

**Influence of Hydrological Heterogeneity on Rotifer Community Structure in Three Different Water Bodies in Shantou Area, Guangdong (China)**

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*Received 15 October 2018 / Accepted 11 July 2019 / Published xx July 2019*

*Communicated by Benny K.K. Chan*

Rotifers, small but essential invertebrates in aquatic ecosystems, are sensitive to environmental changes and are proposed as indicators of trophic state. However, the effects of hydrological heterogeneity on the rotifer community and the ability of rotifer indices to reflect trophic state when comparing different water bodies are still unclear. Here, we investigated rotifer community structure in different seasons in the three types of water bodies: Han river downstream (HD), Reservoir (RE) and Tidal creek (TC) in Shantou City, Guangdong, China. Our findings revealed that rotifer community structure differed significantly among the three water bodies, resulting from a dominance of *Keratella cochlearis*, *Anuraeopsis fissa* and *Polyarthra vulgaris*, who largely accounted for the differences of water bodies. Chlorophyll-*a* and Transparency were the main environmental drivers in RE rotifer communities, while Total Nitrogen, Total Phosphorus and salinity were the main factors in HD and TC communities. Rotifer abundance and the rotifer trophic state index decreased in the order: RE > HD > TC. However, both the Sladeczek's B/T quotient and the *Keratella*-index decreased in the order: HD > RE > TC, which was in accord with the Carlson's trophic index. We conclude that it is efficient to use rotifer composition in water quality assessment when comparing different water bodies. Alpha diversity of rotifers was the highest in HD, which is

consistent with the intermediate disturbance hypothesis. Hydrological heterogeneity were the macro-factors that regulates rotifer community structures in Shantou area.

**Key word:** Rotifera, Different habitats, Environmental factors, Trophic state, Intermediate disturbance.

Citation: Liang D, Wei N, Wang Q, Jersabek CD, He X, Yang Y. 2019. Influence of hydrological heterogeneity on rotifer community structure in three different water bodies in Shantou area, Guangdong (China). *Zool Stud* **58**:0v. doi:-.

## BACKGROUND

Rotifers, a group of essential zooplankton in aquatic ecosystems, are sensitive to changes of the environment, acting as effective indicators of the trophic conditions (Duggan et al. 2002; May et al. 2014; Sládeček 1983). Rotifers connect primary producers and secondary consumers, playing an important role not only in the food chain but also the microbial food web (Arndt 1993; Devetter and Sed'A 2003). Rotifers are also highly adaptive to the changes in the environment and can occupy open niches quickly, and as such they are widely distributed in the world and live in all kinds of water bodies. Consequently, they are adapted to a variety of characteristics of different waterbodies (Segers 2007). In the wide range of environments, rotifer community structure is influenced by the factors such as water temperature, salinity, transparency, trophic status and predators (Devetter 1998; Khaleqsefat 2013). As the biotic and abiotic environmental conditions vary seasonally, the community structure of rotifers also varies with season in a single water body, often with succession of different dominant species (El-Shabrawy and Germoush 2014) and changes in diversity and evenness (Bonecker et al. 2013). It has been shown that rotifer abundance in rivers varies from the rainy season to the dry season periodically (Rougier et al. 2005).

While numerous studies of rotifer community structure have investigated seasonal changes or periodical changes in a single type of water body, few studies compared different water bodies in the same area. Hydrological regimes in different habitats is a more important factor diversifying the living conditions of zooplankton assemblages (Ginders et al. 2016). Tidal creek is a lotic water ecosystem that connects river and sea, similar to an estuary. It is affected by both terrigenous

freshwater flow and tides so that the hydrological conditions and environmental factors change dramatically (Hackney et al. 1976). Reservoir is an artificial facility at the narrow portion of the river, which can be used to generate electricity and for irrigation. Reduction of water velocity and increase of water transparency can result in replacement of lotic species by lentic species after stabilization of the system (Serafim-júnior et al. 2016). It was reported that the open lentic waters harbor a rich variety of rotifers because of the macrophytes (Arora and Mehra 2003). River is a lotic water ecosystem that connects land and sea. Its water level and flow rate fluctuation have been correlated to precipitation and seasonal change (Rougier et al. 2005). Different water bodies have different hydrological characteristics, so the rotifer communities are expected to vary. Since the Shantou area contains different types of water bodies, such as reservoirs, rivers, tidal creeks, it is representative to study the effect of hydrological heterogeneity on rotifer community structure.

Rotifer species composition, total abundance, and diversity indices have been widely used as biological indicators for assessing water quality (May and O'Hare 2005; Wen et al. 2011; Gutkowska et al. 2013). Saprobic species and some rotifer composition indices such as *Brachionus*: *Trichocerca* ratio (Sládeček 1983) and Keratella-index (Gopko and Telesh 2013) have been used as trophic state indicators. Also, the rotifer trophic state index ( $TSI_{ROT}$ ) could be a useful tool for assessing the ecological quality of urban ecosystems (Jurczak et al. 2018). May and O'Hare (2005) claimed that rotifer total abundance is a more sensitive indicator of lake trophic status than species composition. However, there is limited knowledge about whether these indicators are capable of reflecting trophic state when comparing different water bodies.

The aim of this study was to investigate the following questions: (1) which rotifer indicators are suitable to reflect trophic state of the three different water bodies? (2) Which factors are responsible for the difference of rotifer communities? (3) Which water bodies harbor the highest diversity?

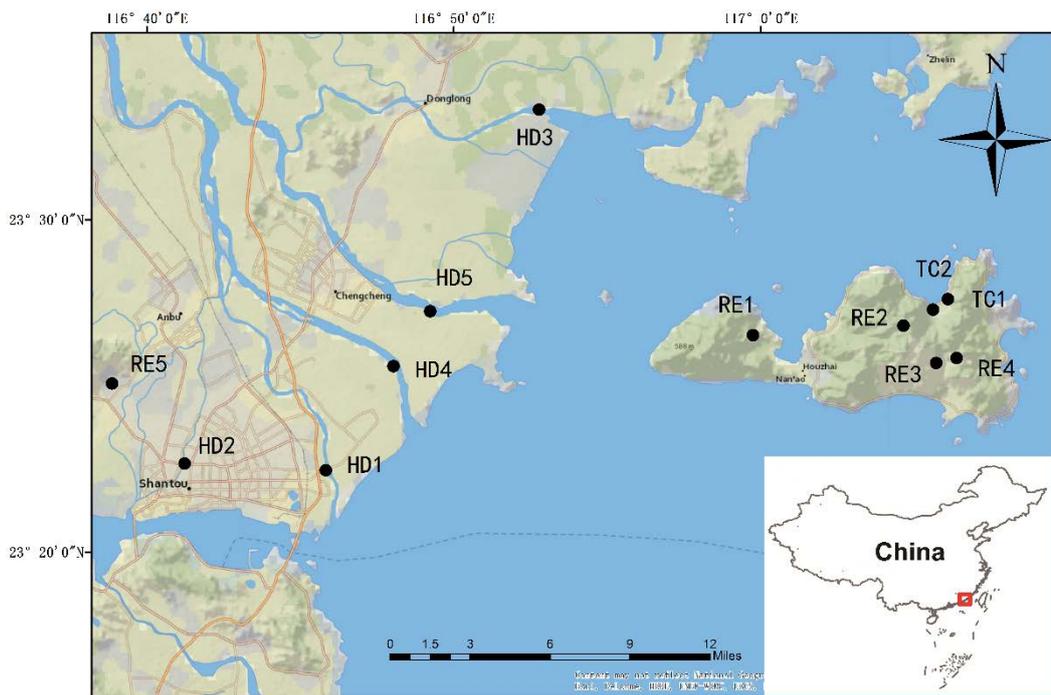
## **MATERIALS AND METHODS**

### **Study sites**

This study was conducted around Shantou, an eastern city of Guangdong province, China, which is affected by the subtropical monsoon climate. The Han River downstream (HD), which

flows through the city, experiences a wet season (April to September) and a dry season (October to March). HD serves as the drinking water supply for the eastern cities of Guangdong province. Its downstream is divided into three main tributaries (northern stream, western stream and eastern stream), running through Shantou city and towards the South China sea. The annual precipitation was 1496 mm in 2015, and 2110 mm in 2016, respectively (Shantou annual climate bulletin). Nanao Island is about 6 km away from the mainland, provided reservoir and tidal creek sites.

Our study was carried out at 12 sampling sites, including 5 Han River downstream sites (HD1, HD2, HD3, HD4, HD5), 5 reservoir sites (RE1, RE2, RE3, RE4, RE5) and 2 tidal creek sites (TC1, TC2) (Fig. 1). Among these, HD3 was located in the northern tributary, HD5 was in the eastern tributary and HD1 HD2 HD4 were in the western tributary of Han River, flowing through downtown Shantou. Apart from RE5, which was on the mainland, the other reservoir sites were located on Nanao Island as freshwater reserves. The tidal creek sites, located in Shenao Town of Nanao Island, receive domestic sewage discharges and are subject to tides.



**Fig. 1.** Location of the study area and the sampling sites covered in Shantou City Abbreviations used in the figures: HD, Han River downstream; RE, reservoir; TC, tidal creek.

### Sampling and analytical procedure

Samples of rotifers were collected 4 times, in July 2015, November 2015, January 2016 and

May 2016. Quantitative samples of rotifers were collected in triplicate each from 5 liters of surface water. These samples were concentrated over a 30  $\mu\text{m}$  mesh. Qualitative samples for species identification were collected by dragging a plankton net with a mesh size of 30  $\mu\text{m}$  on the surface and subsurface water horizontally. Both quantitative and qualitative samples were fixed with 5% formalin solution and preserved in a 50 mL polyethylene bottle immediately. Physical factors such as water temperature, dissolved oxygen, pH, and salinity were measured using a YSI-Plus calibrated multiprobe (USA). Water transparency was measured with a Secchi disc. Chlorophyll-a, total dissolved phosphorus (TP) and total dissolved nitrogen (TN) were determined in the laboratory following the standard analytical methods (GB3838, China, 2002).

Rotifer identification was based on the Koste (1978) classification System. Trophi were isolated from the qualitative samples for further identification and then the list of rotifer species of different water bodies in Shantou area was tabulated. Quantitative samples were concentrated to 10 mL after sedimentation. One mL concentrated solution was taken randomly after mixing and analyzed in a Sedgewick-Rafter chamber. The abundance counts were converted to  $\text{ind.}\cdot\text{L}^{-1}$ .

### **Statistical analysis**

The results were processed by ANOVA using the non-parametric Kruskal-Wallis and Dunn's tests, determining significant differences in rotifer density, rotifer community indices, and physical-chemical factors among the three water bodies. In order to assess the relationship between rotifer abundance and environmental factors, we used SPSS 22.0 to perform the Pearson or Spearman correlation analysis. Correlation coefficients were calculated with Spearman ranks ( $P \leq 0.05$ ). Stepwise multivariate regression was performed to determine the main environmental factors that affected the abundance and rotifer community indices.

The dataset of species was log transformed ( $\log(n + 1)$ ) and standardized, and rotifer community structure was processed using PRIMER 5 to obtain the following indices:

1. Alpha diversity: Margalef species richness index, Simpson dominance index, Shannon-Weiner diversity index, Brillouin diversity index and Pielous evenness index were calculated to evaluate the diversity within water bodies.
2. Beta diversity: Cluster analysis, non-metric multidimensional scaling (NMDS) and analysis of group dissimilarities (ANOSIM), based on Bray-Curtis distance, were analyzed to determine the

differentiation among the three types of water bodies.

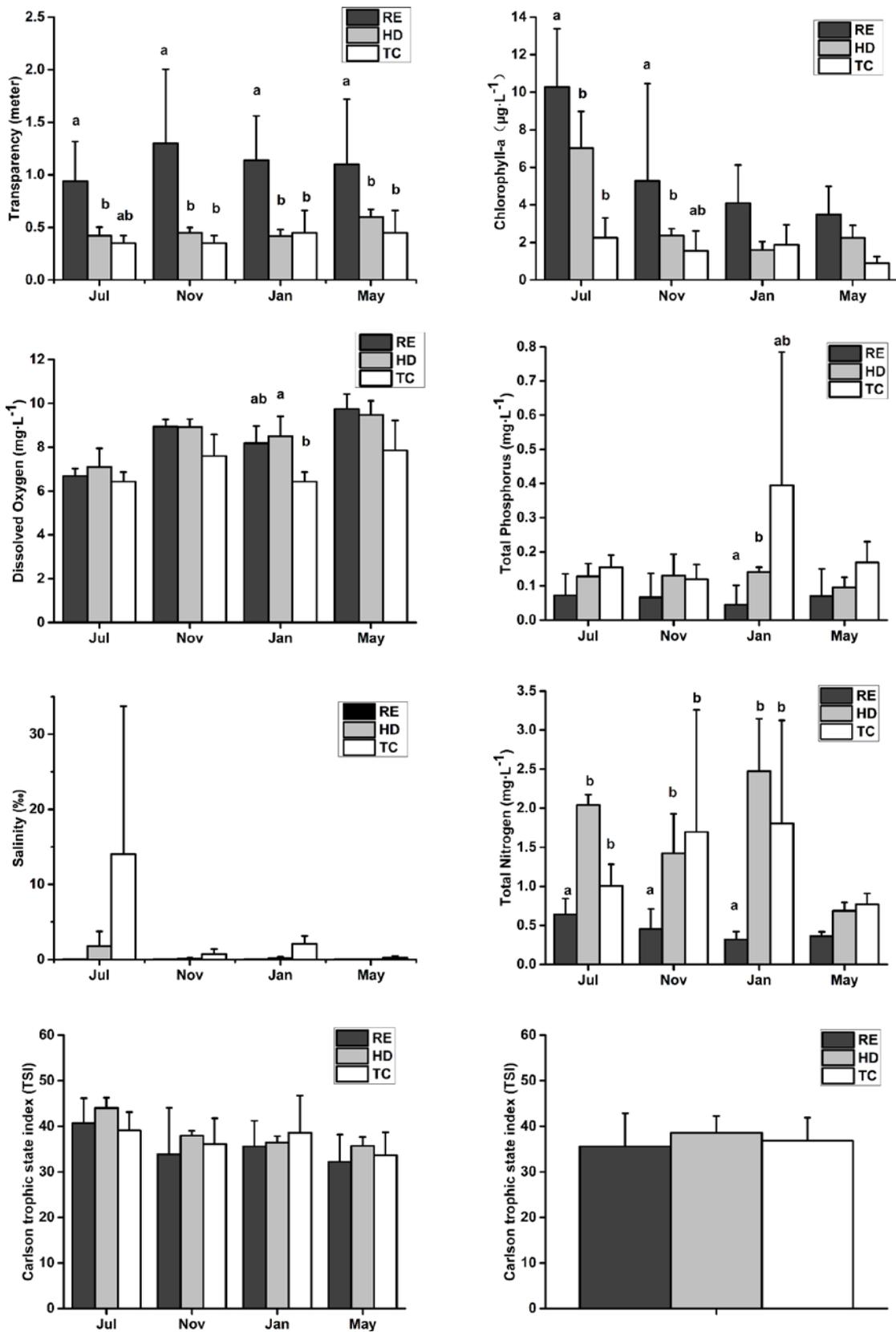
Carlson's trophic state index (Carlson 1977) was applied to evaluate the trophic status of the three water bodies. Brachionus: Trichocerca ratio (Sladeczek's B/T quotient) (Sládeček 1983), *Keratella*-index (KIN) (Gopko and Telesh 2013) and the rotifer trophic state index (TSI<sub>Rot</sub>) (Ejsmont-Karabin 2012) were also used in the trophic state assessment.

The response of rotifer community structure to the environmental variables was analyzed with CANOCO 4.5, a multivariate statistical package. The dataset was log transformed [ $\log(n + 1)$ ] and centered on species. According to the length of gradient (4.53) performed by DCA, we used a CCA (Canonical Correlation Analysis) model to estimate the relationship between rotifer species and environmental factors.

## RESULTS

### Environmental characteristics

The details of the physicochemical data and variations are presented in figure 2. During the study period (2015–2016), water temperatures were similar among all the sampling sites. The lowest temperature occurred in January 2016 at  $16.1 \pm 0.8^\circ\text{C}$ , and the highest temperature occurred in July 2015 at  $32.2 \pm 1.4^\circ\text{C}$ . In November 2015 and May 2016, temperatures were  $23 \pm 0.9^\circ\text{C}$  and  $25.3 \pm 1.1^\circ\text{C}$ , respectively. TC water was lotic water under tidal influence, showing fluctuations of water level and salinity. The pH of all sites ranged from 6 to 9, and on average was slightly alkaline (pH  $7.87 \pm 0.41$ ). The Secchi disc depth (SD) in RE was significantly higher than that in HD and TC ( $P < 0.05$ ) (Fig. 2). The concentrations of chlorophyll-a recorded in RE were significantly higher than that in HD and TC ( $P < 0.05$ ). However, the lowest concentration of TP and TN were found in RE (TP =  $0.06 \pm 0.05 \text{ mg}\cdot\text{L}^{-1}$ ; TN =  $0.44 \pm 0.20 \text{ mg}\cdot\text{L}^{-1}$ ), while the concentration in HD (TP =  $0.12 \pm 0.04 \text{ mg}\cdot\text{L}^{-1}$ ; TN =  $1.65 \pm 0.79 \text{ mg}\cdot\text{L}^{-1}$ ) and TC (TP =  $0.20 \pm 0.18 \text{ mg}\cdot\text{L}^{-1}$ ; TN =  $1.31 \pm 0.91 \text{ mg}\cdot\text{L}^{-1}$ ) were significantly higher than that in RE ( $P < 0.05$ ). The lowest Carlson's trophic state index average value was found in RE, while the highest value was found in HD in most seasons. According to the Carlson's TSI, TC was oligotrophic, while RE and HD were oligotrophic except in July (mesotrophic).



**Fig. 2.** Environmental factors and trophic states in the different water bodies. Letters indicate sample means that are similar (same letter) or significantly different (different letter) among different water bodies; blank represents no significant difference.

**Species composition and abundance of rotifers**

A total of 61 species belonging to 23 genera were identified in all sampling sites during the study period, including subspecies and unidentified bdelloidea. The highest number of taxa occurred in RE (44 species), followed by HD (36 species), and lowest (11 species) in TC. The dominant genera were *Brachionus* (10 species) and *Lecane* (8 species). *Keratella cochlearis*, *Polyarthra vulgaris* and *Anuraeopsis fissa* were distributed in all types of water bodies (Table 1). The prevailing species *K. cochlearis* was widely distributed in all sampling sites of RE, while *A. fissa* and *B. angularis* dominated HD. *B. donneri*, *P. euryptera* and *Hexarthra mira* were only found in RE2, a reservoir on Nanao Island at an elevation of 384 m.

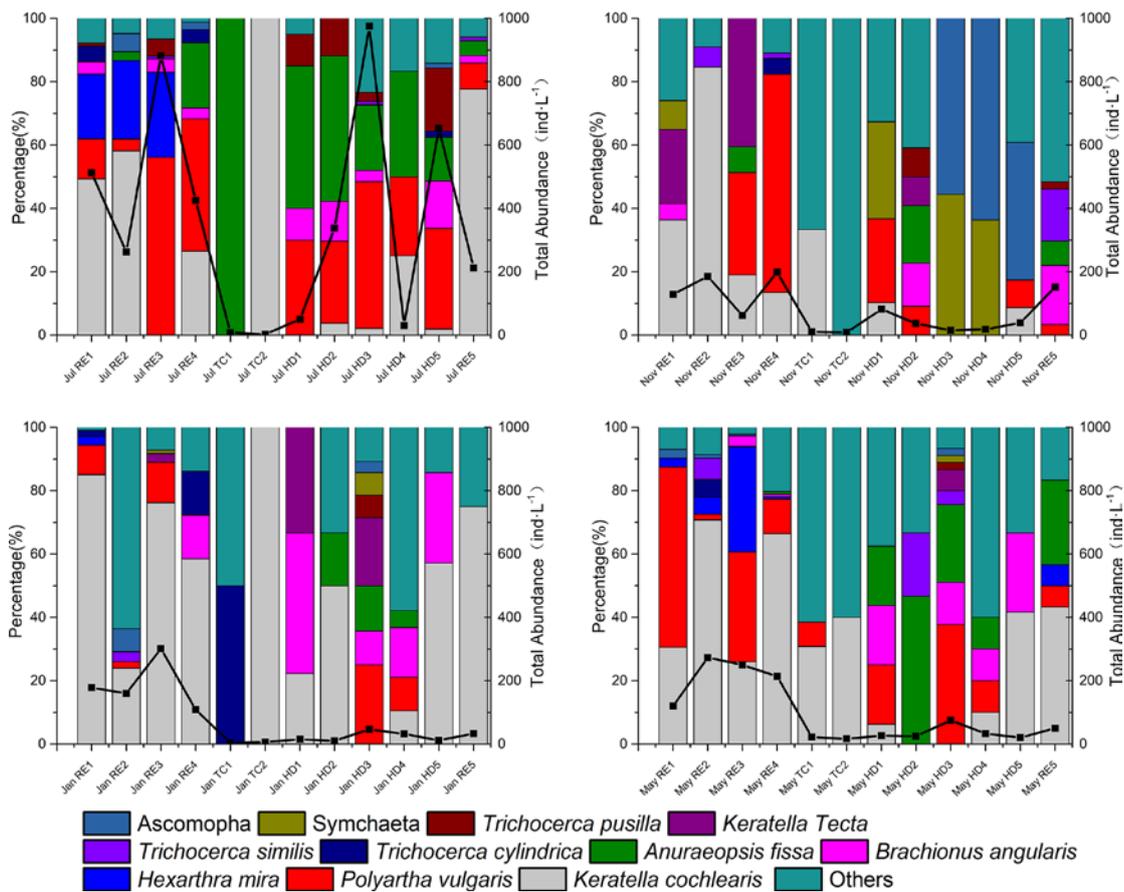
The highest abundance of rotifers occurred at HD3 in July 2015(975 ind.·L<sup>-1</sup>), the rainy season of Guangdong and the lowest abundance occurred at TC1 in January 2016(1 ind.·L<sup>-1</sup>), the dry season of Guangdong. In November, the highest abundance occurred in RE4 (198 ind.·L<sup>-1</sup>) and in May it occurred in RE2 (273 ind.·L<sup>-1</sup>) (Fig. 3).

**Table 1.** The list of rotifer species observed in the three water bodies investigated in Shantou.

Species Names	Abbreviation used in CCA	Water Bodies		
		RE (Reservoir)	HD (Han Downstream)	TC (Tidal creek)
<i>Brachionus calyciflorus</i> f. <i>monstruosus</i> De Ridder, 1987	<i>Bra_cal</i>		+	
<i>B. urceolaris</i> Müller, 1773	<i>Bra_urc</i>	+		
<i>B. angularis</i> Gosse, 1851	<i>Bra_ang</i>	+	+	
<i>B. plicatilis</i> Müller, 1786	<i>Bra_pli</i>			+
<i>B. caudatus</i> Barrois & Daday, 1894	<i>Bra_cau</i>	+	+	
<i>B. falcatus</i> Zacharias, 1898	<i>Bra_fal</i>	+		
<i>B. quadridentatus</i> Hermann, 1783	<i>Bra_qua</i>		+	
<i>B. donneri</i> Brehm, 1951	<i>Bra_don</i>	+		
<i>B. diversicornis</i> Daday, 1883	<i>Bra_div</i>	+		
<i>B. forficula</i> Wierzejski, 1891	<i>Bra_for</i>		+	
<i>Anuraeopsis fissa</i> Gosse, 1851	<i>Anu_fis</i>	+	+	+
<i>Polyarthra dolichoptera</i> Idelson, 1925	<i>Pol_dol</i>	+	+	
<i>P. vulgaris</i> Carlin, 1943	<i>Pol_yul</i>	+	+	+
<i>P. euryptera</i> Wierzejski, 1891	<i>Pol_eur</i>	+		
<i>P. indica</i> Segers & Babu, 1999	<i>Pol_ind</i>		+	
<i>P. remata</i> Skorikov, 1896	<i>Pol_rem</i>	+		
<i>Filinia longiseta</i> Ehrenberg, 1834	<i>Fil_lon</i>		+	
<i>F. opoliensis</i> Zacharias, 1898	<i>Fil_opo</i>	+		
<i>F. cornuta</i> Weisse, 1848	<i>Fil_cor</i>		+	
<i>F. novaezealandiae</i> Shiel & Sanoamuang, 1993	<i>Fil_nov</i>		+	
<i>Asplanchna priodonta</i> Gosse, 1850	<i>Asp_pro</i>	+	+	

<i>A. brightwellii</i> Gosse, 1850	<i>Asp_bri</i>	+	+	
<i>Keratella cochlearis</i> Gosse, 1851	<i>Ker_coc</i>	++	+	+
<i>K. tecta</i> Gosse, 1851	<i>Ker_tec</i>	+	+	
<i>K. tropica</i> Apstein, 1907	<i>Ker_tro</i>	+	+	
<i>Lecane bulla</i> Gosse, 1851	<i>Lec_bul</i>	+		+
<i>L. closteroerca</i> Schmarda, 1859	<i>Lec_clo</i>			+
<i>L. papuana</i> Murray, 1913	<i>Lec_pap</i>	+		
<i>L. stichaea</i> Haring, 1913	<i>Lec_sti</i>	+		
<i>L. unguata</i> Gosse, 1887	<i>Lec_ung</i>		+	+
<i>L. stenroosi</i> Meissner, 1908	<i>Lec_ste</i>	+		
<i>L. hamata</i> Stokes, 1896	<i>Lec_ham</i>	+		
<i>L. luna</i> Müller, 1776	<i>Lec_lun</i>		+	+
<i>Proalides subtilis</i> Rodewald, 1940	<i>Pro_sub</i>		+	
<i>Hexarthra mira</i> Hudson, 1871	<i>Hex_mir</i>	+		
<i>Collotheca</i> sp.	<i>Collotheca</i>	+	+	
<i>Trichocerca pusilla</i> Jennings, 1903	<i>Tri_pus</i>	+	+	
<i>T. similis</i> Wierzejski, 1893	<i>Tri_sim</i>	+	+	
<i>T. stylata</i> Gosse, 1851	<i>Tri_sty</i>	+	+	
<i>T. cylindrica</i> Imhof, 1891	<i>Tri_cyl</i>	+	+	+
<i>T. insignis</i> Herrick, 1885	<i>Tri_ins</i>	+		
<i>T. rousseleti</i> Voigt, 1902	<i>Tri_rou</i>	+		
<i>T. capucina</i> Wierzejski & Zacharias, 1893	<i>Tri_cap</i>	+		
<i>Trichotria tetractis</i> Ehrenberg, 1830	<i>Tri_tet</i>		+	
<i>Wolga spinifera</i> Western, 1894	<i>Wol_spi</i>	+		
<i>Mytilina ventralis</i> Ehrenberg, 1830	<i>Myt_ven</i>	+		
<i>Testudinella patina</i> Hermann, 1783	<i>Tes_pat</i>		+	+
<i>Gastropus hyptopus</i> Ehrenberg, 1838	<i>Gas_hyp</i>	+		
<i>Ascomorpha ovalis</i> Bergendal, 1892	<i>Asc_ova</i>	+	+	
<i>A. saltans</i> Bartsch, 1870	<i>Asc_sal</i>	+	+	
<i>Synchaeta stylata</i> Wierzejski, 1893	<i>Syn_sty</i>		+	
<i>S. oblonga</i> Ehrenberg, 1832	<i>Syn_obl</i>	+	+	
<i>S. tremula</i> Müller, 1786	<i>Syn_tre</i>		+	
<i>S. pectinata</i> Ehrenberg, 1832	<i>Syn_pec</i>		+	
<i>Colurella adriatica</i> Ehrenberg, 1831	<i>Col_adr</i>	+		
<i>Ploesoma truncatum</i> Levander, 1894	<i>Plo_tru</i>	+		
<i>Cephalodella</i> sp.	<i>Cephalodella</i>	+	+	
<i>Cephalodella gibba</i> Ehrenberg, 1830	<i>Cep_gib</i>	+		
<i>Conochilus unicornis</i> Rousselet, 1892	<i>Con_uni</i>	+		
<i>Encentrum</i> sp.	<i>Encentrum</i>	+	+	
<i>Bdelloidea</i> spp.	<i>Bdelloidea</i>	+	+	+

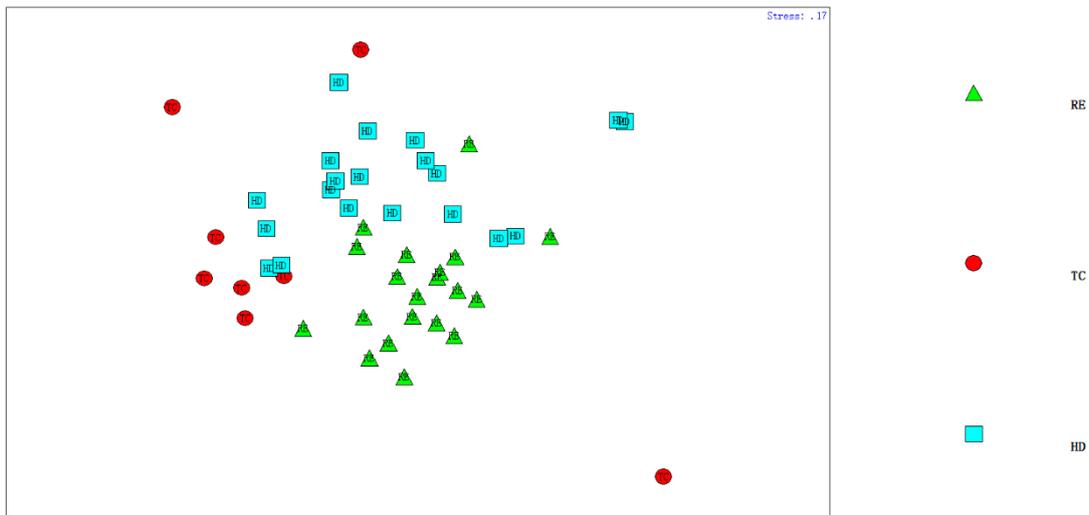
'+' represents the presence of the item; blank represents the absence of the item.



**Fig. 3.** The proportions (Y axis on left) and the abundances (Y axis on right) of the dominant rotifers in different sampling sites in Shantou. The lines represent the total abundances and the column represent the rotifer composition.

### Differentiation of rotifer community structure among different water bodies

A non-metric multidimensional scaling (NMDS) plot was used to reveal the pattern in rotifer community structure in relation to different water bodies. The stress value of NMDS was 0.17 (< 0.2). The RE samples were mostly together and distributed below the First Axis and the HD samples tended to be above the First Axis. TC was mostly on the left but with some points on far right. The result of NMDS revealed that the majority of values from each water body are located in a certain position in the plot and separated from each other, but with some exceptions. (Fig. 4).



**Fig. 4.** The non-metric multidimensional scaling (NMDS) plots of rotifer community in Shantou.

The analysis of group dissimilarity processed by PRIMER using ANOSIM (one-way), demonstrated that rotifer communities were significantly different among three types of water bodies in Shantou area (Global  $R = 0.416$ ,  $P < 0.01$ ). The ANOSIM test indicated that the differences between groups were significantly greater than those within groups, which demonstrated that it is meaningful to group by water bodies. Among them, rotifer community structure in RE was not only significantly different from that in TC ( $R = 0.698$ ,  $P < 0.01$ ), but also significantly different from that in HD ( $R = 0.348$ ,  $P < 0.01$ ). The seasonal differences of rotifer communities were relatively small ( $R = 0.106$ ,  $P < 0.01$ ), when compared to different types of water bodies. This differentiation between HD and RE water bodies was caused by the abundances of their respective dominant species.

According to the contribution rate of rotifers in different water bodies, we found that the variation between RE and HD, the lentic and lotic water, resulted from the abundances of the dominant species in each of the different communities. *Keratella cochlearis*, a dominant species in RE, with the mean abundance significantly higher than that in HD, made the greatest contribution to the variation of different communities (10.43%). In addition, *A. fissa*, *P. vulgaris*, *H. mira* and *B. angularis* also played an important role in their differentiation. On the other hand, the mean abundance of *P. vulgaris* and *H. mira* in RE were significantly higher than that in HD, while abundances of *A. fissa* and *B. angularis* in HD were higher than in RE (Table 2). The abundances of these species were responsible for the differentiation between RE and HD.

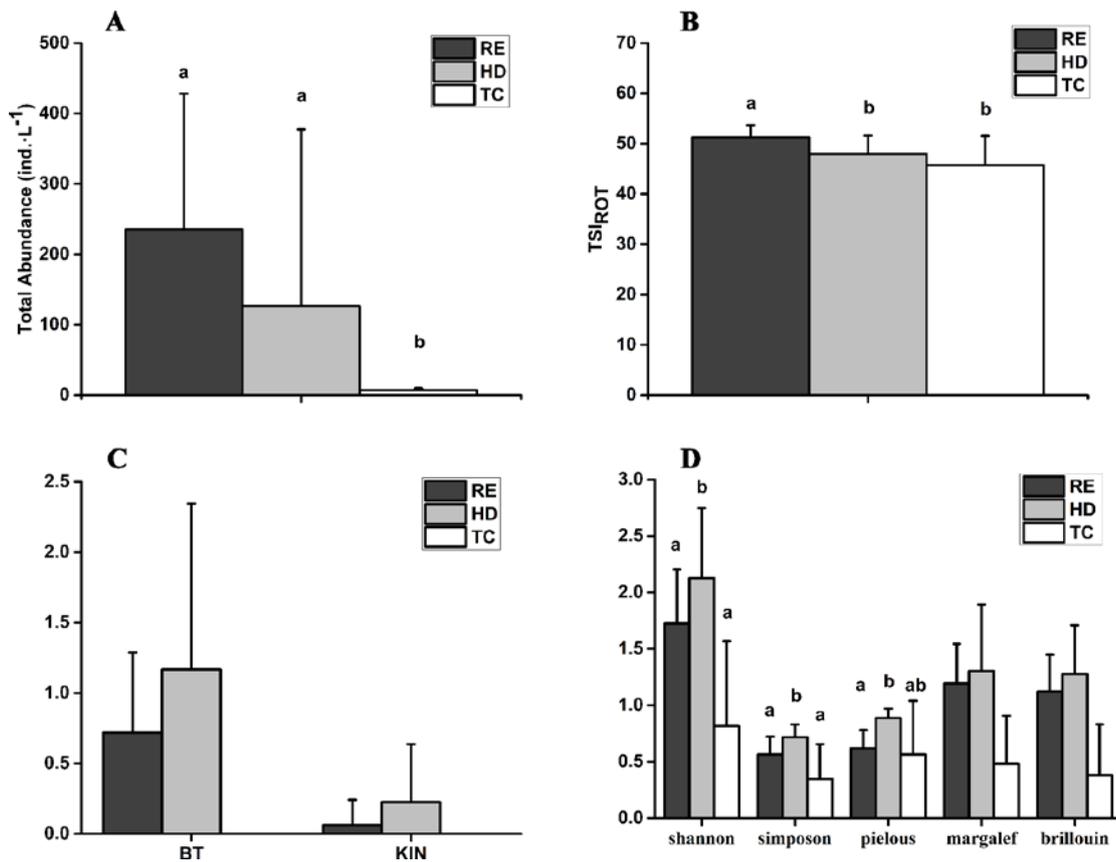
**Table 2.** Dominant species and their contribution rate to differences between RE and HD in Shantou

Species	Average Abundance (Log)		Contribution (%)
	RE (Reservoir)	HD (Han Downstream)	
<i>Keratella cochlearis</i>	94.54	4.79	10.43
<i>Anuraeopsis fissa</i>	6.83	26.63	8.41
<i>Polyarthra vulgaris</i>	58.58	42.50	8.23
<i>Brachionus angularis</i>	6.88	11.42	7.51
<i>Hexarthra mira</i>	25.96	0	4.80
<i>Ascomorpha saltans</i>	1.67	2.42	4.54
<i>Synchaeta stylata</i>	0	2.33	3.92
<i>K. tecta</i>	3.17	1.17	3.79
<i>S. oblonga</i>	0.75	1.83	3.67
<i>Trichocerca pusilla</i>	2.79	10.54	3.50
<i>T. similis</i>	3.67	0.92	3.19
<i>T. cylindrica</i>	4.29	0.75	2.64

### Rotifer community indices

Rotifer abundance in TC ( $7 \pm 3$ ) was significantly lower than that in HD ( $127 \pm 251$ ) and RE ( $235 \pm 193$ ) ( $P < 0.05$ ) (Fig. 5A). The highest  $TSI_{ROT}$  was recorded in RE ( $51 \pm 2$ ;  $P < 0.05$ ), followed by HD ( $48 \pm 3$ ), and lowest ( $46 \pm 5$ ) in TC (Fig. 5B). However, both of the BT and KIN indices were in the order: HD (1.2; 0.22) > RE (0.7; 0.06) > TC (0; 0) (Fig. 5C).

The highest diversity index of both Simpson's in HD and Shannon-Weiner ( $0.72 \pm 0.11$ ;  $2.13 \pm 0.62$ ) were significantly higher than that in RE ( $0.56 \pm 0.15$ ;  $1.72 \pm 0.48$ ) and TC ( $0.35 \pm 0.30$ ;  $0.82 \pm 0.75$ ) ( $P < 0.01$ ;  $P < 0.05$ ). The higher proportion of dominant species in RE resulted in a decrease of the value of Pielou's evenness ( $0.62 \pm 0.16$ ), which was significantly lower than that in HD ( $0.89 \pm 0.08$ ;  $p < 0.01$ ). The Margalef and Brillouin diversities were higher in HD ( $1.30 \pm 0.59$ ;  $1.28 \pm 0.43$ ), but not significantly ( $P > 0.05$ ) (Fig. 5D). The diversity and evenness levels suggested that the species composition of HD water bodies was more even, compared to RE and TC water bodies.



**Fig. 5.** Rotifer community indices among three water bodies. (A) Rotifer total abundance; (B) Ejsmont-Karabin's rotifer trophic state index; (C) Sladeczek's B/T quotient and *Keratella*-index (KIN) values; (D)  $\alpha$  biodiversity indices. Letters indicate sample means that are similar (same letter) or significantly different (different letter) among different water bodies; blank represents no significant difference.

### Relationships between community composition and environment variables

Rotifer abundance showed a significantly positive correlation with chlorophyll-*a* concentration ( $R = 0.52$ ,  $P < 0.001$ ). However, there was no significant correlation between rotifer abundance and other environment factors such as SD, TN, TP, temperature, or salinity ( $P > 0.05$ ). The multivariate regression analysis was conducted with the rotifer abundance as response variable and environment factors as dependent variable including chlorophyll-*a*, SD, TN, TP, temperature (Temp), salinity (Sal), pH and DO. After eliminating the non-significant independent variables, multivariate regression analysis suggested that the total abundance of rotifers was only significantly related to chlorophyll-*a*, SD and Temperature (Table 3). The Shannon-Weiner index was correlated with salinity. Simpson and Pielou's index were associated with salinity, TN, DO, and temperature, and

Margalef was related to DO only.

**Table 3.** Multiple regression of rotifer total abundance and  $\alpha$  diversity

Category	Regression modle	Adjusted $R^2$	$P$
Abundance	$Y = -317.364^{**} + 23.364 \text{ Chla}^{**} + 128.242 \text{ SD}^* + 11.596 \text{ Temp}^*$	0.377	0.00
$\text{TSI}_{\text{ROT}}$	$Y = 48.909^{**} - 16.134 \text{ TP}^{**} + 0.459 \text{ Chla}^{**}$	0.281	0.00
Shannon-Weiner	$Y = 1.804^{**} - 0.067 \text{ SAL}^{**}$	0.119	0.00
Simpson's	$Y = -0.489 - 0.021 \text{ Sal}^{**} + 0.116 \text{ TN}^{**} + 0.081 \text{ DO}^{**} + 0.013 \text{ Temp}^{**}$	0.41	0.00
Pielou's	$Y = -0.055 - 0.022 \text{ Sal}^{**} + 0.157 \text{ TN}^{**} + 0.076 \text{ DO}^{**}$	0.421	0.00
Margalef	$Y = -0.146 + 0.154 \text{ DO}^{**}$	0.104	0.01

\* $P < 0.05$ ; \*\* $P < 0.01$ .

The CCA summarized the relations between the rotifer species composition and environmental variables (Fig. 6) ( $Pseudo-F = 1.628$ ,  $P < 0.001$ ). After forward selection with Monte Carlo permutation tests, Chlorophyll-*a*, TP, TN, SD and temperature were significant contributors to the model (Table 4). The first two ordination axes explained 43.6% of species–environment variability in the ordination of physical and chemical factors (Table 5).

**Table 4.** Correlation coefficient between environmental factors and the first two ordination axes

	SPEC AX1	SPEC AX2	ENVI AX1	ENVI AX2
Chlorophyll- <i>a</i>	-0.6732***	-0.4795***	-0.8076***	-0.5524***
TP	0.3226**	-0.4925***	0.3871***	-0.5674 ***
TN	0.3187**	-0.6504***	0.3824***	-0.7493 ***
SD	-0.3807***	0.6342***	-0.4568***	0.7305***
Salinity	0.1172	-0.3752***	0.1406	-0.4322***
Temperature	-0.2070	-0.5802***	-0.2483	-0.6684 ***

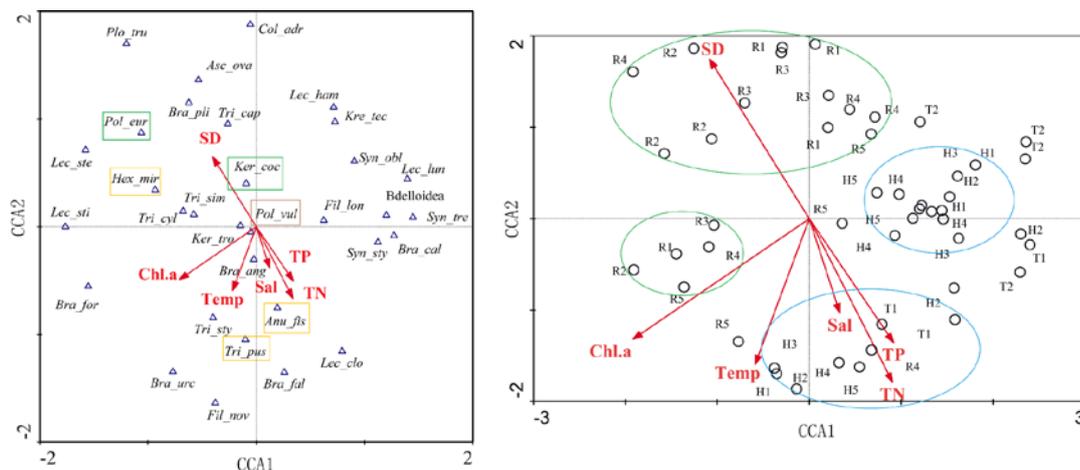
\* $p < 0.05$ ; \*\* $p < 0.01$ ; \*\*\* $p < 0.001$ . SPEC AX1: species axis 1; SPEC AX2: species axis 2; ENVI AX1: environmental factors axis 1; ENVI AX2: environmental factors axis 2.

**Table 5.** Summary statistics for the axes of CCA performed on dominant species

Axes	CCA1	CCA2	Total variance
Eigenvalues	0.223	0.213	1
Species-environment correlations	0.834	0.868	
Cumulative percentage variance: of species data	16.9	14.2	
Cumulative percentage variance: of species-environment relation	76.8	90.1	
Sum of all eigenvalues			1
Sum of all canonical eigenvalues			0.82

From the perspective of species with high contribution rate of community difference,

*Polyarthra vulgaris* stands in the middle of the figure, indicating that it had a wide niche distributed in various sites and a strong adaptability to various environmental factors. *Keratella cochlearis* was positively related to SD, prevailing in RE water bodies with better quality and higher transparency. *Hexarthra mira* was positively related to SD and chlorophyll-a, dominating in RE, the lentic water with higher primary productivity. On the contrary, *Anuraeopsis fissa* and *Brachionus angularis* were positively related to temperature, TN and TP, prevailing in HD, the lotic water with higher concentration of TN and TP (Fig. 6).



**Fig. 6.** Canonical Correlation Analysis (CCA) of rotifer assemblages with environmental variables. Relationship between species and environmental variables (Left). Relationship between sampling site and environmental variables (Right). Abbreviations used in the figures: H, Han River downstream (blue circle); R, reservoir (green circle); T, tidal creek; *Bra\_cal*, *Brachionus calyciflorus*; *Bra\_urc*, *B. urceolaris*; *Bra\_ang*, *B. angularis*; *Bra\_pli*, *B. plicatilis*; *Bra\_fal*, *B. falcatus*; *Bra\_for*, *B. forficula*; *Anu\_fis*, *Anuraeopsis fissa*; *Pol\_vul*, *Polyarthra vulgaris*; *Pol\_eur*, *P. euryptera*; *Fil\_nov*, *Filinia novaezealandiae*; *Fil\_lon*, *F. longiseta*; *Ker\_coc*, *Keratella cochlearis*; *Ker\_tec*, *K. tecta*; *Ker\_tro*, *K. tropica*; *Lec\_clo*, *Lecane closterocerca*; *Lec\_sti*, *L. stichaea*; *Lec\_ste*, *L. stenroosi*; *Lec\_ham*, *L. hamata*; *Lec\_lun*, *L. Luna*; *Hex\_mir*, *Hexarthra mira*; *Tri\_pus*, *Trichocerca pusilla*; *Tri\_sim*, *T. similis*; *Tri\_sty*, *T. stylata*; *Tri\_cyl*, *T. cylindrica*; *Tri\_cap*, *T. capucina*; *Asc\_ova*, *Ascomorpha ovalis*; *Syn\_sty*, *Synchaeta stylata*; *Syn\_tre*, *S. Tremula*; *Syn\_obl*, *S. oblonga*; *Col\_adr*, *Colurella adriatica*; *Plo\_tru*, *Ploesoma truncatum*.

## DISCUSSION

Rotifer community structure and the Carlson's trophic state index have both been used to assess water quality. For example, May and O'Hare (2005) advocated that rotifer total abundance is a more sensitive indicator of lake trophic status than species composition. Based on the rotifer

abundance in relation to trophic state, 500–1000 ind. $\cdot$ L<sup>-1</sup> is considered characteristic of mesotrophic or mesoeutrophic lakes, 1000-2500 ind. $\cdot$ L<sup>-1</sup> eutrophic, and 3000-4000 ind. $\cdot$ L<sup>-1</sup> moderately eutrophic (Ji et al. 2013; Wen et al. 2011). By these criteria, all of the three water bodies we sampled can be considered as oligotrophic due to their low density. However, according to Sladeczek's Q B/T quotient (B/T), values less than 1.0 mean oligotrophic condition, values between 1.0 and 2.0 reflect mesotrophic and values over 2.0 indicate eutrophic conditions (Sládeček 1983). By these criteria, RE can be considered as oligotrophic while HD can be considered as mesotrophic. This result was consistent with the KIN values. A KIN values less than 0.2 means oligotrophic condition, values between 0.2 and 0.3 reflect mesotrophic, and values over 0.3 indicate eutrophic conditions (Gopko and Telesh 2013). Under these definitions, RE can be considered as oligotrophic while HD can be considered as mesotrophic. The Carlson's trophic status of HD in each season was higher than that in RE, which was in accord with the results of B/T and KIN values.

On the other hand, the rotifer abundance of the three water bodies decreased in the order: RE > HD > TC. As TSI<sub>ROT</sub> consists of six different elements including rotifer abundance, it followed the same order with the rotifer abundance. Moreover, the TSI<sub>ROT</sub> indicated that all of the three water bodies were at mesoeutrophic (Jurczak 2018). The rotifer abundance is a useful indicator of lake trophic state within a water body (May and O'Hare 2005; Wen et al. 2011), but it seems that abundance is less useful in comparing the trophic status among different water bodies because of the effects of hydrological conditions. Since most rotifers live in freshwater ecosystems and salinity is negatively correlated with rotifer abundance, fewer rotifers occur in brackish water (Bielańska-Grajner and Gładysz 2010). Thus, it is more efficient to use rotifer composition including B/T, KIN and TSI<sub>ROT</sub> indices in water quality assessment when comparing different water bodies.

Difference of rotifer communities among the three water bodies were mainly due to the abundances of dominant species. Our results showed that *K. cochlearis*, *A. fissa* and *P. vulgaris* greatly contributed to the differences of rotifer communities between RE and HD. This phenomenon can be attributed to the variation of trophic conditions. Lentic waters like reservoirs may be more suitable for the growth of phytoplankton compared to the lotic waters like the Han River downstream, suggesting that biomass of phytoplankton plays a more important role in influencing the rotifer community structure, especially in lentic water (Portinho et al. 2016). According to our results, the concentration of chlorophyll-a was higher, while the concentration of total phosphorus and total nitrogen were lower in the reservoir sites. The average abundance of

rotifers was the highest in the reservoirs, and total abundances were significantly correlated with chlorophyll-a, an indicator of algal abundance, suggesting that rotifers strongly depend on phytoplankton in lentic waters such as reservoirs. Other studies have found that chlorophyll-a is a major factor affecting the rotifer community (Baião and Boavida 2000). Chlorophyll-a in turn can be influenced by phosphorus, which can thus indirectly contribute to the variation of rotifer community structure (May et al. 2014).

Our CCA analysis indicated that *K. cochlearis* tended to dominate in reservoirs with higher transparency and better phytoplankton growth, while Han River downstream, with higher nitrogen and phosphorus levels, was dominated by *A. fissa* and *T. pusilla*. The results presented in this paper are consistent with the results obtained by Wang et al. (2010), who found that *A. fissa*, *T. pusilla* and *B. angularis* dominated in the eutrophic Pearl River.

The variation of rotifer dominance can be attributed to their food collection and selection. *A. fissa* is considered to be a microfilter-feeder, mostly feeding on bacteria-laden detritus particles and nanophytoplankton in suspension (*i.e.*, particles  $\leq 10 \mu\text{m}$ ) (Špoljar et al. 2005). According to trophi and the size of food items, *B. angularis* is classified as fine particle sedimentators, eating food particles less than  $5 \mu\text{m}$  with malleate trophi (Fontaneto et al. 2008; Virro et al. 2009). In addition, *A. fissa* and *T. pusilla* are eutrophication indicators with stronger tolerance to pollution and may therefore dominate in eutrophic water (Wang et al. 2010). Our results also showed that *A. fissa* and *B. angularis* were correlated to the abundance of dissolved organic matter. In moderate flowing water, the suspended matter and bottom sediments are brought to the upper layers, thereby increasing contents of nutrient and biogenic substances, and decreasing water transparency at the same time. The high total nitrogen and phosphorus content in Han River downstream provides abundant dissolved organic detritus and bacteria as food for the microfilter-feeders, which are dominant in rivers. *P. vulgaris* was classified as small raptors who catch food actively in grasping, pumping and piercing actions with virgate trophi (Fontaneto et al. 2008; Obertegger et al. 2011). *P. vulgaris* is strongly adaptable to various environmental factors, and is widely distributed in various types of water. As it feeds on microalgae, better food conditions in RE lead to higher abundances than were observed in HD. Our results suggest that the percentage of species that indicate high trophic state can be a sensitive indicator in water quality assessment when comparing different water bodies.

Stepwise multivariate regression analysis showed that there was a significant correlation

between  $\alpha$  diversity and environmental factors (including salinity, temperature, dissolved oxygen and total nitrogen), indicating that  $\alpha$  diversity was more strongly affected by physical factors than by chemical factors. Salinity (Sal) was negatively correlated with  $\alpha$  diversity, while dissolved oxygen (DO) positively correlated with  $\alpha$  diversity. These results are consistent with that obtained by Bielańska-Grajner and Gładysz (2010); in their study, sewage disposal areas with high Sal had fewest species and lowest abundances. Higher DO was available for the respiration of a large quantity of rotifers. However, the small  $R^2$  suggested that it is not appropriate to consider only one single factor. Hydrological conditions strongly influence the physical parameters of water bodies, including SD, DO and Sal.

The differences of  $\alpha$  diversity in our results can be explained by the intermediate disturbance hypothesis (IDH) (Connell, 1978). Studies on floodplain lakes have shown that constant flow corresponds to frequent disturbance, preventing the communities from developing into climax communities and allowing for a higher biodiversity (Amoros and Bornette 2002; Goździejewska et al. 2016). Since the velocity of flow (Schöll 2011) and salinity represent the strength of disturbance, the pelagic zone in fresh water reservoirs are lentic, the absence of flow corresponds to the lowest level of disturbance. On the contrary, TC not only receives strong flow but also the highest salinity, which restricts most of the rotifer species from thriving. In HD, the consistent flows with lower salinity represent the intermediate hydrological disturbance, resulting in the highest diversities of rotifers. This disturbance was not strong enough to depress rotifer reproduction and sufficiently maintain an intermediate disturbance, which led to more diverse assemblages. On the one hand, moderate disturbance prevents some strongly competitive species with a stronger tolerance like *P. vulgaris* and *K. cochlearis* from complete dominance, and provides more niche support for the less competitive species to avoid extinction. On the other hand, water flows could be likened to a mixer, which brings the sedimentary organic detritus and benthic rotifers such as *Lecane* and bdelloids to the upper layers, enhancing the species evenness and diversity of the rotifer assemblages.

## CONCLUSIONS

Hydrological heterogeneity is the macro-factor that regulates rotifer community structures in the Shantou City area. HD presented the highest  $\alpha$  diversity, which was likely due to the

intermediate disturbance of hydrological conditions. RE and TC can be considered as oligotrophic while HD as mesotrophic. It is necessary to combine the rotifer community structure and the Carlson's trophic state index in water quality assessment. Rotifer abundance alone failed in comparing the trophic status with different water bodies because of the effects of hydrological conditions. It is more efficient to use rotifer composition including B/T, KIN and TSI<sub>ROT</sub> indices in water quality assessment when comparing different water bodies.

**Acknowledgments:** We thank National Natural Science Foundation of China (41673080) for financial support. Logistic support of Shantou Water Bureau and W. Z. Chen from Shantou University is gratefully acknowledged. The paper benefited greatly from editing of the scientific language by Senjie Lin and George B. McManus (University of Connecticut, USA).

**Authors' contributions:** Yufeng Yang conceived and designed the research. Diwen Liang carried out the experiment and wrote the manuscript, and other authors revised the manuscript.

**Competing interests:** DL, NW, QW, CJ, XH and YY declare that they have no conflict of interest.

**Availability of data and materials:** We do not want to open up our data.

**Consent for publication:** Not applicable.

**Ethics approval consent to participate:** Not applicable.

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