic ecosystem. It is the faunal component that occupies the primary consumer level and forms a link between microscopic photosynthetic algae and fish (Madin *et al.* 2001; Sundaresan and Kumar 2013). Zooplankton feed on phytoplankton and are, in turn, consumed by fish and macroinvertebrates (Sharma 1998; Biswas 2015). Zooplankton communities depend on the nutrient content of an ecosystem and rapidly respond to changes in nutrient content (Dodson 1992). Their diversity and abundance are very sensitive to any changes in physicochemical conditions, thus providing information about the health of an aquatic system and of any damage and threat to the ecosystem (Rosenberg and Resh 1993; Biswas 2015; Sanyal *et al.* 2015). Zooplankton is not only a pertinent bioindicator of pollution load, but can also be used to assess the changing trophic status of an aquatic system (Mukhopadhyay *et al.* 2007; Kushwaha and Agrahari 2014; Sanyal *et al.* 2015).

The Sfax southern coast (southern Mediterranean Sea, Tunisia) has been under increased anthropogenic pressure for many years due to industrial activities, including phosphate treatment, ship activity in commercial and fishing harbors, agricultural activity, including salt marshes and the storage of olive oil waste (margins), and the dumping of considerable volumes of urban waste in the Thyna landfill and municipal wastewater treatment plants (Zaghden *et al.* 2005; Gargouri 2006; Choura *et al.* 2015). Hence, in the southern part of Sfax, many different sources release diverse compounds into marine waters, including

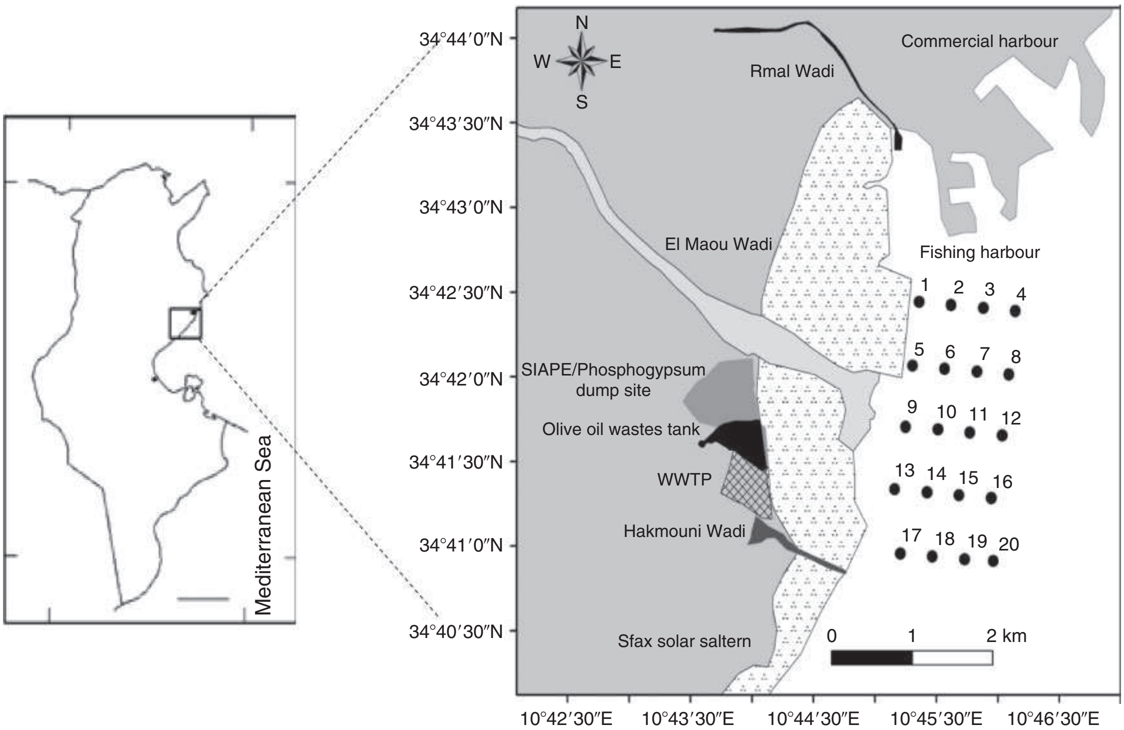


**Fig. 1.** Location of the study stations along the southern coast of Sfax (Tunisia) in the southern Mediterranean Sea, sampled in spring (March 2013).Stations 1–10, northern stations; Stations 11–20, southern stations. SIAPE, phosphoric acid and fertiliser plant; WWTP, wastewater treatment plant (located in front of Station 9). The fishing harbour is located close to Stations 1 and 2.

phosphates, fluorides, sulfates, naturally occurring radionuclides, trace metals and other trace elements, hydrocarbons and polyphenolic and flavonoid compounds (Zaghden *et al.* 2005; Tayibi *et al.* 2009; Mezghani-Chaari *et al.* 2011). These discharges have led to increases in biological oxygen demand (BOD), dissolved oxygen (DO) and suspended particulate matter (SPM), in addition to changes in pH, in the receiving water (Davis and Marshall 1998; Negi and Rajput 2013).

A recent study evaluated the water quality along the Sfax southern coast by analysing physicochemical parameters (temperature, salinity, pH), major anions ($Cl^-$, $HCO_3^-$, $SO_4^{2-}$, $F^-$), major cations ($Ca^{2+}$, $K^+$, $Mg^{2+}$, $Mn^{2+}$, $Fe^{2+}$ and $Fe^{3+}$) and biogeochemical parameters (nutrients, turbidity, SPM, total polyphenolic and flavonoid compounds, chemical oxygen demand (COD), 5-day BOD ($BOD_5$); Drira *et al.* 2016). The Sfax northern coast has been partly restored as part of the Taparura Project, including rehabilitation of the site of a former industrial complex, cleaning up of beaches and restoration of the area (Callaert *et al.* 2009). This project led to significant improvements in plankton communities and water quality (Rekik *et al.* 2013*a*). For this reason, a future project restoring the southern area is deemed necessary to ameliorate the biological quality of the seawater (primarily planktonic aspects).

The aims of the present study were to: (1) investigate the spatial distribution of zooplankton (copepod) assemblages along the Sfax southern coast; and (2) assess relationships between zooplankton composition and the physicochemical parameters of seawater, which were determined in the same samples and published previously (Drira *et al.* 2016), using multivariate analysis. To the best of our knowledge, the present study is the first to evaluate the effects of anthropogenic inputs on the structure and spatial distribution of copepod assemblages in along the southern Sfax coast.

## Materials and methods

### *Study area and sampling*

The study area was along the coastline of Sfax city (34°44′N, 10°46′E), located in the south-east of Tunisia in the northern part of the Gulf of Gabes (southern Mediterranean Sea; Fig. 1). The area is characterised by shallow waters and an important biodiversity with endemic *Posidonia oceanica* seagrass beds, which were replaced by green tides caused by coastal *Ulva rigida* (Ben Brahim *et al.* 2010). Despite its ecological significance, the southern Sfax coast has been user serious threat for many decades from maritime, urban and industrial activities. The major sources of pollution include urban and industrial wastewaters, the fishing harbour and phosphate-processing coproducts (Aloulou *et al.* 2012). The main industries in Sfax are phosphates, chemical products, textiles, olive oil, food, soap and paint (Barhoumi *et al.* 2009).

In the present study, 20 stations were sampled at high tide during a spring cruise conducted on 19 March 2013 along the Sfax southern coast on the vessel *Taparura*. Station depths ranged from 0.3 to 3.5 m (Figs 1, 2*a*). Stations located in the northern part of this area (S1–10) were the most affected by sewage from the El Maou wadi, the phosphate industry and the waste water treatment plant (WWTP), whereas stations located in the southern part of this area (S11–20), bordering the solar

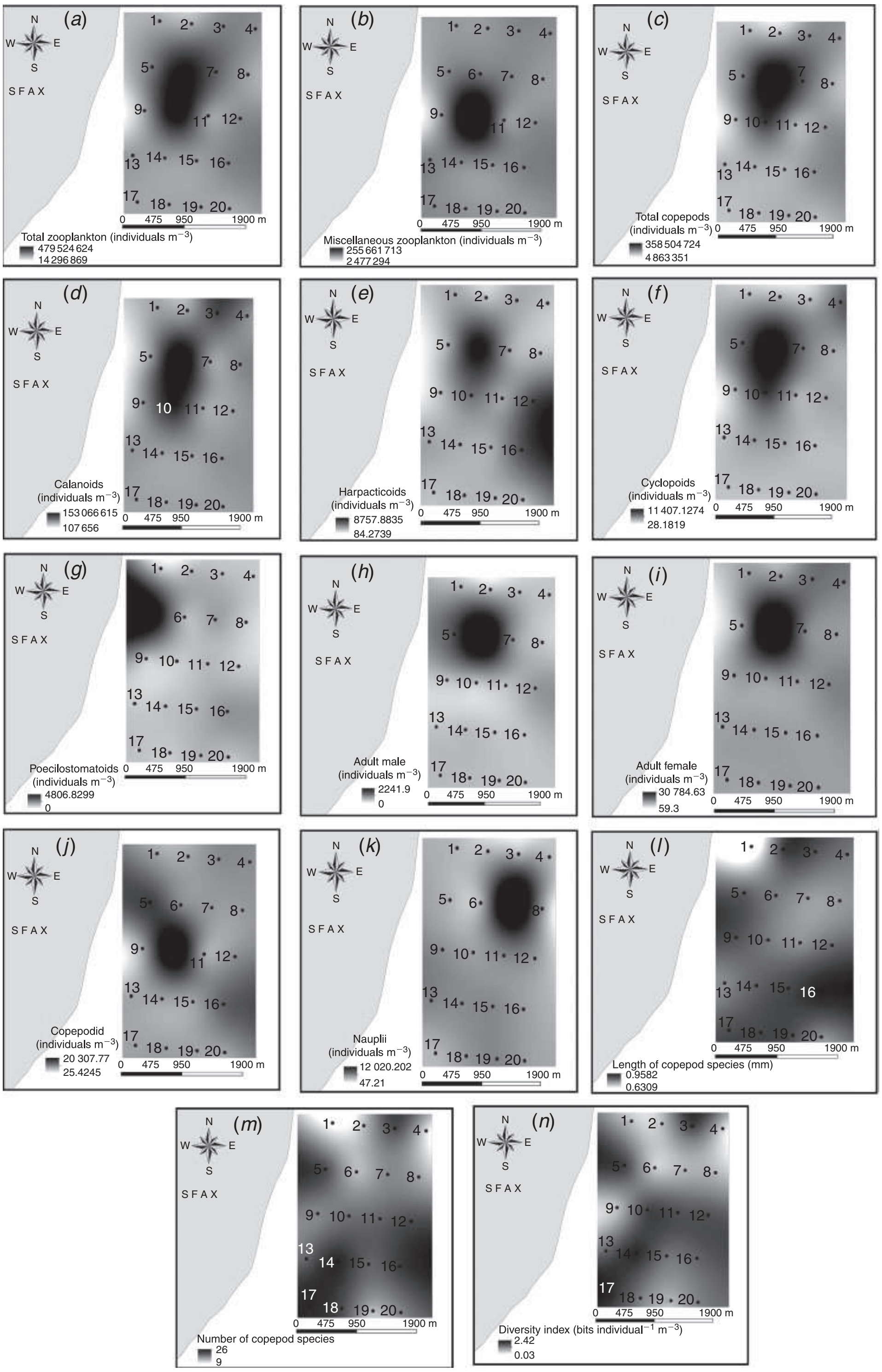


**Fig. 2.** Spatial variation of zooplankton parameters at Stations 1–20 sampled off the southern coast of Sfax in spring (March 2013): abundance of (*a*) total zooplankton, (*b*) miscellaneous zooplankton, (*c*) total copepods, (*d*) calanoids, (*e*) harpacticoids, (*f*) cyclopoids, (*g*) poecilostomatoids, (*h*) adult males, (*i*) adult females, (*j*) copepodids and (*k*) nauplii; (*l*) size of copepod species; (*m*) number of copepod taxa; and (*n*) diversity index.

saltern marsh, were less affected although they did receive inputs from the Hakmouni wadi (Fig. 1).

Zooplankton was collected using a cylindroconical net (aperture 30 cm, height 100 cm, mesh size 100 µm) equipped with a Hydro-Bios flowmeter (mechanical flow meter HB 438 111, Muchtar Tabrani, Bekasi, Indonesia, see https://www.jualo.com/elektronik/iklan-hydro-bios-flow-meter_2, accessed 14 September 2017). The net was towed obliquely from near bottom to the surface at each station at a mean speed of 1 m $s^{-1}$ over a period of 4 min. After collection, zooplankton samples (200 mL) were rapidly preserved in a buffered formaldehyde solution (2%). Samples were subsequently stained with Rose Bengal to identify the internal tissues of the different zooplankton species and to facilitate copepod dissection.

### *Zooplankton analysis*

Zooplankton samples were identified according to Rose (1933), Bradford-Grieve (1999) and Costanzo *et al.* (2000). The different copepod species were sorted into four demographic classes (nauplii, copepodids, adult males and adult females). Miscellaneous zooplankter were also counted according to Tregouboff and Rose (1978*a*, 1978*b*). Enumeration of the entire zooplankton sample was performed under a vertically mounted deep-focus dissecting microscope (TL 2; Olympus Australia Pty Ltd, Melbourne, Vic., Australia) and numerical density was expressed as the number of individuals per cubic metre. The total length of the body of adult copepods was measured for each species in each station sampled (10 individuals for each species in each sample). The body lengths of the different copepod species were measured using an eyepiece micrometer and taking into consideration the several body parts that make up the whole body. The sex ratio (expressed as a percentage) was estimated for each copepod species according to Wiwatanaratanabutr and Grandjean (2016) as follows:

$$\text{number of males} \div (\text{number of males} + \text{number of females}) \times 100$$

### *Data processing and statistical analysis*

Geographic information systems (GIS) tools through ArcGIS, ver. 10.2 (Esri, Redlands, CA, USA), were used to create contour plots. Kriging was the method used to build maps relative to spatial distribution for all dataset parameters. Mesozooplankton diversity was measured using a range of univariate and multivariate diversity measurements. Species were assessed using the Shannon–Wiener index, $H'$ (Shannon and Weaver 1949):

$$H' = -\sum_{ni}^{i=1} \frac{ni}{N} \log_2 \frac{ni}{N}$$

where $n_i$ is the number of individuals of taxon $i$ in the sample and $N$ is the total number of organisms of all species. The dominance index, δ, for copepod species was calculated using to the following formula:

$$\delta = (n_1 + n_2) \div N$$

where δ is the dominance index equal to the percentage contribution of the two most important species ($n_1 + n_2$) of the total standing stock and $N$ is total individual abundance. Unpaired Student's $t$-tests were used to evaluate differences between the northern (S1–10) and southern (S11–20) stations in terms of physical, physicochemical and biogeochemical parameters with a confidence level of 95%. Differences were considered significant at $P < 0.05$.

The spatial variability of copepod communities in relation to environmental variables was assessed using multivariate analysis after $\log_{10}(x + 1)$ data transformation (Sokal and Rohlf 1981). Analyses were conducted using two datasets, the first containing the frequencies of copepod taxa and the second containing the environmental and trophic variables, with the diversity index ($H'$) considered as an independent variable. Factorial correspondence analysis (FCA) and principal component analysis (PCA) were performed on these two datasets respectively. The results of the two analyses were associated through a co-inertia analysis (Dolédec and Chessel 1994). Analyses were performed using Ade-4 software (Laboratory of Biometry, Genetics and Population Biology, University of Lyon 1, Lyon, France) (Thioulouse *et al.* 1997). Pearson's rank correlations were conducted using XLStat 2014 (Thierry Fahmy, University of California—Berkeley, Berkeley, CA, USA) to determine potential correlations between the copepod community and the physicochemical variables.

Data are given as the mean ± s.d.

## Results

### *Spatial distribution of zooplankton according to environmental conditions*

Table 1 summarises the mean values of zooplankton parameters and selected environmental parameters using data from Drira *et al.* (2016) in the northern (S1–10) and southern (S11–20) stations.

Total zooplankton abundance varied from $1.43 \times 10^3$ individuals $m^{-3}$ at S20 to $47.95 \times 10^3$ individuals $m^{-3}$ at S10, with a mean of $10.25 \pm 12.55 \times 10^3$ individuals $m^{-3}$ (Fig. 2*a*; Table 1). Zooplankton assemblages were dominated by copepods, which represented 73% of total zooplankton abundance, whereas the abundance of miscellaneous zooplankton did not exceed 27% (Table 2). The density of non-copepod zooplankton varied from $0.25 \times 10^3$ individuals $m^{-3}$ at S20 to $25.57 \times 10^3$ individuals $m^{-3}$ at S10, with a mean of $3.07 \pm 5.40 \times 10^3$ individuals $m^{-3}$ (Fig. 2*b*; Table 1). Polychaete larvae, cirriped larvae, ostracods, jellyfish, zoea and fish eggs were also permanent components of meroplankton contributing to 81% of the non-copepod abundance. Conversely, appendicularians, cladocerans and foraminifera were permanent components of the holoplankton, but did not exceed 19% of miscellaneous zooplankton abundance (Table 2). Total copepod abundance varied from $0.49 \times 10^3$ individuals $m^{-3}$ at S17 to $35.85 \times 10^3$ individuals $m^{-3}$ at S6, with a mean of $7.18 \pm 8.89 \times 10^3$ individuals $m^{-3}$ (Fig. 2*c*; Table 1).

In all, 27 different copepod species were identified during a single cruise in March 2013 belonging to four different orders: Calanoida, Cyclopoida, Harpacticoida and Poecilostomatoida (Table 2). Calanoida was the most diverse order (12 species), followed by Poecilostomatoida (6 species), Cyclopoida (5 species) and Harpacticoida (4 species), contributing to 39, 10, 29 and 22%

**Table 1. Summary of zooplankton parameters (present study) and environmental variables (from Drira *et al.* 2016) along the Sfax southern coast in spring (March 2013) at the northern and southern stations**

Data are the mean ± s.d. The last column shows results of Student's *t*-tests for comparisons between northern (Stations 1–10) and southern (Stations 11–20) stations. Asterisks denote significant differences between Northern and Southern stations: *, $P < 0.05$; **, $P < 0.001$; ***, $P < 0.0001$. NTU, nephelometric turbidity units; SPM, suspended particulate matter; COD, chemical oxygen demand; $BOD_5$, 5-day biochemical oxygen demand; Chl-*a*, chlorophyll-*a*

| | Northern stations | Southern stations | $|t|$ (d.f) |
|---|---|---|---|
| Physical and chemical parameters | | | |
| Depth (m) | 1.59 ± 1.06 | 0.97 ± 0.44 | 1.95 (18) |
| Temperature (°C) | 17.9 ± 2.20 | 15.20 ± 0.80 | 4.75 (18)*** |
| Salinity | 37.4 ± 1.00 | 40 | 4.20 (18)*** |
| pH | 7.86 ± 0.14 | 8.39 ± 0.07 | 9.00 (18)*** |
| Biogeochemical parameters | | | |
| $NO_3^-$ (μM) | 6.44 ± 2.70 | 6.56 ± 1.74 | 0.26 (18) |
| $NO_2^-$ (μM) | 0.31 ± 0.11 | 0.32 ± 0.14 | 0.16 (18) |
| $NH_4^+$ (μM) | 3.59 ± 1.71 | 4.25 ± 1.36 | 1.05 (18) |
| Total N (μM) | 21.04 ± 3.57 | 21.22 ± 2.82 | 0.43 (18) |
| $PO_4^{3-}$ (μM) | 12.72 ± 5.19 | 9.65 ± 2.72 | 0.74 (18) |
| Total P (μM) | 32.78 ± 12.86 | 27.73 ± 11.99 | 0.47 (18) |
| $Si(OH)_4$ (μM) | 33.15 ± 16.64 | 28.66 ± 13.90 | 0.04 (18) |
| N/P ratio | 0.99 ± 0.53 | 1.26 ± 0.51 | 0.66 (18) |
| Turbidity (NTU) | 5.47 ± 1.65 | 0.58 ± 0.24 | 9.64 (18)*** |
| SPM (mg $L^{-1}$) | 29.45 ± 6.95 | 29.17 ± 2.57 | 1.04 (18) |
| COD (×$10^3$ mg $L^{-1}$) | 2.13 ± 0.58 | 1.66 ± 0.16 | 1.71 (18)* |
| $BOD_5$ (mg $L^{-1}$) | 65.30 ± 31.90 | 55.30 ± 26.50 | 0.07 (18) |
| $BOD_5$/COD | 0.03 ± 0.01 | 0.03 ± 0.01 | 0.77 (18) |
| COD/$BOD_5$ | 42.26 ± 27.11 | 37.94 ± 19.07 | 0.73 (18) |
| SPM/$BOD_5$ | 0.63 ± 0.47 | 0.67 ± 0.35 | 0.30 (18) |
| Chl-*a* (×$10^{-2}$ μg $L^{-1}$) | 0.64 ± 0.56 | 0.33 ± 0.25 | 1.60 (18) |
| Zooplankton | | | |
| Abundance (×$10^3$ individuals $m^{-3}$) | | | |
| Total zooplankton | 15.40 ± 16.28 | 5.10 ± 2.92 | 1.69 (18) |
| Non-copepod zooplankton | 4.26 ± 7.57 | 1.88 ± 0.97 | 0.60 (18) |
| Total copepods | 11.14 ± 11.23 | 3.21 ± 2.35 | 2.10 (18)* |
| Calanoids | 3.96 ± 5.40 | 0.30 ± 0.36 | 2.00 (18) |
| Harpacticoids | 1.76 ± 2.69 | 1.68 ± 2.01 | 0.62 (18) |
| Cyclopoids | 3.60 ± 3.75 | 0.61 ± 0.64 | 2.49 (18)* |
| Poecilostomatoids | 0.68 ± 1.47 | 0.28 ± 0.32 | 0.21 (18) |
| Adult males | 0.38 ± 0.72 | 0.05 ± 0.09 | 0.65 (18) |
| Adult females | 5.09 ± 9.25 | 0.68 ± 1.08 | 3.11 (18)** |
| Copepodids | 3.95 ± 6.37 | 1.02 ± 1.92 | 1.84 (18) |
| Nauplii | 1.70 ± 3.68 | 1.44 ± 0.61 | 1.84 (18) |
| Length of copepod species (mm) | 0.84 ± 0.08 | 0.88 ± 0.04 | 1.34 (18) |
| Ratio males/adults (%) | 13 ± 30 | 20 ± 36 | 0.46 (18) |
| Shannon index (bits $individual^{-1}$) | 0.75 ± 0.89 | 1.54 ± 0.63 | 1.70 (18)* |
| Number of copepod species | 16.70 ± 4.59 | 21.70 ± 3.70 | 2.59 (18)* |
| Dominance index (%) | 53.54 ± 18.08 | 65.96 ± 14.16 | 2.61 (18)** |

of the total abundance respectively (Table 2). Among the calanoid copepods, *Paracalanus parvus*, *Calanus helgolandicus* and *Paracartia latisetosa* were the most abundant species, accounting for 14, 7.5 and 6.5% of total copepod abundance respectively. Among the cyclopoid copepods, *Oithona nana* and *Oithona plumifera* were the most abundant species, accounting for 14 and 7.5% of total copepod abundance respectively. *Tisbe battagliai*, *Euterpina acutifrons* and *Harpacticus littoralis* were the dominant harpacticoid species and accounted for 13, 9 and 3.5% of total copepod abundance respectively. Among the poecilostomatoid copepods, *Corycaeus clausi* was dominant, accounting for 1.5% of total copepod abundance.

The peak abundance of copepod community recorded at S6 was associated with a high density of calanoids, harpacticoids, cyclopoids, adult males and adult females (Fig. 2*d–f*, *h*, *i*, *k*; Tables 1, 2). However, poecilostomatoid density and the percentage of males reached maximum values at S5 (Fig. 2*g*; Table 1). In all sampled stations, a low percentage of larval-stage (copepodids 35%, nauplii 22%) individuals (Table 2) and a high percentage of adults (43% of total copepod abundance) were recorded. There was a greater number of adult females than males (Table 2), which affected the sex ratio (% males). In fact, the sex ratio did not exceed 20% (Table 1). The Shannon–Wiener index ($H'$) for copepods was low, with values ranging

**Table 2. Taxonomic composition and quantitative aspects of the zooplankton community along the Sfax southern coast in spring (March 2013)**
Data are the mean ± s.d. Stations with maximum abundance are listed for each taxon. RA, relative abundance; TL, total length; FO, frequency of occurrence; SWMSA, station with maximum species abundance

| Zooplankton | Abbreviation | RA (%) | TL (mm) | FO (%) | Density (individuals $m^{-3}$) | SWMSA |
|---|---|---|---|---|---|---|
| Copepods (73%) | | | | | | |
| Small copepods (≤1.45 mm) | | 92.74 | | | | |
| Calanoids | | | | | | |
| *Acartia clausi* (Giesbrecht, 1889) | *A cl* | 0.49 | 1.010 ± 0.001 | 55 | 31.83 | 5 |
| *Acartia longiremis* (Lilljeborg, 1853) | *A lo* | 0.06 | 1.220 ± 0.004 | 35 | 3.99 | 3 |
| *Acartia danae* (Giesbrecht, 1889) | *A da* | 0.05 | 1.120 ± 0.008 | 45 | 3.42 | 3 |
| *Acartia* sp. (Dana, 1846) | *A sp* | 0.15 | 1.060 ± 0.006 | 50 | 9.45 | 14 |
| *Paracalanus parvus* (Claus, 1863) | *Par p* | 14.01 | 1.010 ± 0.005 | 100 | 905.80 | 10 |
| *Paracartia grani* (Sars, 1904) | *Par g* | 0.14 | 1.150 ± 0.007 | 40 | 8.99 | 3 |
| *Paracartia latisetosa* (Kritchagin, 1873) | *Par l* | 6.32 | 1.200 ± 0.007 | 75 | 408.90 | 6 |
| *Temora longicornis* (Müller, 1792) | *Tem l* | 1.20 | 1.020 ± 0.011 | 75 | 77.34 | 3 |
| *Centropages typicus* (Kroyer, 1849) | *Cen t* | 3.09 | 1.400 ± 0.010 | 25 | 200.10 | 6 |
| *Isias clavipes* (Boeck, 1865) | *Isi* | 0.10 | 1.240 ± 0.015 | 50 | 6.22 | 17 |
| Harpacticoids | | | | | | |
| *Clytemnestra scutellata* (Dana, 1847) | *Cly* | 1.34 | 0.600 ± 0.010 | 25 | 86.84 | 10 |
| *Euterpina acutifrons* (Dana, 1847) | *Eut a* | 8.93 | 0.390 ± 0.014 | 65 | 576.97 | 12 |
| *Harpacticus littoralis* (Sars, 1910) | *Har l* | 3.42 | 0.320 ± 0.014 | 75 | 221.45 | 11 |
| *Tisbe battagliai* (Volkmann-Rocco, 1972) | *Tis* | 13.00 | 1.400 ± 0.090 | 95 | 840.12 | 6 |
| Cyclopoids | | | | | | |
| *Oithona brevicornis* (Giesbrecht, 1891) | *Oit br* | 1.06 | 0.650 ± 0.156 | 95 | 68.45 | 4 |
| *Oithona nana* (Giesbrecht, 1892) | *Oit n* | 14.87 | 0.560 ± 0.041 | 95 | 961.05 | 10 |
| *Oithona plumifera* (Baird, 1843) | *Oit pl* | 7.49 | 1.410 ± 0.034 | 95 | 483.79 | 6 |
| *Oithona similis* (Claus, 1866) | *Oit s* | 4.27 | 0.410 ± 0.037 | 90 | 276.30 | 7 |
| *Oithona* sp. (Baird, 1843) | *Oit* sp. | 5.11 | 0.880 ± 0.071 | 100 | 330.68 | 6 |
| Poecilostomatoids | | | | | | |
| *Oncaea mediterranea* (Claus, 1863) | *Onc m* | 0.93 | 0.650 ± 0.003 | 65 | 60.27 | 7 |
| *Oncaea minuta* (Giesbrecht, 1892) | *Onc mi* | 0.23 | 0.620 ± 0.006 | 60 | 14.88 | 3 |
| *Triconia conifera* (Giesbrecht, 1891) | *Tri* | 0.26 | 0.650 ± 0.006 | 70 | 16.87 | 19 |
| *Corycaeus clausi* (Dahl, 1894) | *Cor cl* | 1.14 | 1.210 ± 0.035 | 90 | 73.56 | 16 |
| *Corycaeus latus* (Dana, 1849) | *Cor la* | 0.38 | 0.690 ± 0.007 | 85 | 24.40 | 5 |
| *Corycaeus speciosus* (Dana, 1849) | *Cor sp* | 0.05 | 0.800 ± 0.029 | 90 | 300.20 | 5 |
| Large copepods (1.45–2.5 mm) | | 7.26 | | | | |
| Calanoids | | | | | | |
| *Calanus helgolandicus* (Claus, 1863) | *Cal h* | 7.23 | 1.780 ± 0.047 | 90 | 467.69 | 6 |
| *Temora stylifera* (Dana, 1849) | *Tem s* | 0.03 | 1.490 ± 0.008 | 30 | 1.84 | 11 |
| Miscellaneous zooplankton (27%) | | | | | | |
| Meroplankton | | 81 | | | | |
| Polychaeta larvae | *Pla* | 18.36 | 0.612 ± 0.011 | 95 | 564.34 | 10 |
| Cirripedia larvae | *Cla* | 16.46 | 0.766 ± 0.015 | 90 | 505.69 | 10 |
| Ostracoda | *Ost* | 1.30 | 0.502 ± 0.001 | 15 | 40.05 | 6 |
| Medusa | *Med* | 0.61 | 0.120 ± 0.001 | 50 | 18.61 | 10 |
| Zoea | *Zoe* | 0.50 | 0.402 ± 0.002 | 55 | 15.21 | 5 |
| Fish eggs | *Egg* | 43.64 | 0.510 ± 0.005 | 100 | 1341.06 | 10 |
| Holoplankton | | 19 | | | | |
| Appendicularia | *App* | 16.12 | 1.025 ± 0.004 | 100 | 495.30 | 10 |
| Cladocera | *Cla* | 0.12 | 0.515 ± 0.003 | 35 | 3.75 | 18 |
| Foraminifera | *For* | 2.90 | 0.306 ± 0.006 | 70 | 89.06 | 10 |

between 0.03 bits individual$^{-1}$ (14 species, S6) and 2.42 bits individual$^{-1}$ (26 species, S17; Fig. 2*m*, *n*).

There were significant differences between the groups of stations in the two zones (northern *v.* southern) for several environmental parameters. Water temperature, turbidity and COD were significantly higher in the northern than southern stations, whereas salinity and pH were higher in southern than northern stations. However, there were no significant differences for most zooplankton parameters between the northern and southern stations, except for total copepods, cyclopoids and total adult females, which were more abundant in the northern than southern stations. However, the number of copepod species, the Shannon–Wiener index ($H'$) and the dominance index (δ) were higher in the southern than northern stations (Student's *t*-test, $P < 0.05$; Table 1). There was a tendency for δ to decrease from onshore to offshore stations (Fig. 3).

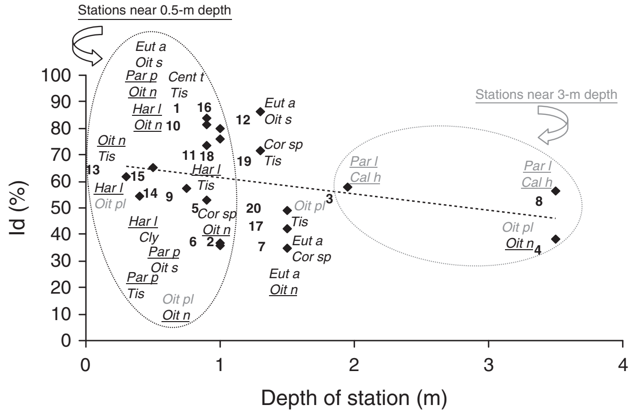


**Fig. 3.** Relationships between the dominance index of the planktonic copepod species and station depth off the southern coast of Sfax in spring (March 2013). *Cal h*, *Calanus helgolandicus*; *Cen t*, *Centropages typicus*; *Cly*, *Clytemnestra scutellata*; *Cor sp*, *Corycaeus speciosus*; *Eut a*, *Euterpina acutifrons*; *Har l*, *Harpacticus littorallis*; *Oit pl*, *Oithona plumifera*; *Oit s*, *Oithona similis*; *Par l*, *Paracartia latisetosa*; *Par p*, *Paracalanus parvus*; *Tis*, *Tisbe battagliai*; *Oit n*, *Oithona nana*. Numbers next to diamonds indicate station numbers, dominant species in 'coastal' areas (stations near 0.5-m depth) are in black and underlined text, in 'intermediate' areas (stations 0.5–3-m depth) are in light grey, in 'offshore' areas (stations near 3-m depth) are in light grey and underlined, and the rest of species are in black text.

The coastal area (stations of ~0.5-m depth; southern stations exclusively; S11, S13–15) was characterised by 25 small planktonic copepod species (<1.45 mm) contributing to 93% of total copepod abundance, namely *O. nana* (14%), *P. parvus* (14%) and *H. littoralis* (3.5%; Fig. 3; Table 2). However, *P. latisetosa* (<1.45 mm; 6.5%) and *C. helgolandicus* (1.45–2.5 mm; 7.5%) were exclusively dominant at depths of ~3 m (northern stations exclusively; S3, S4 and S8). *O. plumifera* (<1.45 mm; 7.5%) was found in substantial amounts at depths between 0.5 and 3 m (Fig. 3; Table 2). Two larger copepods (1.45–2.5 mm), namely *C. helgolandicus* (1.780 ± 0.047 mm) and *Temora stylifera* (1.490 ± 0.008 mm), were identified in the samples (*T. stylifera* in S14; *C. helgolandicus* in S3 and S8) and accounted for 7.3% of total copepod abundance (Fig. 3; Table 2).

### Factors regulating spatial distribution of copepod assemblages

The first factorial plane of the co-inertia analysis explained 69% of the variance, of which 51% was attributed to the first axis and 18% to the second. Therefore, the co-inertia analysis demonstrated a co-structure between the two datasets (i.e. copepod abundance and environmental–trophic variables). The correlation between the environmental and biological coordinates of the stations on the first factorial plane, reflecting the degree of association between the environmental and copepod systems, was highly significant, with $R$-values of 0.76 and 0.73 for the two factorial planes (Axis F1 environment/Axis F1 biology and Axis F2 environment/Axis $F_2$biology respectively; Fig. 4*c*, *d*). In both the 'environment' and 'copepod' structures, Axis 1 showed a distinction between northern stations (S1–10, except S2) and southern stations (S11–20; Fig. 4*c*). Northern stations were characterised by higher depth, turbidity, COD and phosphorus ($PO_4^{3-}$ and total P) and lower taxonomic diversity. Northern stations were also associated with several copepod taxa (*P. latisetosa*, *C. helgolandicus*, *P. parvus*, *Centropages typicus* and several *Oithona* species; Fig. 4*a*, *b*), including the smallest copepod species, such as *O. nana* (14%; 0.560 ± 0.041 mm) and *P. parvus* (14%; 1.010 ± 0.005 mm). These species were associated with a high COD level, as indicated by the significant positive correlations ($r$ = 0.38–0.50; $P < 0.05$).

Southern stations were characterised by high salinity, pH, $NO_3^-$ and N/P ratio relative to the northern stations, and were associated with other copepod taxa, namely Acartidae (*Acartia clausi*, *Acartia danae*, *Acartia longiremi*, *Paracartia grani*), *Triconia conifera*, *Oncaea minuta*, *T. stylifera*, *Corycaeus latus*, *C. clausi* and *H. littoralis* (Fig. 4*a*, *b*), including large copepod species such as *T. stylifera* (0.03%; 1.490 ± 0.008 mm). The association of these latter copepod species with high SPM and low COD, total P and $PO_4^{3-}$ was particularly strong for stations situated in the last transect (S17–20) in front of Hakmouni wadi, removing the release of the olive oil wastes from margins and the phosphate industry releases to the sea, which was a source of pollutant discharge from the phosphogypsum wastes issued from the SIAPE manufactory and the olive oil wastes from margins. On the second axes of the two systems, in contrast with the other stations, S10 and S13 were characterised by high Si $(OH)_4$ concentrations and by the presence of *Clytemnestra scutellata*.

## Discussion

Sfax (733 687 inhabitants) is a site of intense maritime, urban and industrial activities that contribute to the degradation of coastal water quality (Ben Salem *et al.* 2015; Drira *et al.* 2016). In the southern coastal area, the major sources of pollution include urban, industrial and municipal wastewaters, the fishing harbor and waste-processing phosphates (Ben Abdallah *et al.* 2006; Aloulou *et al.* 2012; Choura *et al.* 2015). Previously, this area was thoroughly characterised with regard to physico-chemical and biogeochemical analyses of the surface waters (Drira *et al.* 2016). In the present study, we focused on copepods that, according to Rajagopal *et al.* (2011) and Ekpo (2013), play an integral role and serve as bioindicators of water pollution (Mukhopadhyay *et al.* 2000). Indeed, species variation, distribution and abundance of zooplankton highly depend on the chemical and physical properties of water (Patra *et al.* 2011). The species diversity index is a relevant tool for water quality assessment and a basis for biomonitoring by evaluating the structural complexity of zooplankton assemblages (Jafari *et al.* 2011; Patra *et al.* 2011). In the present study, the Shannon–Wiener index ($H'$) for copepods was fairly low: $H'$ was <1.5 bits individual$^{-1}$ for 55% of stations, including most northern stations (S1–10), except S3, S5 and S10, which had pioneer communities at the colonisation stage, and ranging between 1.5 and 2.5 bits individual$^{-1}$ for the other stations, demonstrating a transition ecosystem at the diversification stage (Shannon and Weaver 1963). According to Wilhm (1975) and Mukhopadhyay *et al.* (2007), a high $H'$ suggests rich diversity and therefore a healthier (less polluted) ecosystem, whereas a low $H'$ indicates

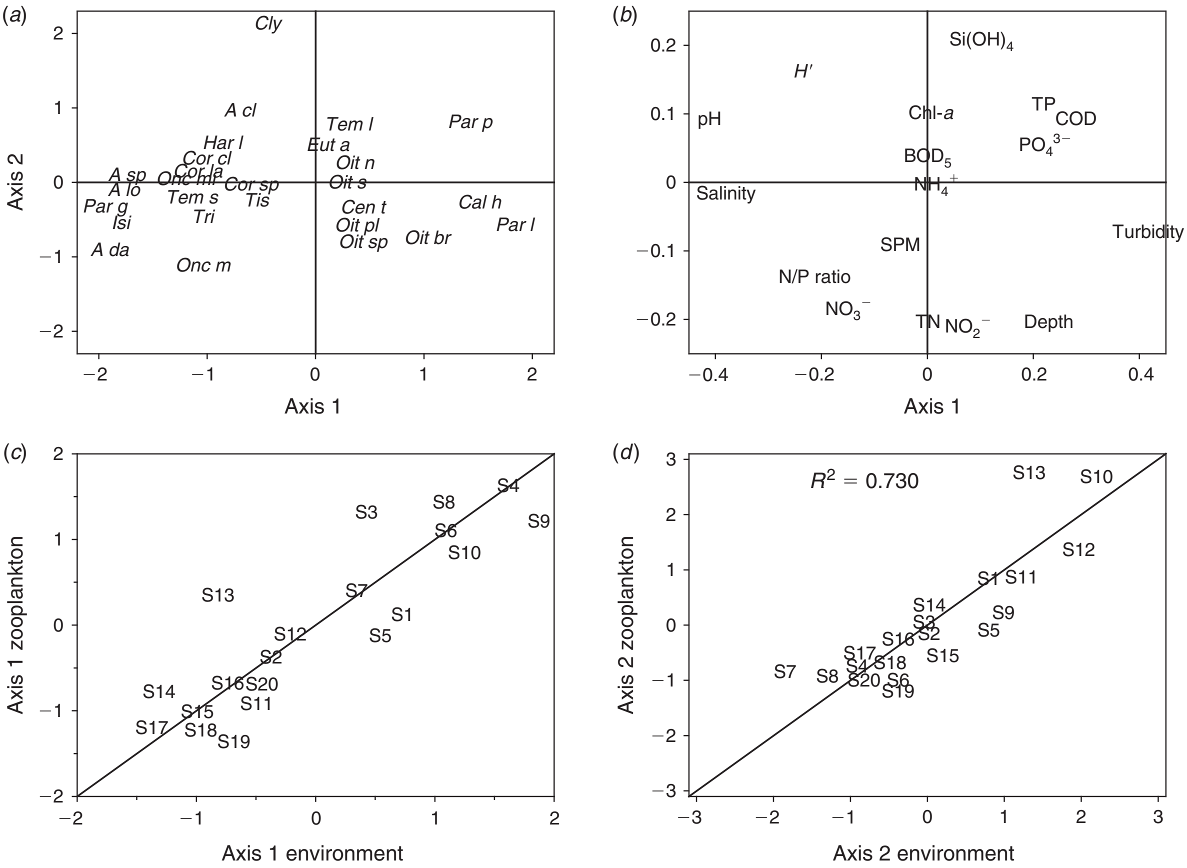


**Fig. 4.** Co-inertia analysis. Ordination on the axes (1, 2) of (*a*) copepod taxa and (*b*) environmental variables and plots of the sampling points on the (*c*) first and (*d*) axes second of the two systems. The diagonal lines represent equality between the coordinates on the two systems. (*a*) Symbols used to indicate taxa are defined in Table 2. S1–20, Stations 1–20; Chl-*a*, chlorophyll-*a*; $H'$, Shannon–Wiener diversity index; TP, total P; TN, total N; $BOD_5$, 5-day biochemical oxygen demand; SPM, suspended particulate matter; COD, chemical oxygen demand.

poor diversity. Quite low $H'$ values for copepod communities (0.48–0.89 bits individual$^{-1}$) were reported in 2008 in the same area (Ben Salem *et al.* 2015). In contrast, the Sfax northern coast (north of the commercial harbour), partly rehabilitated through the TaparuraProject (including confinement of phosphogypsum and depollution of coastal waters; Callaert *et al.* 2009), was characterised by much higher $H'$ values in the same season (most values >2 bits individual$^{-1}$, with a maximum of 3.5 bits individual$^{-1}$; Rekik *et al.* 2012). Note the difference in $H'$ between the southern (present study) and northern (investigated by Rekik *et al.* 2012) coasts in the same season, spring, after the rehabilitation process through the TaparuraProject. The nutrient ($NO_3^-$ and $PO_4^{3-}$) concentrations in the present study are considerably (2- and 20-fold respectively) higher than those reported by Rekik *et al.* (2012). Indeed, high nutrient inputs in a mesotrophic system hinder the stability of the ecosystem, thus leading to a reduction in species richness (Bell 2005; Ben Salem *et al.* 2015). In the present study, with regard to $H'$ values, we may also consider that the water quality in the northern zone of the study area ($H' = 0.75 \pm 0.89$) was substantially degraded compared with that in the southern zone ($H' = 1.54 \pm 0.63$), less affected by the sewage from El Maou wadi, the phosphate industry and WWTP, even though nutrient concentrations were not significantly different between the two zones (Student's *t*-test, $P > 0.05$).

The difference in copepod biodiversity between the northern (S1–10) and southern (S11–20) stations could be explained, in part, by the spatial distribution of dissolved trace metals. Indeed, Ben Salem and Ayadi (2016) showed that the highest contents of dissolved Cd (0.21 μg L$^{-1}$), Cu (4.34 μg L$^{-1}$), Fe (30.74 μg L$^{-1}$), Ni (10.21 μg L$^{-1}$), Pb (3.43 μg L$^{-1}$) and Zn (5.78 μg L$^{-1}$) were found in stations situated in front of the WWTP (i.e. S9 in the present study). The Cu and Ni concentrations exceeded the continuous concentration and maximum concentrationin the USEnvironmental Protection Agency water quality criteria (US Environmental Protection Agency 1999).

COD was more important in the northern than southern stations (Drira *et al.* 2016). The highest values were recorded at S9, located in front of the phosphate industry plant, which releases large amounts of $PO_4^{3-}$, $Cl^-$ and $SO_4^{2-}$ into the sea (Drira *et al.* 2016). Inputs of phosphates and organic matter may contribute to the increase in COD, $BOD_5$ and SPM in surface waters (Davis and Marshall 1998; Negi and Rajput 2013; Drira *et al.* 2016) and subsequently to changes in the community structure of zooplankton (Tallberg *et al.* 1999; Arora and Mehra 2009). In the present study, a $BOD_5$/COD ratio <0.5 indicates pollution of a chemical origin (Ohwoghere-Asuma and Aweto 2013; El Rhaouat *et al.* 2014; Jamwal *et al.* 2015) whereas COD/$BOD_5$ ratios > 3 indicate an organic load of a low biodegradability (Drira *et al.* 2016). In addition, the highest density of copepods ($35.85 \times 10^3$ individuals $m^{-3}$; S6) was associated with a high $BOD_5$ (60 mg $L^{-1}$). $BOD_5$ is a key parameter that regulates the population of copepods (Jagadeeshappa and Kumara 2013). The increase in the total numerical density of zooplankton has been shown to be related to a decrease in SPM load (Bhattacharya *et al.* 2014). The same result was found in the present study for S10, which had the highest zooplankton density ($47.95 \times 10^3$ individuals $m^{-3}$) and the lowest SPM (17.9 mg $L^{-1}$).

The present study showed that there were more adult females than males. Several studies have shown that pollution may affect the population dynamics of zooplankton species by controlling individual survival and reproduction, and by altering the sex ratio. For example, Medina *et al.* (2002) reported that adult males of *Acartia tonsa* were much more sensitive than females to contamination by pyrethroids. This is consistent with the results of the present study showing a preponderance of females and thus suggesting a high mortality of males in a highly polluted environment. The preponderance of females over males, which reduces the sexratio (Kiørboe 2006), may be due to the higher mortality of males because of their increased vulnerability to predation during their search for mates (Mendes-Gusmão *et al.* 2013). However environmental factors driving differential physiological longevity of males and females may be more important in determining the sex ratio (Mendes-Gusmão *et al.* 2013). Comparisons of the size structure and composition of zooplankters can indicate the nature and extent of pollutant loads (Sarma 1996; Mukhopadhyay *et al.* 2000). In the present samples, dominant species such as *O. nana*, *P. parvus*, *H. littoralis* and *T. battagliai* were spread in the shallow waters (~0.5-m depth) along the coast and thus appeared more adapted to the coastal anthropogenic inputs. In addition, the small cyclopoid *O. nana* had clear eurythermal and euryhaline distributions (Drira *et al.* 2014). Nonetheless, large copepods (>1.45 mm), such as *C. helgolandicus* (relative abundance 7.23%, total length $1.78 \pm 0.05$ mm) dominated in the more off-shore zone at ~3-m depth. The coastal anthropogenic inputs affected the copepod species by decreasing their body size. *O. plumifera*, a small copepod species (<1.45 mm; 7.5%), was found in substantial amounts between the inshore and offshore areas. Copepod species can be classified into different ecological groups based on water quality affinities (Hooff and Peterson 2006; Bi *et al.* 2012). In the present study, we identified three different groups that are good indicators of water pollution, as discussed below.

The first group is composed of *O. nana* (14.9% of the total adult copepod count), *P. parvus* (14%) and *H. littoralis* (3.4%), which were the dominant small (<1.45 mm) copepod species. These species were spread along the coast, proliferated primarily in shallow waters (~0.5-m depth) and were thus better adapted than other species to coastal anthropogenic inputs. These species are resistant to unfavourable conditions and have a high capacity to develop in eutrophicated and polluted areas (Arfi *et al.* 1981). *O. nana* was reported to be dominant in the Gulf of Gabes (Drira *et al.* 2014) and in the Tunis North Lagoon (Annabi-Trabelsi *et al.* 2005), accounting for 87 and 31% of total copepod abundance respectively. Several studies have demonstrated that *O. nana*, which has a certain ability to endure environmental perturbations because of its low respiratory rate and omnivorous diet (Gallienne and Robins 2001; Rekik *et al.* 2013*b*; Ben Ltaief *et al.* 2015), is able to proliferate in unstable ecosystems and capable of colonising polluted environments (Williams and Muxagata 2006; Drira *et al.* 2010, 2014; Rekik *et al.* 2013*b*) and areas subjected to eutrophication (Richard and Jamet 2001). A possible link between phosphate and the abundance of *O. nana* in the Gulf of Gabes, and along the southern coast of Sfax in particular, was demonstrated in previous studies (Drira *et al.* 2010, 2014; Ben Salem *et al.* 2015). Thus, this species is cosmopolitan in many marine ecosystems (Fernández de Puelles *et al.* 2007) and dominant in coastal areas of several Mediterranean regions (Mazzocchi and Ribera d'Alcala 1995; Christou 1998), including the Algerian basin (Gaudy 1985; Riandey *et al.* 2005). Tolerance to pollution, as observed in the Gulf of Gabes, may also explain the high proliferation of *O. nana* observed in the Bay of Toulon (north-western Mediterranean) by Jamet *et al.* (2001), who suggested that this species may be used as a biological indicator of such unhealthy systems.

The second group of good indicators of water pollution consists of *P. latisetosa* (<1.45 mm) and *C. helgolandicus* (7.5%; 1.45–2.5 mm), which were dominant only in waters of ~3-m depth (northern stations exclusively; i.e. S3, S4 and S8). The large copepod *C. helgolandicus* is rare in inshore waters, with maximum abundance in the present study observed in offshore waters, as reported for the Irish Sea (Gowen *et al.* 1999) and the Mediterranean Sea (Zakaria *et al.* 2016).

The third group is represented by *O. plumifera* (<1.45 mm; 7.5%), which was found in substantial amounts at depths between 0.5 and 3 m (Fig. 3; Table 2). *O. plumifera* was shown to be the most abundant component of coastal and oceanic copepod assemblages in shallow nearshore waters off the southern coast of South Africa ($6 \times 10^3$ individuals $m^{-3}$; Porri *et al.* 2007). This species exhibits very particular swimming and feeding behaviour (Paffenhöfer and Mazzocchi 2002), allowing it to exploit the large variety of available food resources, such as phytoplankton, ciliates and detritus (Ribera d'Alcalà *et al.* 2004). A perusal of literature has shown that this species is tolerant of organic pollution, which may favour its dominance when COD values are high (Bhattacharya *et al.* 2014).

Along the Sfax southern coast, anthropogenic inputs as well as tidal and current conditions may explain the high level of organic debris preferentially consumed by *O. plumifera*. Furthermore, it was demonstrated that suspended matter was homogeneously distributed in neritic and open sea in the Gulf of Gabes (Drira *et al.* 2010). In summary, the present study demonstrates that the nearshore coastal waters appear to be more responsive or sensitive to anthropogenic inputs, resulting in the different copepod species assemblages.

## Conclusions

Zooplankton composition may reflect water quality in coastal marine ecosystems. The present study was undertaken to assess zooplankton species diversity in relation to anthropogenic inputs along the southern coastline of Sfax. Copepod assemblages could move along the tides and currents, which may partially mask the effects of water chemistry (anthropogenic inputs) on zooplankton diversity and composition. The results of the present study revealed that the most abundant mesozooplankton copepod species (*O. nana*, *P. parvus*, *H. littoralis* and *T. battagliai*) were spread along the coast and were more adapted and tolerant of coastal anthropogenic inputs. The copepod diversity was higher in the southern, less-affected stations ($H'$ between 1.5 and 2.5 bits individual$^{-1}$) than in the northern stations, which were more affected by sewage ($H' \leq$ 1.5 bits individual$^{-1}$). In addition, a shift in the planktonic copepod community between the two zones was linked to a deterioration in water quality (high phosphorus content, high turbidity, high COD) in the northern zone directly subjected to higher levels of pollutant discharge. Increased nutrient loading from expanding anthropogenic activities along the southern coast of Sfax must be carefully considered in order to maintain the stability of the ecosystem and increase plankton richness. Therefore, integrated management of the southern coast of Sfax remains an important need and public awareness is required to improve the water quality and biodiversity in the region.

## Conflicts of interest

The authors declare that they have no conflicts of interest.

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