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# Responses of macrobenthic communities to patchy distributions of heavy metals and petroleum hydrocarbons in sediments: A study in China's Zhoushan Archipelago

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**Abstract**

This study conducted four cruises during 2014–2017 to investigate relationships between macrobenthic communities and sediment contaminations in sea area around the Zhoushan Archipelago. Fourteen sites were categorized into three groups: high total heavy metal contamination content (HHMC), high total petroleum hydrocarbon content (HTPH), and low content ratio of heavy metal contamination content to total petroleum hydrocarbon content (HMC/TPH) areas. Four main taxa of macrofauna (polychaetes, bivalves, gastropods, and crustaceans) were determined to respond to environmental factors differently. While tolerant polychaetes being the minimal impact by environmental factors, bivalves were threated by heavy metal pollutions in sediment. Additionally, body size distribution frequency demonstrated that macrofauna in the low HMC/TPH areas were less disturbed by contamination than those in the HHMC and HTPH areas. The result represented the presentation of sensitive species while tolerant species are usually considered as small size organisms. Overall, this study confirmed the hypothesis that the contamination levels of small-scale patches is indicated by the condition of macrobenthic communities.

**Key words:** macrobenthos, heavy metal, petroleum hydrocarbon, contaminant effect, Zhoushan Archipelago

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## 1 Introduction

Sediments in coastal zones are typically dominated by terrigenous material derived through aeolian and alluvial processes (Delta, 1990). It causes the accumulation of land-derived contaminants (Bryan and Langston, 1992). Therefore, coastal sediments usually act as the sink and source of contaminants in aquatic systems (Rowlatt and Lovell, 1994; Mucha et al., 2003). This makes these sediments an essential source of information in major marine monitoring programs. Heavy metals and petroleum hydrocarbons are considered as the major anthropogenic contaminants in coastal sediments; their contents tend to increase with industrial development. The high toxicity of the aforementioned metals and hydrocarbons can severely damage organisms from the cellular level to the population level (Fernandes et al., 1997; Gopalakrishnan et al., 2008). Contents of heavy metals and petroleum hydrocarbons vary considerably through the coastal zone, primarily due to differences in sediment types and input from anthropogenic contaminants (Gao and Chen, 2008; Bastami et al., 2014; Fang et al., 2016). Thus, a coastal area can be considered as a discrete habitat, whose inhabitants are differentiated by contaminant levels and ecophysiological boundaries. The benthic communities living in such vulnerable ecosystems exhibit less diversity but high resistance to changes in such extremely variable environment (Dauvin, 2007).

Macrobenthic organisms play a major role in nutrient (Braeckman et al., 2014) and carbon cycling (van Oevelen et al., 2006; Hunter et al., 2012). Moreover, some macrobenthic species are a protein source for human beings (Frid and Caswell, 2015). Macrobenthic organisms use sediment as their habitat and food source, and thus, their biodiversity tends to be lower and their community structure tends to be altered by sediment contamination (Johnston and Roberts, 2009). The properties of macrobenthos, such as deposit feeding and limited mobility, make them a cost-effective environmental indicator (Thompson and Lowe, 2004). Macrobenthos have been used as bioindicators to assess the ecological status of water systems in many monitoring programs globally (Thompson and Lowe, 2004; Van Hoey et al., 2010; Parmar et al., 2016). Although many macrobenthic species remain undescribed, some groups, such as amphipods and polychaetes, have been used as environmental stress indicators (Giangrande et al., 2005).

Heavy perturbation caused by heavy metals and petroleum hydrocarbons typically results in changes in benthic community

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structure, abundance, and biomass (Dauer et al., 2000; Thompson and Lowe, 2004; Cibic et al., 2012). The Pearson and Rosenberg model describes benthic responses to sediment contamination, particularly how the number of tolerant taxa increase, and the number, abundance, and biomass of contamination-sensitive taxa relationships decrease when perturbation levels increase (Pearson and Rosenberg, 1978). Mechanisms underlying the model descriptions have been proposed; for instance, more contaminated sediment has more organic material, which provides nutrition for contaminant-tolerant species. However, even the abundance and biomass of tolerant species decline, and they eventually die after their toxic thresholds are exceeded. In addition, a benthic community subject to pollution exhibits a decrease in mean species size with infauna occupying the superficial sediments (Weston, 1990).

To test the hypothesis that the condition of macrobenthic communities indicate contamination levels in small-scale patches, this study investigated the ecological response of benthic macrofauna to contamination by assessing them in patches of contamination generated by contents of heavy metals and petroleum hydrocarbons in sea area around the Zhoushan Archipelago, China. This study aimed to (1) assess the contamination of sediment by heavy metals and petroleum hydrocarbons in sediments and (2) analyse the response of macrobenthos to sediment contamination. In addition, diversity indices suitable for environmental assessment were determined.

## 2 Materials and methods

### 2.1 *Study site*

The Zhoushan Archipelago, comprising 1 390 islands, is located at the mouth of Hangzhou Bay, East China Sea. Some of the most densely populated and industrialized cities in China, such as Shanghai, Hangzhou, and Ningbo, are within 150 km of it. The Zhoushan Harbor is one of the most important and busiest harbors in China. Crude petroleum and ore are stored at this temporary base. Contaminants leaking from storage depots and maritime transports have accumulated in sediments in areas around the nearby sea, with such accumulation causing severe marine environmental crises (Ma, 2012). Environmental conditions in the sea area around the Zhoushan Archipelago have considerably deteriorated over the past two decades (Xu et al., 2015). Several studies have assessed the contamination of water (Liu et al., 1991), sediment (Dong et al., 2012; Hu et al., 2009; Wang et al., 2015) and biota (Shou et al., 2009; Wang et al., 2014). These studies have revealed that the grain size (Zhuo et al., 2019), organic matter and C/N ratio (Xu et al., 2016) in sediment typically appear to be homogeneous throughout the sea area around the Zhoushan Archipelago. However, none of these studies have analyzed the effect of marine contamination on the distribution of organisms at the community level.

### 2.2 *Sampling*

Macrobenthos were collected during four cruises in July and November 2014, May 2016, and February 2017. Each cruise visited approximately 27 stations around several main islands of the Zhoushan Archipelago (Fig. 1), and two replications were collected using a Van Veen grab sampler (surface area: 0.10 $m^2$) at each station. With the exception of the cruise in February 2017, the other cruises collected sediment. Approximately 100 g of sediment scraped from the sea floor of each station was added to a clean polythene bag and kept in a deep-freezer for subsequent chemical analyses. The remaining sediments were washed through a 0.5 mm mesh sieve. All residues on the sieve were fixed in a 10% formalin solution. The general environmental parameters, such as pH, salinity, and temperature of bottom seawater conditions were measured simultaneously using a dissolved oxygen meter and a water quality meter, and approximately 500 mL bottom water samples were collected from each station and analyzed in a laboratory to determine their suspended solids (SS)

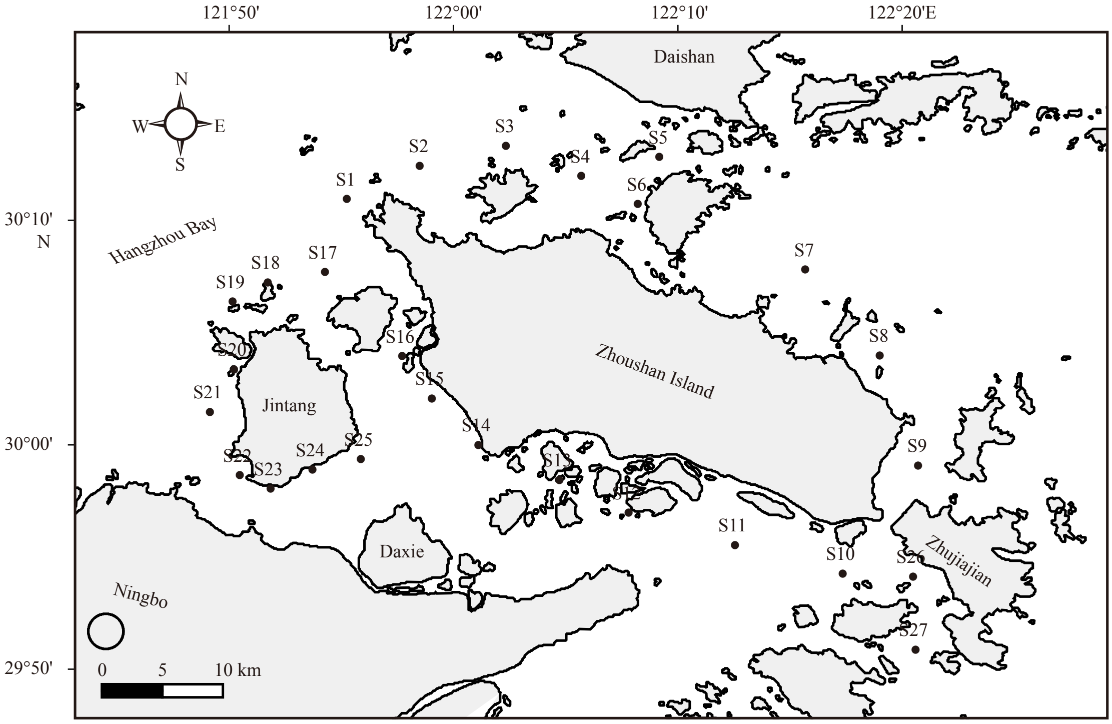

**Fig. 1.** Map of the 27 sampling locations in the sea area around the Zhoushan Archipelago.

and chemical oxygen demand (COD) properties.

### 2.3 *Laboratory analysis*

In the laboratory, macrobenthos were identified, sorted, and counted by their lowest practical taxonomic level. Unidentified taxa were considered only if they could not be mistaken for other identified taxa. All identified specimens were confirmed by at least 2 macrobenthic taxanomist. For analyzing of the biomass and body size distribution of individual specimens, only intact animals were selected for weighing—after being blotted with absorbent paper. This study used an electronic precision balance (Sartorius, BSA, Germany) accurate to 1 mg. Fragments of specimens were collectively weighed to estimate the total biomass at each station. Biomass measurements are represented in terms of wet weight. Before being weighed, polychaetes were removed from their tubes. However, the calcified structures of mollusks and crabs were included.

Bottom water was filtered through 0.45 μm GF/F glass filter paper to collect SS. SS were subsequently weighed using an electronic analytical balance (Mettler Toledo, MX5, USA). The COD in bottom water was measured using the $\mathrm{COD_{OH}}$ method (Goh and Lim, 2008). The total organic carbon (TOC) in sediments was estimated using a TOC analyzer (Shimadzu, TOC-L, Japan). Cd, Cr, Cu, Pb, and Zn contents in sediments were measured using a flame atomic absorption spectrophotometer (Perkin Elmer, AA-800, USA) after digestion with a mixed solution of acids. As and Hg contents were evaluated using an atomic fluorescence spectrometer (Skyray Instrument, AFS200, China). Petroleum hydrocarbon content was measured using a fluorescence spectroscope (Shimadzu, RF-6000, Japan). All instruments were calibrated before use. Heavy metal and petroleum hydrocarbon contents are expressed as milligram per kilogram of dry weight.

### 2.4 *Data analysis*

To quantify the biodiversity of macrobenthic communities, two community ecological indices, species richness ($S$) (Barrio Froján et al., 2008) and Shannon–Wiener diversity ($H'$) (Shannon and Weaver, 1964) were calculated. One-way ANOVA analysis of variance (ANOVA) based on abundance was used to compare differences among the microbenthic communities in different seasons.

To represent the level of heavy metal contamination, the potential ecological risk index (RI) was calculated. The RI can be expressed as follow:

$$C_f^i = C_i / C_n^i, \tag{1}$$

$$\mathrm{RI} = \sum_{i=1}^{n} T_i C_f^i, \tag{2}$$

where $T_i$ is the toxic coefficient of the various heavy metal: for Zn is 1, for Cr is 2, for Cu and Pb is 5, for As is 10, for Cd is 30, and for Hg is 40; and $C_i$ is the dry weight of these various heavy metal in the sediment (Hakanson, 1980). $C_f^i$ is the pollution index of each heavy metal; $C_n^i$ is the background contents of heavy metals in the sediment of the East China Sea (Ji, 2011), as shown in Table 1.

**Table 1.** Background contents of heavy metals in sediment (Ji, 2011)

| Heavy metal | Cu | Pb | Zn | Cd | Cr | As | Hg |
|---|---|---|---|---|---|---|---|
| Background content/(μmol·kg$^{-1}$) | 95.12 | 47.29 | 686.15 | 0.18 | 223.08 | 55.07 | 0.04 |

Analysis of similarities (ANOSIM) was used to test the hypothesis that there is a significant difference among groups with different levels of contamination. Nonmetric multidimensional scaling (MDS) ordination was used to represent sampling locations in two-dimensional space in terms of species composition and abundance. The same similarity matrix for ANOSIM, cluster, and nonmetric MDS was calculated using Bray-Curtis similarity; abundance data were transformed to their fourth root. The preference of macrobenthic taxa to environmental parameters was tested using canonical correspondence analysis (CCA). Before CCA analysis, variance inflation factors (VIF) of each variable were calculated. Variables with VIF higher than 10 were removed from CCA. One-way ANOVA was conducted using SPSS (IBM, version 20.0, USA); ANOSIM, cluster, and MDS were conducted using PRIMER 6 (PRIMER-e Ltd, UK); and CCA was conducted using Canoco 5 (Microcomputer Power, USA).

## 3 Results

### 3.1 *Contaminant content in sediment*

The heavy metal contamination contents (HMC) of sediments ranged from 3 978.49 μmol/kg to 2 047.03 μmol/kg. Stations S17 and S18 had the highest seasonal average HMC values. The total heavy metal contamination content (THM) of Station S17 reached its maximum in July 2014. Station S9 had the lowest seasonal THM value, reaching its minimum on July 2014. The contents of major elements were similar in different seasons. Of the HMC, average contents of Zn, Cr, Cu, As and Pb were (1 551.86±376.26) μmol/kg, (949.09±97.17) μmol/kg, (418.95±64.01) μmol/kg, (172.15±41.97) μmol/kg, and (115.63±15.39) μmol/kg, respectively, and those of Cd and Hg were less than 1.5 μmol/kg. Regarding to contents in different seasons, of the THM, Zn, Cr, Cu, As and Pb contents varied from 1 500.45 μmol/kg to 1 615.82 μmol/kg, 932.47 μmol/kg to 960.73 μmol/kg, 404.93 μmol/kg to 425.08 μmol/kg, 161.57 μmol/kg to 175.88 μmol/kg, and 114.50 μmol/kg to 116.88 μmol/kg, respectively.

The total petroleum hydrocarbon content (TPH) of sediments ranged from 1.42 mg/kg to 25.73 mg/kg. Stations S11, S12, S13 and S14 had the highest seasonal average TPH value. The TPH in Station S13 reached its maximum value on November 2017. Stations S18 and S19 had the lowest seasonal average TPH values, reaching its minimum values in May 2016.

The classification of high RI was based on the criteria suggested by Wang et al. (2014) while the low RI was based on Jiang et al. (2011), but Jiang et al. (2011) only studied five heavy metals, so this study raised the criteria of low RI; classification of petroleum hydrocarbons was based on UNEP (1992); specifically, sites with a seasonal RI higher than 440 were categorized as a high HMC (HHMC) areas. Sites with a seasonal TPH higher than 15 mg/kg were categorized as high TPH (HTPH) areas, and sites with seasonal RI lower than 360 and seasonal TPH lower than 8 mg/kg were categorized as low HMC/TPH areas. The HHMC area comprised Stations S1, S2, S5, S14 and S18; the HTPH area comprised Stations S11, S12, S13, S14 and S20; the low HMC/TPH area comprised Stations S3, S9, S19, S26 and S27. The RI, TPH and TOC values for each area are displayed in Table 2.

**Table 2.** Contaminants measured in surface sediments in the sea area around the Zhoushan Archipelago

| Area | RI range | RI mean | TPH range/(mg·kg$^{-1}$) | TPH mean/(mg·kg$^{-1}$) | TOC range/% | TOC mean/% |
|---|---|---|---|---|---|---|
| Overall | 245.73–491.61 | 400.56±57.90 | 1.42–25.73 | 9.63±6.05 | 0.59–1.75 | 1.00±0.26 |
| HHMC | 458.13–491.61 | 476.49±11.35 | 1.64–20.17 | 7.35±6.65 | 0.94–1.36 | 1.09±0.12 |
| HTPH | 342.58–484.03 | 391.86±46.68 | 14.47–25.73 | 18.41±3.72 | 0.67–1.44 | 0.89±0.32 |
| Low | 308.56–354.94 | 337.68±18.76 | 2.29–7.26 | 4.95±2.02 | 0.70–1.73 | 1.01±0.40 |

### 3.2 *Macrobenthic assemblage*

Over the seasonal survey, 842 individual macrobenthos, belonging to 108 marine taxa, were enumerated. Polycheates were predominant among the macrobenthos, comprising 48.1% of the total assemblage, followed by crustaceans (28.7%), mollusks (12.0%), others (7.4%), and echinoderms (3.7%).

The mean overall site abundance was 72 ind./m$^2$ (5–230 ind./m$^2$) and that of site biomass was 1.05 g/m$^2$ (0.01–9.83 g/m$^2$) (Table 3). Biomass had more outliers than the other parameters (Fig. 2), likely due to the numerous phyla to which the macrofauna in sea area around the Zhoushan Archipelago, and it implies a wide range in individual weights.

The most dominant species were defined by both their abundance and occurrence frequency. Eight species with the highest abundances, and enumerated in more than two seasons, were defined as dominant species. They accounted for 63.1% of the total macrofaunal abundances. Six of these species were polychaetes: *Micronephthys oligobranchia*, *Sternaspis scutata*, *Amaeana occidentalis*, *Euclymene lombricoides*, *Heteromastus filiformis*, and *Goniada japonica*. Several of these dominant polychaetes, particularly *S. scutata*, *E. lombricoides*, and *H. filiformis* are considered typical tolerant species. Of the other two domin-

**Table 3.** Quantitative characteristics and significance of seasonal variation of the macrobenthos within sea area around the Zhoushan Archipelago

| Area | Abundance range/(ind.·m$^{-2}$) | Abundance mean/(ind.·m$^{-2}$) | Biomass range/(g·m$^{-2}$) | Biomass mean/(g·m$^{-2}$) | One-way ANOVA | |
|---|---|---|---|---|---|---|
| | | | | | *F* | *p* |
| Overall | 5–230 | 72±61 | 0.01–9.83 | 1.05±1.90 | 5.530 | 0.2 |
| HHMC | 10–230 | 84±85 | 0.02–6.44 | 1.10±1.78 | 5.298 | 1.1 |
| HTPH | 10–215 | 66±60 | 0.01–9.09 | 1.86±2.94 | 1.689 | 21.8 |
| Low | 30–210 | 94±51 | 0.21–9.83 | 2.74±3.56 | 14.601 | 0.2 |

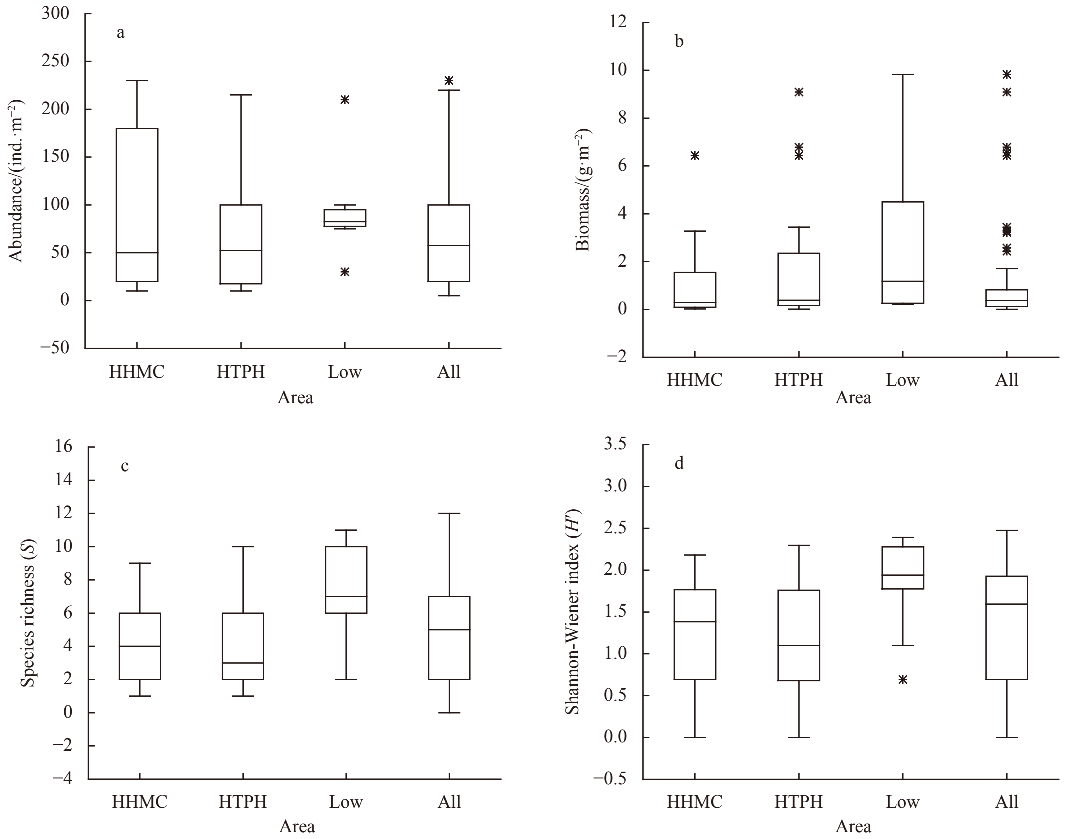

**Fig. 2.** Boxplots illustrating the range for values of selected benthic macrofaunal assemblage parameters recorded in sea area around the Zhoushan Archipelago. Measured parameters include abundance (a), biomass (b), species richness (c), and Shannon-Wiener index (d).

ant species, one was a crustacean (*Apseudes nipponicus*), and the other was a mollusk (*Ennucula niponica*).

### 3.3 *Macrobenthos under different levels of contamination*

Univariate analyses revealed that during summer, assemblages were significantly dissimilar between sampling areas (*R*=0.102, *p*=0.04) at significant level 0.05. An MDS plot of samples from all three areas over all four seasons also revealed that compared with those taken from a low HMC/TPH area, the replicates taken from the HHMC area had tighter clustering (Fig. 3). Although samples taken from each area overlap on the MDS plots, statistical analysis revealed that the assemblages from the HHMC and low HMC/TPH areas were significantly dissimilar in both spring (*R*=0.45, *p*=0.04) and autumn (*R*=0.63, *p*=0.02; Table 4) at significant level 0.05. No significant dissimilarity was revealed between the HTPH area and the other two areas, which possibly indicating that the macrofauna in the HTPH area were in transition from original fauna to altered fauna. The cluster results also confirmed differences in the macrofauna of the HHMC area with those of the low THM/HPH area (Fig. 4). According to cluster results, data from stations in the HTPH area and HHMC area did not obviously differ (Fig. 4). ANOSIM and cluster analysis furnished similar results.

Opportunistic species are generally characterized by small body sizes, thus, macrobenthic communities in polluted areas are typically exhibit relatively small animal sizes. The size frequency of HHMC and HTPH areas had a similar single peak distribution, whereas the low HMC/TPH area had a second peak in Class 4 (Fig. 5). The frequency distribution indicates that a relatively high proportion of large-sized individuals was presented in the low HMC/TPH area.

The four main taxa of macrofauna responded to environment factors differently (Fig. 6). Polychaetes exhibited superior environmental endurance, with no obvious preference for particular environment parameters. Bivalves tended to be distributed proximate to SS—their main food source—in bottom water and heavy metals in the sediments. By contrast, gastropods completely avoided the sediments contaminated by heavy metals and petroleum hydrocarbons. Crustaceans were not influenced by the contents of heavy metals or petroleum hydrocarbons, but they were sensitive to salinity.

## 4 Discussion

The contents of heavy metal contaminants in sediments around the Zhoushan Archipelago were close to the results of investigations in similar bays and coastal zones (Fang et al., 2013; Bastami et al., 2014). Ecological risk assessments reported that the total risk to ecosystem from heavy metals in surface sediment were moderate to high (Chai et al., 2015). However, historical data since 1996 reported increasing contents of heavy metals in the study area (Chai et al., 2015). The lack of historical data on contents of petroleum hydrocarbons in this area makes estimat-

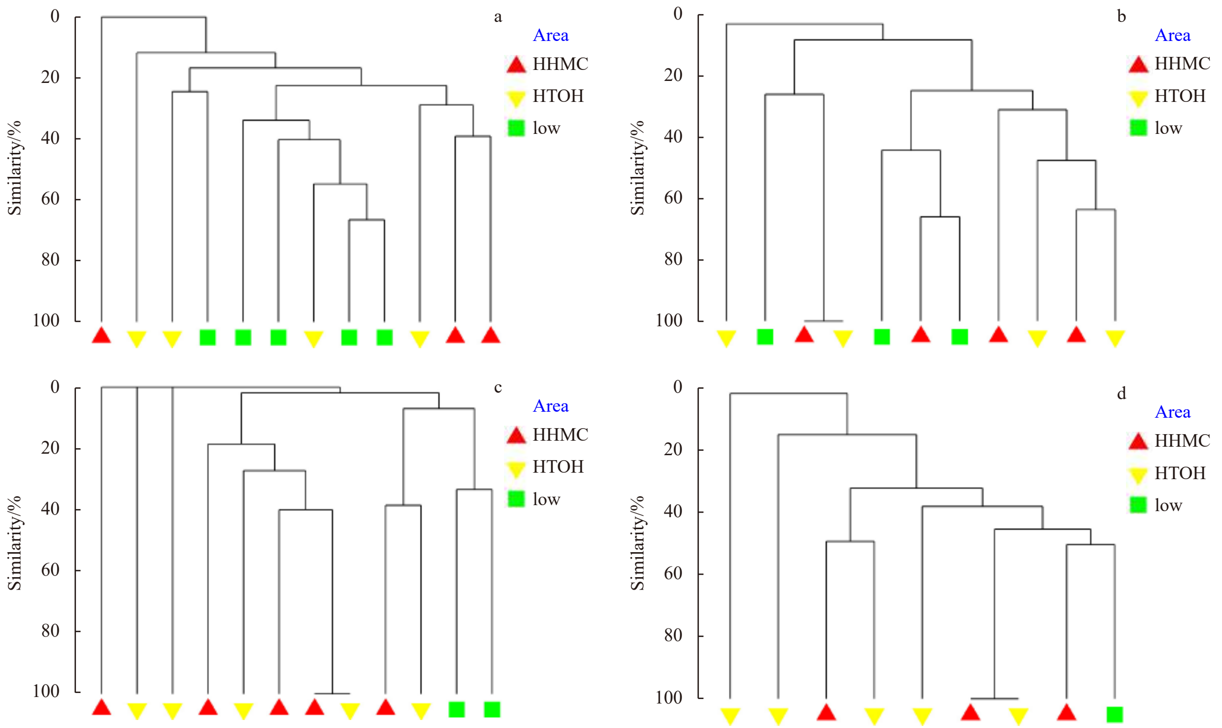

**Fig. 3.** Season clusters of Bray-Curtis similarity of assemblage samples from three areas: a. spring, b. summer, c. autumn, and d. winter (macrofaunal data transformed to their fourth root were used).

**Table 4.** Results of comparing community structure of macrobenthos from three areas over four seasons

| Area | Spring | | Summer | | Autumn | | Winter | |
|---|---|---|---|---|---|---|---|---|
| | *R* | *p* | *R* | *p* | *R* | *p* | *R* | *p* |
| HHMC & HTPH | 0.18 | 0.20 | −0.07 | 0.66 | −0.01 | 0.91 | −0.32 | 1.00 |
| HHMC & Low | 0.45 | 0.04 | −0.24 | 0.86 | 0.63 | 0.02 | −0.11 | 0.75 |
| HTPH & Low | 0.04 | 0.42 | −0.06 | 0.60 | 0.18 | 0.24 | −0.56 | 1.00 |

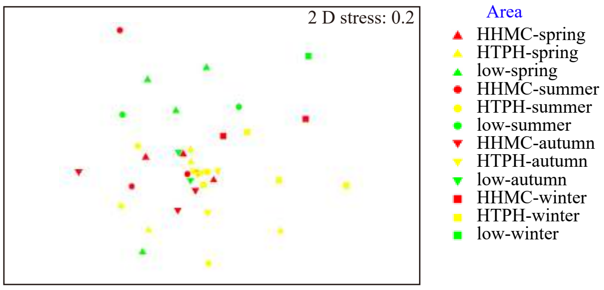

**Fig. 4.** Nonmetric multidimensional scaling ordination from three areas over four seasons in terms of species composition and abundance (macrofaunal data transformed to their fourth root were used).

ing the current level of contamination difficult. However, similar studies have suggested a higher rate of potential aggregation of petroleum hydrocarbons in this area from human activity, compared with the offshore area around the East China Sea (Li et al., 2005). Nevertheless, the general condition of hydrocarbon contamination in the sea area around the Zhoushan Archipelago is not as severe as that in the other nearshore area of China (Gao and Chen, 2008; Wang et al., 2015).

Environmental parameters of sediment such as grain size, organic matter, and C/N content ratio may also influence the distribution of macrobenthos. The grain size of sediment in the sea area around the Zhoushan Archipelago was highly uniform, and most of the sediment was clay and silt (Zhang et al., 2015; Zhuo et al., 2019). A continuous trend of C/N content ratio in surface sediment was observed in the Hangzhou Bay and the adjacent sea area, but as part of the Hangzhou Bay, the sediment around the Zhoushan Archipelago showed exhibited no dramatic changes of differences in total organic carbons TOC and/or C/N content ratio (Xu et al., 2016). Therefore, the characteristic of sediment in the sea area around the Zhoushan Archipelago can be characterized as homogeneous and stable. The distribution of macrobethons macrobenthos in the sea area around the Zhoushan Archipelago is more likely influenced by contamination rather than environmental parameters. Some studies have observed continuous gradients of contaminants in the Hangzhou Bay and its adjacent sea area (Che et al., 2003; Adeleye et al., 2016). However, with regard to any specific area, measurements of such gradients tend to be disrupted by local human activities. The results of this study indicate that the high contents of heavy metals and petroleum hydrocarbons had a patchy distribution through the sea area around the Zhoushan Archipelago. The Zhoushan Archipelago is one of the largest harbors in China, with chemical factories and oil storage tanks scattered throughout the entire archipelago (Zhang and Zhu, 2013). Sediment resuspension caused by hydrodynamic forces, anthropogenic activities, and scattered contamination sources has contributed to the patchy contaminant distribution. Our ANOSIM revealed significant differences between the benthic communities in the HHMC and low THM/TPH ratio areas, but only in spring and autumn. This study discerned no statistical evidence of dissimilarity between the macrobenthic communities in the HTPH and low THM/TPH ratio areas. However, the MDS plots suggested that the HTPH area was in a transitional zone between a low THM/TPH ratio and HHMC area. The HTMP, HTPH, and low HMC/TPH areas were all discrete habitats, each comprising several nonadjacent sites. Macrobenthos frequently aggregate into clusters separated from each other (Eleftheriou and McIntyre, 2005). Moreover, their limited mobility prevents their regular migration to other habitats. Therefore, this study considers the status of macorbenthic community is a suitable biological indicator to assess the health of small-scale habitat patches.

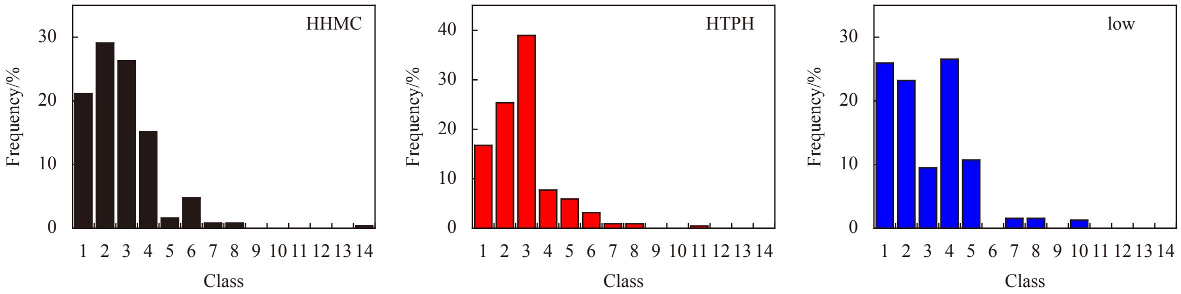

**Fig. 5.** Frequencies of various sizes of macrofauna specimens by geometric size class (Class 1, 1 mg; Class 2, 2–3 mg; Class 3, 4–7 mg; Class 4, 8–15 mg; Class 5, 16–31 mg; Class 6, 32–63 mg; Class 7, 64–127 mg; Class 8, 128–255 mg; Class 9, 256–511 mg; Class 10, 0.512–1.023 g; Class 11, 1.024–2.047 g; Class 12, 2.048–4.095 g; Class 12, 4.096–8.191 g; Class 14, 8.192–16.383 g).

A series of sites with both heavy metal contamination and organic enrichment were discovered in sea area around the Zhoushan Archipelago. Enriched organic matter in sediment provides additional food for benthic deposit-feeders; however, they must endure the concomitant content of heavy metals. Some researchers have proposed that tolerant species is expected to increase in abundance at moderate to relatively high contaminant contents (Thompson and Lowe, 2004). However, our abundance analysis of the HHMC area did not indicate potential aggregation of tolerant species. This was likely because the content level of heavy metals remained under the threshold at which biological effects become evident.

The macrofauna investigated in this study comprised taxa with high morphological diversity; moreover, their body sizes varied widely (Ryu et al., 2011). The total biomasses collected from some sampling sites was heavily influenced by a few large individuals. Two goby fish of *Odontamblyopus rubicundus* and *Amblyotrypauchen arctocephalus* accounted for 99% and 93% of the total biomass of their sites, respectively, and the two sites with gobies were also two of the three sites with the highest biomass over all four seasons. Large-sized organisms such as goby are more mobile than the small sized organisms studied. They migrate between habitats to avoid hostile environments instead of staying in one place (Monte, 2002). This supports our hypothesis that mobile macrobenthic organisms are less affected by the presence of discrete contaminated habitats in the sea area

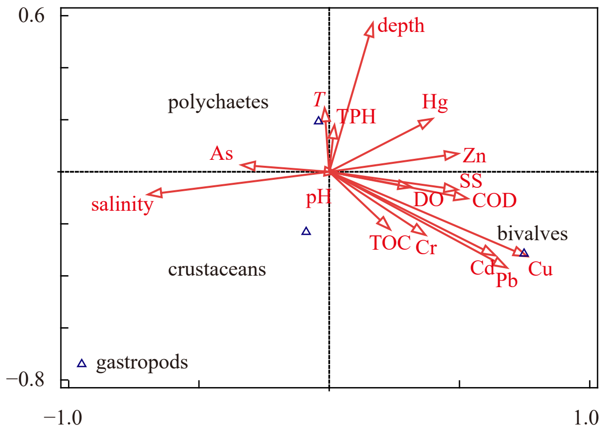

**Fig. 6.** Canonical correlation analysis of four main taxa of macrobenthos to environmental parameters in bottom water, namely depth, pH, temperature (*T*), salinity, suspended solids (SS), chemical oxygen demand (COD) and dissolved oxygen (DO) and contaminants in surface sediment, specifically total organic carbon (TOC), total petroleum hydrocarbon (TPH) and heavy metals within sea area around the Zhoushan Archipelago.

around the Zhoushan Archipelago. Therefore, an environmental assessment (particularly that of a small area) that uses the total biomass of macrobenthos may overestimate the health condition of the macrobenthic community.

The size frequency data on macrobenthic communities were used for detailed examination of the effects of pollution. Most individuals weighed 1–15 mg, and clear differences were evident between the three areas after individuals weighing larger than 15 mg were excluded from our data analysis. The HHMC and HTPH areas had the same trend, but the frequency in HTPH was more evenly distributed. Petroleum hydrocarbons can provide energy and carbon to microorganisms (Xue et al., 2015; Varjani, 2017). However, no evidence suggests that macrobenthos can use hydrocarbons, which are regarded as a common contaminant in coastal areas. The distribution of size frequencies also indicate that the content of petroleum hydrocarbon causes the degradation of a macrobenthic community, however, the ANOSIM analysis did not demonstrate this. The structure of benthic communities in the HTPH area did not change as substantially as that of those in the HHMC area. The effect of the content of petroleum hydrocarbons was observed after comparing benthic communities in the HTPH area with those in a less contaminated area. Size frequency in the low HMC/TPH area was distributed bimodally. The second peak in Class 4 indicates a considerable presence of large-sized organisms in the low HMC/TPH area (Warwick and Clarke, 1984). According to the Pearson and Rosenberg model, moderate contamination results in an ecotone overlap of environmental sensitive and opportunistic communities (Pearson and Rosenberg, 1978). Most opportunistic species are small because of selective pressures for morphological simplicity and rapid maturation (Ryu et al., 2011). Thus, the presence of large-sized groups indicates relatively healthy environmental conditions.

The responses of macrofauna in the Zhoushan Archipelago to environment parameters and levels of contamination varied among taxa. The macrofauna in the sea area around the Zhoushan Archipelago were dominated by polychaetes. Unlike the other taxa, polychaetes were not influenced by heavy metals significantly (Fig. 6). Many polychaetes were regarded as pollution tolerant species (Giangrande et al., 2005), but recent toxicity studies showed they were still disturbed by high content of contaminations (Ghribi et al., 2019; Tong et al., 2019). Although the abundance, biomass, and species richness of polychaetes in the HHMC and HTPH areas did not increase, most polychaetes in these areas belonged to typical macrobenthic families with high pollution tolerance (Rouse and Pleijel, 2001). Some dominant polychaetes, such as *S. scutata*, *E. lombricoides*, and *H. filiformis,* are considered opportunistic species. Previous study found the abundance of polychaetes usually decrease with depth (Carvalho et al., 2013), which is different from our result. This may because the coastal area of the Zhoushan Archipelago were strongly disturbed by human activities, polychaetes stayed the deep sea area to avoid anthropogenic disturbations. Temperature is another import environment factor for polychaetes. Growth rate of polychaetes increased significantly at high temperature (Yokoyama, 1988). This may interpret the polychaetes prefer to high temperature. Mollusks preferred habitats with either a high TOC in the sediments or a presence of SS in bottom water. Two main classes (bivalve and gastropod) of mollusks responded to heavy metals through various approaches, which have a primary influence on their life strategies. Most gastropods are deposit-feeders, and they avoid most heavy metals in sediment except As, which is a pervasive environmental toxicant and widely distributed in the aquatic environment due to natural or anthropogenic processes (Sharma and Sohn, 2009). But in some cases, As content detected in gastropods did not correlate strongly to metal content in bottom sediment (Loder et al., 2016), which suggested that gastropod metabolic processes could regulate the uptake and accumulation of As. Therefore, gastropods have high tolerance to As. Growth of marine gastropods could be restrained by low salinity (Pechenik et al., 2003). Sea areas around the Zhoushan Archipelago were dominated by low salinity waters, so the gastropods prefer to high salinity in this area. Bivalves primarily comprise filter-feeders that take organic particles from water, their tolerance to sediment contamination appeared superior to that of gastropods (Rumisha et al., 2012). The distributions of bivalves were correlated with SS in bottom water event. These habitats were accompanied with high heavy metals contents in sediments. DO was also important to bivalves. Bivalves absorb oxygen from the water, low DO was lethal to bivalves (Long et al., 2008). CCA demonstrated that Pb, Cd, Cr and Cu were associated with the distribution of bivalves (Fig. 6). All heavy metals could be toxic at certain contents, but Pb, Cd and Cr are very toxic even at low dose (Jović and Stanković, 2014). Cu is an essential element in trace amount for a normal growth and development for aquatic organisms, but it is potentially harmful to most of marine organisms at some level of exposure and absorption (Yılmaz et al., 2017). Therefore, the local commercial bivalves may need risk assessment because these heavy metals accumulate in bivalves are potential threats to human health. Crustaceans shared similar life strategy with gastropods for they are deposit-feeder and have relative strong mobility. Like gastropods, crustaceans avoided habitats with high heavy metal contamination and preferred high salinity. Salinity is a key parameter of crustaceans, especially to the larva (Anger, 2003). The preference to salinity might implied the crustaceans in the sea area around the Zhoushan Archipelago were dominated by brackish species, which is common within the macrobenthic community in nearshore area (Obolewski et al., 2018).

## 5 Conclusions

This study revealed that sediments with high contents of heavy metals and petroleum hydrocarbons in the sea area around the Zhoushan Archipelago are small discrete patches.

These contaminated patches influenced the community structure of macrobenthos in the sea area around the Zhoushan Archipelago. The study also revealed that the main taxa of macrobenthos exhibit specific responses to environmental factors due to their different life strategies. Bivalves are the main taxa threatened by heavy metal pollutions in sediment. Analyses of size frequency demonstrated specific differences between areas with varying levels of contamination and indicated the presence of a higher percentage of relatively large organisms in the low HMC/TPH ratio area than in the HHMC and HTPH areas, whereas the ANOSIM only indicated a difference in the spring and autumn community structures between HHMC and low HMC/TPH areas. Thus, this study suggests that researchers conduct a detailed examination of the body sizes of macrobenthos before using them as environmental indicators, particularly when assessing a small-scale area.

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