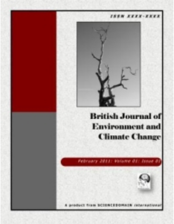

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# Are Diatom-based Indices from Europe Suitable for River Health assessment in China? A Case Study from Taizi River, Northeastern China

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### *Authors' contributions*

*This work was carried out in collaboration among all authors. Authors Qu, Yin and Zhang conducted the field trip including collecting sample of diatom and water quality. Author Yin conducted the identification of diatom. Authors Qu and Zhou designed the study and conducted the calculation of diatom-based indices. Author Zhao conducted main data analysis and modeling. The manuscript was prepared by author Qu and revised by CB. Author Qu oversaw the whole process as the research advisor. There were series of interactions among the authors to prepare and revise the manuscript.*

***Original Research Article***

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## ABSTRACT

**Aims:** Diatom-based indices are widely used for river health assessment. Many such indices were originally developed in European countries based on a specific taxa list of benthic diatoms. Thus, the transferability of these indices to other rivers and geographic locations has been questioned.

**Design, Place and Duration of Study:** In this study, we sampled benthic diatoms in the Taizi River, a temperate river in northeastern China during May 2009 to evaluate the applicability of eight commonly used diatom-based indices for assessing the principal water quality gradient and relationship with different water chemical parameters.

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**Methodology:** Sensitivities of the eight indices were evaluated by applying the principle component analysis (PCA), the box-plot map and multiple comparisons of the Kruskal–Wallis nonparametric test (K-W test).

**Results:** The results showed that all eight of the tested indices showed significant correlations with the principal contamination gradient of both nutrient enrichment and organic pollution. The contamination gradient was extracted through principal component analysis and the first three axes explained 40.19%, 18.72%, and 9.77% of the total variation, respectively.

**Conclusion:** Our results confirmed that the diatom-based indices did not properly reflect the current Chinese surface water quality classes. However, these indices showed consistent trends with chemical parameters that reflected general water quality condition, such as electric conductivity (EC); organic pollution, as reflected by dissolved oxygen (DO) and chemical oxygen demand (COD) and nutrient enrichment, as reflected by total nitrogen (TN) and total phosphorus (TP). The results indicated that both BDI and SHE were the most suitable diatom-based indices among the eight tested indices, although SPI, DES and ROTT were also suitable for river health assessment in the Taizi River.

*Keywords: Diatom-based index; BDI; river health; OMNIDIA; Taizi River.*

## 1. INTRODUCTION

Benthic algae are important aquatic biota in riverine ecosystems. As primary producers, algae provide the main food resources for macro invertebrates and fish and play an important role in energy cycling [1,2]. Due to their short life span, relatively complex community composition and high sensitivity, benthic algae can integrate both short-term and long-term environmental variation and a broad range of human stressors [3]. Thus, many monitoring programs in the United States [4] and Europe [5-7] include diatoms as main elements to assess river health.

There are several diatom-based indices that have been developed in Europe using the Zelinka and Marvan formula [8]. The indices take into account the variations of relative abundance and tolerance of each taxon in the community, and have been widely accepted as important indictors in river health assessment. The Descy Index [9] was the first diatom index developed based on the pollution tolerance classification for diatoms. The standardized Biological Diatom Index (BDI) [10] was first developed and applied in France for the surveillance of water quality. The BDI index was revised (BDI-2006) [11] for better suitability to the European Water Framework Directive (WFD). Other widely used indices include the Specific Pollution sensitivity Index (SPI) [12], Generic Diatom Index (GDI) [13], Trophic Diatom Index (TDI) [14], Commission for Economical Community Index (CEE) [15], Schiefele and Schreiner’s Index (SHE) [16] and Rott Saprobic Index (ROTT) [17].

Even though diatom indices are widely applied, there are two aspects that still present conflicts. The first is the suitability and applicability of different indices to different geographic regions [18]; the second is the sensitivity of diatoms to different kinds of contamination, such as ionic stresses, organic pollution, heavy metal contamination and eutrophication. Hering et al. [19] recommended that benthic diatom indices were better indicators to reflect eutrophication and land use gradients in European countries: the 25 tested diatom indices (e.g., SPI, SLA, DES, LMI, SHE, Watanabe index, TDI, BDI, DAIpo, CEE, GDI, Rott) showed similarly strong correlations with eutrophication/organic pollution and land use gradients.

However, there are also several examples of unsuccessfully applied indices. For example, Rott et al. [20] found the BDI may not be suitable for use in Austrian rivers and Gomà et al. [21] found the correlations between BDI and water quality variables were not significant in the upper Segre basin of the Oriental Pyrenees, Spain. Deng et al. [22] also compared seven indices of diatom sensitivity and applicability in the Dongjiang River, southeast China. The results showed that BDI and GDI were the most suitable indices for river health assessment in this area. Tan et al. [18] compared the usefulness of 14 indices in a subtropical river of central China. They identified strong, significant correlations of BDI and EPI to trophic status and ionic content, and strong correlation of Watanabe's index to organic pollution and conductivity gradients. Except for the Index Diatom Artois–Picardie (IDAP) [23,24], all other indices were suitable to reflect the water quality in the Han River, China during base-flow conditions.

Regarding sensitivity, Prygiel and Coste [25] confirmed six indices, including SPI, DES, CEE, GDI, Sládecek's saprobic index (SLA) [26] and Leclercq and Maquet's index (LMI) [27], as suitable indicators to detect the dominant stressor of organic pollution in the Artois–Picardie water basin in France. Among them, SPI, GDI and CEE also showed significant correlations with ionic strength and eutrophication. Rimet et al. [28] designed an experiment to evaluate the sensitivity of nine diatom indices by transferring the epilithic diatom biofilms from polluted rivers to an unpolluted stream. The results confirmed that five indices (SPI, TDI, CEE, Eutrophication Pollution Index [EPI] and ROTT) had higher sensitivities than the other four indices (GDI, BDI, LMI and SLA).

Currently, diatom-based indices are applied with greater frequency for river health assessment in China [29-31]. However, the assessments of suitability and sensitivity of these indices are rare for Chinese rivers [22]. Furthermore, there is still no research relating diatom indices to the national water quality standards, which currently represent the main aspect of the river health assessment method in China [32]. Biological river health assessment has become one of the most efficient management tools to date. Currently, both the Ministry of Environmental Protection and the Ministry of Water Resources of the People's Republic of China processed corresponding researching programs of river health assessment. And selecting suitable indices of benthic diatom, macro invertebrates and fish is one of the hot topics of the programs. It is also interested in for all the river management authorities in China. In this study, we conducted research to evaluate the applicability of eight diatom indices for water quality assessment; compare the sensitivity of each index to the Chinese national water quality and water contamination gradients; establish quantitative correlations between the stressor gradient and indices and establish suitable references values for each index for the temperate continental rivers in China.

## 2. MATERIALS AND METHODS

### 2.1 Study Region

The study was conducted in the Taizi River basin, which is the part of the Liao River basin located in northeastern China. Taizi River is located in the mid and high latitudes of China in a temperate, continental monsoon climate zone. It has a watershed area of $1.39\times10^4$ $km^2$ and stream length of 413 km. Main features of the local climate include a hot rainy summer, cold sunny winter, and a short spring and autumn. The annual temperature is around 7.9ºC and annual precipitation ranges from 650–950 mm across the basin [33]. Taizi River basin includes nine main tributaries and supplies drinking water for 5.5 million people as well as water for domestic, industrial and irrigation purposes.

Human disturbances are unevenly distributed from upstream to downstream. Forest dominates the main land cover in the eastern upper stream regions, while agriculture and urban areas dominate the main land uses in the middle and western downstream regions (Fig. 1) [34]. Across the whole watershed, the key threats to river health include clearing of riparian vegetation, construction of in-stream barriers and dams and in-stream extraction of sand and gravel [35]. In the middle and downstream areas, urban and rural non-point source pollution and industrial point source discharge have caused severe contamination of water quality in the Taizi River since the 1950s. Investments have been made for riverine restoration and protection during the last decade to improve water quality.

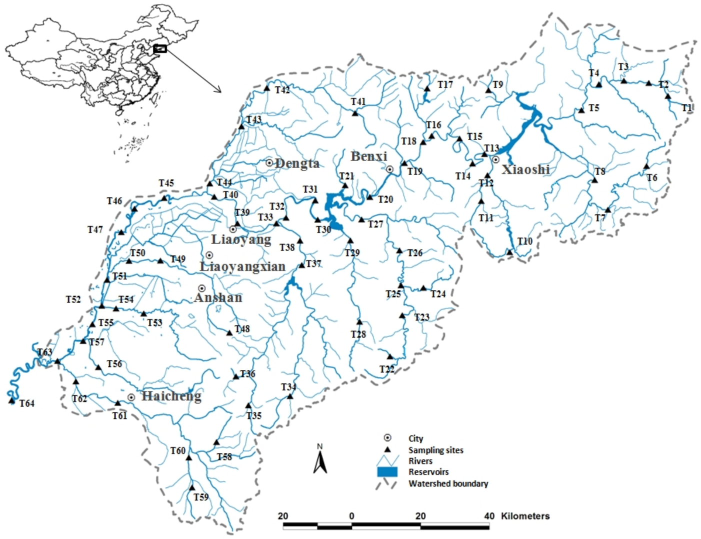

**Fig. 1. Distribution of the 64 sites sampled for benthic diatoms in the Taizi River basin**

## 2.2 Chemical Characteristics of Water Quality

Water samples were collected during the field trip before the processing of benthic diatom samples. Chemical parameters of electrical conductivity (EC) and dissolved oxygen (DO) were measured using a handheld YSI multiparameter instrument (professional plus) and pH was measured using a pen-style YSI pH tester instrument in situ. River water at each site was collected in polyethylene plastic bottles pre-rinsed three times with distilled water and kept below 4ºC for laboratory analysis. In the laboratory, ammonia-nitrogen ($NH_3$) was measured with Nessler's reagent. Nitrate-nitrogen ($NO_3$) and total nitrogen (TN) were determined through the alkaline potassium persulfate oxidation–UV spectrophotometric method. Concentrations of total phosphorus (TP) and orthophosphate-phosphorus ($PO_4$) were measured by the ammonium molybdate spectrophotometric method. Five-day biological oxygen demand ($BOD_5$) was calculated by quantifying the dissolved oxygen of the samples before and after the 5-day incubation at 20ºC and the permanganate index ($COD_{Mn}$) was measured by the potassium permanganate method. Chemical oxygen demand (COD) was measured by the standard potassium dichromate method. Suspended solids (SS) were measured by the standard operating procedure for total suspended solids analysis. The collection, transportation and analysis of water samples were performed according to the Chinese Standard Methods for Examination of Water and Wastewater [36] (Table 1).

## 2.3 Evaluation of Chinese Environmental Quality Standards for Surface Water

China manages its surface freshwater quality based on ambient water quality standards. Currently, national surface freshwater quality standards are used for the protection of water quality, decreasing the risk of water contamination, concerning human health of drinking water and maintenance of aquatic ecosystems. Five grades were used to evaluate from good to bad water quality conditions. The evaluation of Chinese environmental quality standards for surface water is based on the 'one-out, all-out' principle. In this way, the lowest water quality grade from any parameter represents the final water quality grade or class. According to the different water resource use aims, the water quality standards are divided into five classes in China [37]. These classes refer to headwater streams and national nature reserves (Class I); key zones for protection of surface drinking water, habitat for rare freshwater organisms and spawning regions for fish and shrimps (Class II); surface drinking water sources, wintering and migration regions for fish and shrimps (Class III); industrial and recreational water (Class IV) and agricultural and landscape water (Class V).

In this research, we selected six of the measured parameters for the water quality evaluation, excluding EC, $NO_3$ and TN. The EC does not have a national-level water quality standard since it is broadly affected by local geological conditions. The standard value of $NO_3$ is 10 mg/L, applied as a supplementary parameter for surface drinking water. The national water quality standard for TN is only applicable for lentic water in China; a TN standard for lotic waters has yet to be developed.

## 2.4 Benthic Algae Sampling and Identification

Benthic algae samples were collected from 64 sampling sites in the Taizi River before the monsoon season in May 2009 (Fig. 1). Nine samples were collected at each site from multiple substrates (e.g., stones, boulders, moss, vascular plants, sand or silt) and from multiple habitats (e.g., riffles, runs, shallow pools) within a 300 m sampling region [38]. All of the surface area of hard substrate within a 3.5-cm diameter corer was brushed thoroughly, and surface areas of soft substrate were collected using a core sampler with a 3.5 cm diameter, then all the samples were rinsed with a total of 500 ml distilled water and combined. The samples were preserved in a 4% formalin aqueous solution in a 500 ml plastic bottle. In the laboratory, diatom slides were kept in a glass jar to oxidize the organic material with acid disposal. Within each diatom slide, a minimum of 300 valves were counted under high magnification oil emersion. The 'soft' algae were identified directly using a 0.1-ml counting chamber. Most of the benthic algae were identified to the species level and several taxa were only identified to the genus level, according to the classification manuals of Hu and Wei [39] and Zhu and Chen [40].

## 2.5 Diatom Indices

Eight indices, including BDI, SHE, SPI, DES, GDI, CEE, TDI and ROTT were selected for evaluation in the river health assessment in China. The selected diatom indices mainly use weighted average values of sensitivity, indicator value and relative abundance of each taxon. The eight diatom indices (Table 2) were calculated using the OMNIDIA 7 software (Version 8.0) [24,41]. Zelinka and Marvan [8] served as a basis for the calculation of four indices, including DES, BDI, SPI and GDI, as follows:

$$ID = \frac{\sum A_i v_j i_j}{\sum A_i v_j},$$

where $A_j$ is the relative abundance of species j; $v_j$ is the indicator value ($1 \leq v \leq 3$) and $i_j$ is sensitivity.

The DES uses 106 species of diatoms that have been classified into five categories of sensitivity. The BDI is based on the ecological profiles of 209 taxa for which presence probabilities are defined from 14 physical and chemical parameters (e.g., organic pollution, nutrients, conductivity and pH) within seven water quality classes. The SPI is calculated from the Saprobic system and comprises around 13,000 taxa, which usually includes all observed taxa [23]. The GDI also uses all observed species and five categories of sensitivity. This index was developed in order to have an easily usable index for river health assessment that utilizes genera, instead of species for cases of limited taxonomic skills [23].

The Differentiating Species System (DSS) takes into account 100 freshwater diatom species with worldwide abundance [42]. Similar to the DSS, SHE has been modified to use 386 species from seven classifications based on trophic state and pollution resistance. The CEE is based on a double entry table and 208 species are used. Low indicator taxa are horizontally ranked by increasing tolerance and high indicator taxa are vertically ranked by increasing tolerance. Correspondingly, the taxa are classified in different groups (low indicator) and subgroups (high indicator). The intersection of the groups and subgroups results in the values that correspond to the quality class. TDI uses 1603 species of diatoms, five classes of sensitivity to trophic state and three classes of reliability. This index is suitable to detect eutrophication in rivers caused by large, predominantly lowland sewage works.

Values calculated for each index were linearly adjusted on a scale from 0–20, with 0 representing the worst water quality and 20 the best water quality. In this study, we classified the index scores into five categories corresponding to different water quality statuses of excellent (16–20), good (12– <16), fair (8– <12), poor (4– <8) and critical (0–4).

## 2.6 Data Analysis

Spearman rank correlation analysis was used to detect the correlations among different indices. Principal component analysis (PCA) was performed to detect the major water quality gradients. Before PCA, water quality parameters with relatively high partial correlation coefficients ($r > 0.75$) and variance inflation factors ($F > 20$) were eliminated. Through the PCA, the water quality parameters were reduced to a few interpretable principal components. Simple linear regression analyses were applied to evaluate the quantitative relationship between the disturbance gradients and diatom indices. The Kruskal–Wallis nonparametric test (K-W test) was used to evaluate the variation of chemical parameters among different diatom-based health indices and the general variation of each parameter within the groups was analyzed using ANOVA. The correlation analysis, PCA analysis and K–W test were performed with STATISTICA software (Version 7.0).

## 3. RESULTS

A total of 109 species and subspecies of diatoms within 24 genera and 8 families were identified in the Taizi River. The three most dominant species were *Achnanthes minutissima*, *Ceratoneis arcus* and *Diatoma vulgare var. lineare* Grun with relative abundances of 33%, 8%

and 7% respectively. For the 64 sampling sites, six species—*Nizschia palea*, *Synedra ulna*, *Cymbella ventricosa*, *Synedra acus*, *Melosira varians* and *Navicula rhynchocephala* — comprised more than 60% of the occurrences.

Nine water chemical parameters were measured to evaluate the usefulness of eight diatom-based indices for river health assessment in the Taizi River. According to the surface water quality standards, eutrophication was considerable, with relatively high levels of $NH_3$ (average 0.92 mg/L), TN (average 5.18 mg/L) and TP (average 0.31 mg/L). Meanwhile, the organic pollution was relatively lower compared with eutrophication. Most of the sampling sites had relatively high DO (average 5.70 mg/L) and low $COD_{Mn}$ (average 5.71 mg/L). However, the COD was high with an average 27.74 mg/L. The variation of EC was considerable, ranging from a minimum of 57μs/cm to a maximum of 1126 μs/cm (average 336 μs/cm) in the Taizi River (Table 1).

**Table 1. Values of water quality parameters and diatom-based indices for the Taizi River**

| | Code | Mean ± SD | Min–Max | Standards* |
|---|---|---|---|---|
| **Chemical parameters** | | | | |
| Suspended solids | SS | 36.160±27.855 | 5–124 | |
| Electric conductivity | EC | 336.070±244.259 μs/cm | 57–1126 | – |
| Dissolved Oxygen | DO | 5.700±2.044 mg/L | 0.4–10.3 | 7.5–2 |
| Chemical Oxygen Demand | COD | 27.740±19.449 mg/L | 0.0–100.5 | 15–40 |
| Permanganate index | $COD_{Mn}$ | 5.710±5.030 mg/L | 2.65–29.25 | 2–15 |
| 5-days Biological Oxygen Demand | $BOD_5$ | 5.440±5.207 mg/L | 0.0–28.5 | 3–10 |
| Ammonia-nitrogen | $NH_3$ | 0.920±1.401 mg/L | 0.00–6.21 | 0.15–2.00 |
| Nitrate-nitrogen | $NO_3$ | 2.880±2.620 mg/L | 0.00–12.25 | < 10** |
| Total nitrogen | TN | 5.180±3.617 mg/L | 0.68–17.75 | 0.2–2.0§ |
| Total phosphorus | TP | 0.310±0.560 mg/L | 0.00–3.03 | 0.02–0.40 |
| **Diatom indices** | | | | |
| Biological Diatom Index | BDI | 12.850±3.731 | 5.0–20.0 | |
| Schiefele and Schreiner's Index | SHE | 11.810±4.720 | 1.0–19.9 | |
| Specific Pollution sensitivity Index | SPI | 12.010±4.735 | 1.0–19.7 | |
| Descy Index | DES | 9.480±4.486 | 1.0–16.0 | |
| Generic Index of Diatom assemblage | GDI | 12.680±4.176 | 1.0–19.4 | |
| Commission for Economical Community Index | CEE | 7.930±.027 | 1.0–17.7 | |
| Trophic Diatom Index | TDI | 7.650±3.436 | 1.0–19.6 | |
| RottSaprobic Index | ROTT | 11.340±3.691 | 3.7–16.5 | |

**Chinese environmental quality standards for surface water (NEPB, 2002b)*

***Supplementary parameter of surface water for drinking water*

*§TN standards are currently available only for lentic but not lotic, waters in China*

Comparisons among the different diatom-based indices showed that GDI had a relatively high level of evaluation with an average value of 12.68, while TDI had relatively low level of evaluation with an average value of 7.65. Among the eight indices, BDI showed generally higher evaluation with all values higher than 4. In contrast, DES and ROTT showed generally lower evaluation with all values lower than 16 (Table 1). Significant correlations were found among the eight diatom indices using Spearman correlation analysis and all the P-values were less than 0.01 (Table 2). The correlation coefficient was highest for SPI and GDI (r = 0.951) and lowest for SHE and CEE (r = 0.666).

**Table 2. Correlation coefficient matrix for the eight diatom indices (n = 64, P< 0.01)**

| | BDI | SHE | SPI | DES | GDI | CEE | TDI | ROTT |
|---|---|---|---|---|---|---|---|---|
| BDI | 1 | | | | | | | |
| SHE | 0.829 | 1 | | | | | | |
| SPI | 0.903 | 0.884 | 1 | | | | | |
| DES | 0.917 | 0.864 | 0.878 | 1 | | | | |
| GDI | 0.857 | 0.872 | 0.951 | 0.857 | 1 | | | |
| CEE | 0.691 | 0.666 | 0.691 | 0.671 | 0.677 | 1 | | |
| TDI | 0.708 | 0.744 | 0.756 | 0.795 | 0.822 | 0.686 | 1 | |
| ROTT | 0.883 | 0.883 | 0.885 | 0.932 | 0.861 | 0.667 | 0.822 | 1 |

The first three factorial axes were constructed to represent the dominant water quality gradients on the basis of the decline in eigen values in the PCA (Fig. 2). Three axes accounted for 68.68% of the total variance (Fig. 2), among which the first axis accounted for 40.19% and the second for 18.72%. The main explanatory parameters for PCA Factor 1 were $NH_3$, DO, EC and TP. The $NH_3$, EC, TP, COD, $COD_{Mn}$ and $BOD_5$ were negatively correlated with PCA Factor 1, while DO was positively correlated. Main explanatory parameters for PCA Factor 2 were $NO_3$ and TN, both of which were negatively correlated. SS explained the largest amount of variance on PCA Factor 3 and was positively correlated.

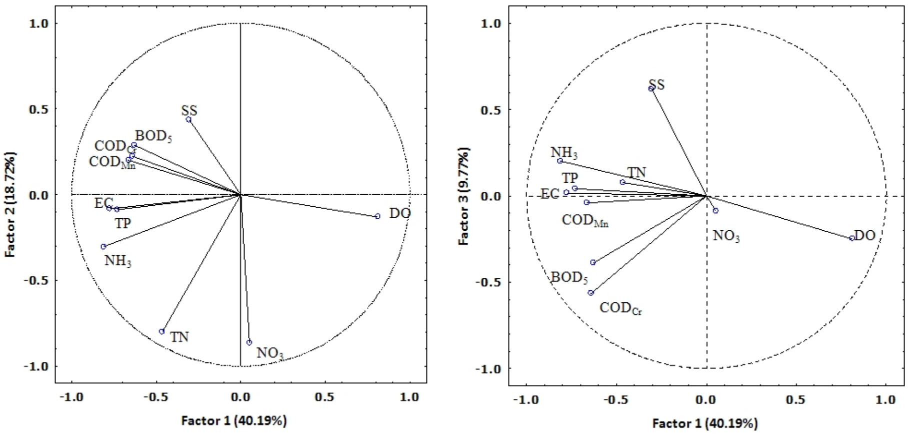

**Fig. 2. Principal component analysis (PCA) ordination plots for 10 water quality parameters at the 64 sites sampled in the Taizi River basin. (a) Factor 1 × Factor 2 and (b) Factor 1 × Factor 3**

The regression analyses revealed that all eight of the diatom indices could be explained significantly by PCA Factor 1. The negative direction of the PCA Factor 1 represented the main water quality stressor gradient. All indices showed positive linear relationships with PCA Factor 1. Among the eight indices, BDI, SPI and ROTT showed relatively higher levels of explanation (adjusted $R^2$ = 0.46–0.48). GDI also showed a significant linear model with PCA Factor 2 but at low levels of variance ($R^2$ = 0.06, P=0.04). GDI was positively correlated with PCA Factor 2. There were no indices that had significant correlations with PCA Factor 3 (Table 3).

**Table 3. Adjusted coefficients of determination ($R^2$) from linear regressions used to analyze the response of eight diatom indices to the multiple stressor gradients (PCA Factors 1 and 2)**

| Equation | Adjusted $R^2$ | F-value | P-value |
|---|---|---|---|
| BDI = −1.28*PCA Factor 1+12.85 | 0.47 | 56.50 | < 0.001 |
| SHE = 1.40*PCA Factor 1+11.81 | 0.34 | 33.69 | < 0.001 |
| SPI = 1.64*PCA Factor 1+12.01 | 0.48 | 58.17 | < 0.001 |
| DES = 1.33*PCA Factor 1+9.48 | 0.35 | 33.90 | < 0.001 |
| GDI = 1.29*PCA Factor 1+12.68 | 0.38 | 38.82 | < 0.001 |
| CEE = 1.22*PCA Factor 1+7.93 | 0.36 | 36.13 | < 0.001 |
| TDI = 0.98*PCA Factor 1+7.65 | 0.32 | 30.03 | < 0.001 |
| ROTT = 1.26*PCA Factor 1+11.34 | 0.46 | 54.99 | < 0.001 |
| GDI = 0.83*PCA Factor 2+12.68 | 0.06 | 4.93 | 0.04 |

In general, the variation of diatom-based indices did not correspond to water quality classes from relative clean water (Class II) to contaminated water (Class V) in the Taizi River. Among the eight indices, SHE, DES and TDI had no significant variation overall and among the different groups in relation to the decreasing water quality classes (Fig. 3). The other five indices of BDI, SPI, GDI, CEE and ROTT showed significant differences of general variation through the one-way ANOVA test (P< 0.05). However, the differences were not significant among Classes II– IV. These indices only showed significant differences between the lowest water quality grade (Class V) and the other classes using the multiple comparison K-W test (Fig. 3).

The sensitivities reflected by the correlations of chemical parameters to the river health categories of diatom-based indices varied significantly among different indices. Generally, chemical parameters all showed significant increasing water quality trends with increasing health categories of diatom-based indices except for $COD_{Mn}$ and $NO_3$. Among the nine chemical parameters, EC, $NH_3$ and TP showed broad, significant correlations with all of the diatom-based indices (P< 0.001). DO also showed broad correlations to indices except for TDI. COD and $BOD_5$ were significantly correlated with five indices and TN with two indices. The multiple comparisons K-W test showed considerable variations for BDI, DES and GDI with six chemical parameters; SHE, TDI, CEE and ROTT showed variations with seven chemical parameters and SPI showed significant variations with eight chemical parameters (Table 4).

As the health categories improved, the average values of chemical parameters gradually changed in the positive direction or for DO, the negative direction. The variation was significant between the highest (i.e., very good) and lowest (i.e., critical) health class when the multiple comparison of K-W test was used. There were 31 chemical parameters in total that showed these types of significant variations, identified as Type I and Type II. These indices were suitable bio-indicators to reflect the changing concentrations of chemical parameters. Except for the gradual, significant increasing or decreasing trends, there were also 14 chemical parameters that showed abrupt variation from the best to the worst health category. These indices may not be sufficient or feasible to fulfill the assessment objectives of chemical parameter variations.

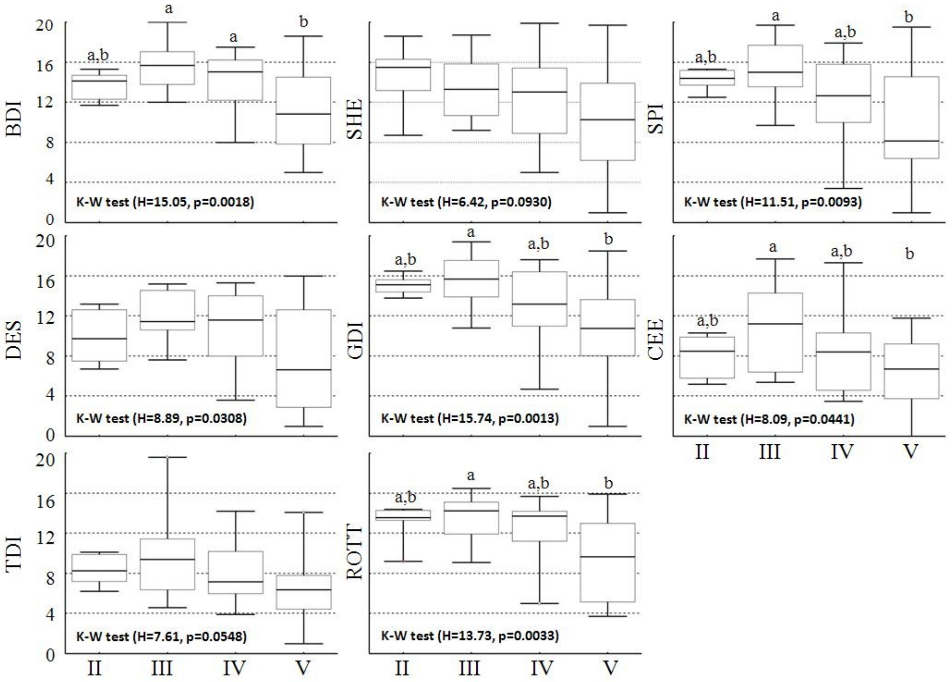

**Fig. 3. Box plots for the eight diatom indices in relation to the Chinese surface water quality standards established for Classes II–V. The general variation and multiple comparisons among different water quality grades were conducted with the Kruskal–Wallis test (K-W test), with associated H- and P-values shown for each index. Different letters above the boxes indicate significant differences (P< 0.05) among the groups based on the multiple comparison K-W test. The dashed lines represented the health classes explained at the diatom-based indices in details**

Eight sub-types were identified for those classifications that showed similar, significant variations between river health categories (Fig. 4). Sub-type I (Type I– I) represented when the variation was significant between the first category (very good for SHE; good for BDI, DES and ROTT) and the critical category. However, the variations were not significant among the middle three categories (or two categories for BDI, DES and ROTT) with the very good or critical categories (Fig. 4A and 4B). In total, 11 indices showed the same sub-types, including DO, COD, $COD_{Mn}$ and $BOD_5$ for BDI; DO, $COD_{Mn}$ and $BOD_5$ for SHE; TN for SPI and DES and EC and $NH_3$ for ROTT (Table 4). Sub-type II (Type I – II) showed similar variations as Type I – I, however the variation was significant between the very good category and poor and critical categories, such as TN and TP for SHE (Fig. 4C). For the four health classes of BDI and DES, the variations of EC, $NH_3$ and TP showed significance between the good category and poor and critical categories (Fig. 4D and Table 4). Sub-type III (Type I – III) was also similar to Type I – I but the variation was significant between the good and critical categories of DO for SPI (Fig. 4E). For the four health categories of DES and ROTT, the variations of EC and TP were significant between very good and good categories with the poor category (Fig. 4F and Table 4). For sub-type IV (Type I – IV) the variation was significant for very good and good from poor and critical, such as the EC for SPI (Fig. 4G). For the four health categories of BDI, the variation of $NH_3$ was significant for the very good to the poor and critical (Fig. 4H and Table 4). Sub-type V (Type I – V) showed that variation was significant between the very good category and the rest of the categories, only represented as TP for TDI (Fig. 4J). Sub-type VI (Type I – VI) had significant variation between the very good category and fair, poor and critical categories, only represented as COD for SHE (Fig. 4K). Sub-type VII (Type I – VII) showed significant variation for very good, good and fair categories with the critical category, represented as EC for GDI (Fig. 4L). Finally for sub-type VIII (Type I – VIII), the variation was significant for very good, good and fair categories with the poor category, represented as the $BOD_5$ for DES (Fig. 4M).

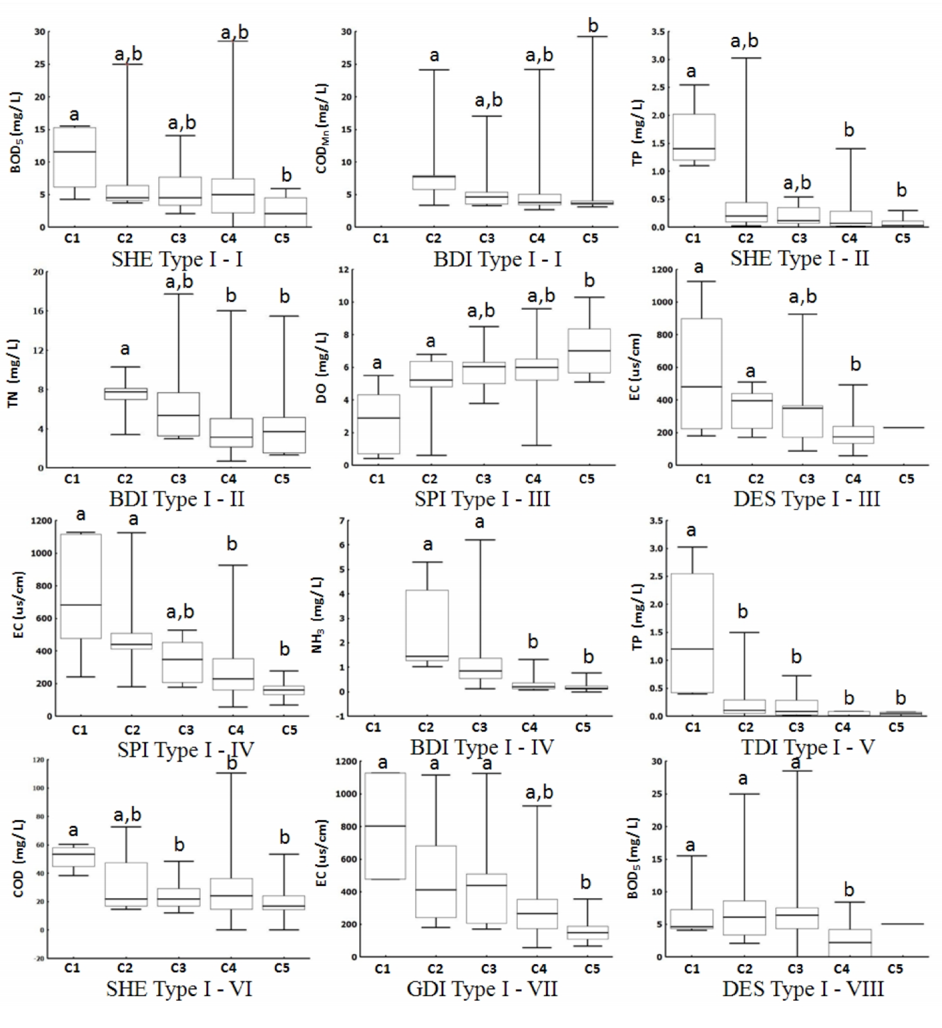

**Fig. 4. Box plots for select indices of different sub-types within Type I. Different letters above the boxes indicate significant differences (P< 0.05) based on the multiple comparison K-W test**

**Table 4. Differences of nine chemical parameters, excluding suspended solids, among the five river health categories for each diatom-based index. Different letters indicate significant differences among different categories based on the multiple comparisons K-W test. The bold text indicates significant differences of the five categories with the K-W test, with associated F- and P-values**

| | | **C1 (very good)** | **C2 (good)** | **C3 (fair)** | **C4 (poor)** | **C5 (critical)** | **F-value** | **P-value** |
|---|---|---|---|---|---|---|---|---|
| **Sampling sites** | | **0** | **9** | **15** | **24** | **16** | | |
| BDI | EC | | 690.90 ± 125.38$^{a}$ | 371.60 ± 51.43$^{b}$ | 255.80 ± 24.33$^{b}$ | 223.60 ± 31.33$^{b}$ | 13.42 | < 0.001 |
| | DO | | 3.20 ± 0.87$^{a}$ | 5.50 ± 0.40$^{b}$ | 5.90 ± 0.27$^{b}$ | 7.00 ± 0.46$^{b}$ | 9.64 | < 0.001 |
| | COD | | 44.30 ± 7.18$^{a}$ | 34.30 ± 7.00$^{ab}$ | 23.00 ± 2.48$^{b}$ | 19.50 ± 2.68$^{b}$ | 4.96 | 0.004 |
| | $COD_{Mn}$ | | 8.40 ± 2.06 | 5.60 ± 0.93 | 5.00 ± 0.87 | 5.40 ± 1.60 | 1.02 | 0.3910 |
| | $BOD_5$ | | 10.10 ± 2.74$^{a}$ | 6.40 ± 1.43$^{ab}$ | 4.70 ± 0.73$^{b}$ | 3.00 ± 0.72$^{b}$ | 4.60 | 0.006 |
| | $NH_3$ | | 2.70 ± 0.57$^{a}$ | 1.50 ± 0.48$^{b}$ | 0.40 ± 0.08$^{c}$ | 0.20 ± 0.05$^{c}$ | 12.84 | < 0.001 |
| | $NO_3$ | | 2.90 ± 0.72 | 2.80 ± 0.72 | 3.10 ± 0.60 | 2.60 ± 0.59 | 0.13 | 0.941 |
| | TN | | 7.40 ± 0.62 | 6.10 ± 0.96 | 4.30 ± 0.75 | 4.20 ± 0.92 | 2.35 | 0.081 |
| | TP | | 1.00 ± 0.25$^{a}$ | 0.50 ± 0.20$^{ab}$ | 0.10 ± 0.03$^{b}$ | 0.10 ± 0.02$^{b}$ | 8.92 | < 0.001 |
| **Sampling sites** | | **4** | **10** | **15** | **22** | **13** | | |
| SHE | EC | 960.50 ± 161.18$^{a}$ | 404.60 ± 51.31$^{b}$ | 297.70 ± 35.96$^{b}$ | 297.00 ± 43.72$^{b}$ | 201.50 ± 31.81$^{b}$ | 15.04 | < 0.001 |
| | DO | 7.40 ± 0.53$^{a}$ | 4.90 ± 0.45$^{b}$ | 4.30 ± 0.37$^{b}$ | 6.80 ± 1.37$^{b}$ | 5.60 ± 1.97$^{b}$ | 5.99 | < 0.001 |
| | COD | 51.40 ± 4.71$^{a}$ | 31.70 ± 6.30$^{ab}$ | 24.00 ± 2.52$^{b}$ | 29.20 ± 5.36$^{b}$ | 19.40 ± 3.43$^{b}$ | 2.60 | 0.05 |
| | $COD_{Mn}$ | 7.40 ± 0.53 | 4.90 ± 0.45 | 4.30 ± 3.67 | 6.80 ± 1.37 | 5.60 ± 1.97 | 0.73 | 0.57 |
| | $BOD_5$ | 10.70 ± 2.73$^{a}$ | 7.10 ± 2.05$^{ab}$ | 5.70 ± 0.80$^{ab}$ | 5.40 ± 1.29$^{ab}$ | 2.30 ± 0.67$^{b}$ | 2.73 | 0.04 |
| | $NH_3$ | 3.80 ± 0.85$^{a}$ | 1.80 ± 0.58$^{b}$ | 0.60 ± 0.13$^{ab}$ | 0.60 ± 0.25$^{ab}$ | 0.20 ± 0.08$^{c}$ | 10.30 | < 0.001 |
| | $NO_3$ | 2.80 ± 1.29 | 2.90 ± 1.01 | 3.80 ± 0.71 | 2.90 ± 0.63 | 1.90 ± 0.23 | 0.84 | 0.504 |
| | TN | 8.60 ± 0.57$^{a}$ | 5.80 ± 1.45$^{ab}$ | 5.60 ± 0.90$^{ab}$ | 4.80 ± 0.82$^{ab}$ | 3.80 ± 0.66$^{b}$ | 1.59 | 0.188 |
| | TP | 1.60 ± 0.32$^{a}$ | 0.50 ± 0.29$^{b}$ | 0.20 ± 0.04$^{b}$ | 0.20 ± 0.07$^{b}$ | 0.10 ± 0.02$^{b}$ | 10.92 | < 0.001 |
| **Sampling sites** | | **5** | **12** | **8** | **27** | **12** | | |
| SPI | EC | 728.00 ± 174.72$^{a}$ | 491.70 ± 72.04$^{b}$ | 339.90 ± 48.09$^{bc}$ | 269.80 ± 33.04$^{bc}$ | 163.80 ± 15.99$^{c}$ | 10.17 | < 0.001 |
| | DO | 2.80 ± 0.99$^{a}$ | 4.80 ± 0.59$^{b}$ | 5.90 ± 0.50$^{bc}$ | 5.90 ± 0.31$^{bc}$ | 7.20 ± 0.50$^{c}$ | 6.69 | < 0.001 |
| | COD | 34.90 ± 8.75$^{ab}$ | 42.60 ± 6.54$^{a}$ | 22.10 ± 2.70$^{b}$ | 25.70 ± 4.09$^{ab}$ | 18.30 ± 1.09$^{b}$ | 3.30 | 0.02 |
| | $COD_{Mn}$ | 6.60 ± 0.86 | 6.40 ± 1.67 | 4.70 ± 1.63 | 6.40 ± 6.55 | 3.80 ± 0.12 | 0.73 | 0.57 |
| | $BOD_5$ | 7.90 ± 1.89$^{ab}$ | 10.10 ± 2.55$^{a}$ | 3.90 ± 0.97$^{bc}$ | 4.70 ± 0.58$^{bc}$ | 2.50 ± 0.71$^{c}$ | 4.89 | 0.001 |
| | $NH_3$ | 4.00 ± 0.78$^{a}$ | 1.40 ± 0.37$^{b}$ | 0.90 ± 0.16$^{bc}$ | 0.50 ± 0.21$^{bc}$ | 0.20 ± 0.03$^{c}$ | 14.64 | < 0.001 |
| | $NO_3$ | 4.60 ± 1.92$^{a}$ | 2.10 ± 0.45$^{a}$ | 4.30 ± 1.29$^{a}$ | 3.00 ± 0.51$^{a}$ | 1.70 ± 0.19$^{a}$ | 2.17 | 0.09 |
| | TN | 10.10 ± 2.02$^{a}$ | 5.10 ± 0.64$^{ab}$ | 6.60 ± 1.59$^{b}$ | 4.50 ± 0.66$^{b}$ | 3.80 ± 0.72$^{b}$ | 3.85 | 0.008 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | TP | 1.10 ± 0.41$^{a}$ | 0.60 ± 0.25$^{ab}$ | 0.20 ± 0.07$^{b}$ | 0.20 ± 0.06$^{b}$ | 0.10 ± 0.01$^{b}$ | 5.53 | < 0.001 |
| Sampling sites | | 12 | 13 | 16 | 22 | 1 | | |
| DES | EC | 565.30 ± 106.65$^{a}$ | 349.20 ± 31.93$^{b}$ | 353.40 ± 56.53$^{b}$ | 195.40 ± 23.14$^{b}$ | 231.0 | 7.89 | < 0.001 |
| | DO | 4.20 ± 0.68$^{a}$ | 5.60 ± 0.28$^{ab}$ | 5.60 ± 0.58$^{ab}$ | 6.60 ± 0.33$^{b}$ | 9.5 | 4.46 | 0.007 |
| | COD | 36.20 ± 4.52 | 29.70 ± 4.98 | 30.10 ± 7.02 | 20.80 ± 2.67 | 16.9 | 1.86 | 0.15 |
| | COD$_{Mn}$ | 5.70 ± 0.56 | 4.50 ± 0.53 | 6.50 ± 1.45 | 5.90 ± 1.45 | 5.5 | 0.38 | 0.76 |
| | BOD$_{5}$ | 6.80 ± 1.20$^{a}$ | 7.50 ± 1.74$^{ab}$ | 7.00 ± 1.58$^{b}$ | 2.40 ± 0.54$^{b}$ | 5.1 | 4.41 | 0.007 |
| | NH$_{3}$ | 2.80 ± 0.55$^{a}$ | 0.70 ± 0.11$^{b}$ | 0.70 ± 0.35$^{b}$ | 0.30 ± 0.06$^{b}$ | 0.2 | 14.06 | < 0.001 |
| | NO$_{3}$ | 3.30 ± 0.93 | 2.90 ± 0.41 | 2.80 ± 0.70 | 2.80 ± 0.61 | 1.6 | 0.1 | 0.96 |
| | TN | 7.90 ± 1.06$^{a}$ | 4.70 ± 0.50$^{ab}$ | 4.90 ± 0.92$^{b}$ | 4.40 ± 0.83$^{b}$ | 1.8 | 2.99 | 0.038 |
| | TP | 1.00 ± 0.28$^{a}$ | 0.20 ± 0.06$^{b}$ | 0.20 ± 0.09$^{b}$ | 0.10 ± 0.02$^{b}$ | 0.1 | 10.72 | < 0.001 |
| Sampling sites | | 2 | 6 | 15 | 26 | 15.0 | | |
| GDI | EC | 801.50 ± 324.50$^{a}$ | 507.30 ± 140.86$^{ab}$ | 428.80 ± 60.15$^{bc}$ | 307.70 ± 37.17$^{bc}$ | 162.00 ± 19.68$^{c}$ | 7.04 | < 0.001 |
| | DO | 3.10 ± 2.40$^{a}$ | 3.90 ± 0.79$^{b}$ | 5.60 ± 0.46$^{ab}$ | 5.60 ± 0.35ab | 7.10 ± 0.47$^{b}$ | 4.73 | 0.002 |
| | COD | 44.80 ± 6.30 | 32.80 ± 7.16 | 31.80 ± 4.75 | 27.70 ± 4.79 | 19.50 ± 1.60 | 1.35 | 0.26 |
| | COD$_{Mn}$ | 6.80 ± 1.00$^{a}$ | 5.00 ± 0.95$^{a}$ | 5.00 ± 0.40$^{a}$ | 7.30 ± 1.46$^{a}$ | 3.80 ± 0.20$^{a}$ | 1.32 | 0.27 |
| | BOD$_{5}$ | 6.20 ± 1.93$^{a}$ | 8.20 ± 2.02$^{b}$ | 7.20 ± 1.55$^{ab}$ | 5.00 ± 1.11$^{b}$ | 3.20 ± 0.70$^{b}$ | 1.64 | 0.18 |
| | NH$_{3}$ | 2.90 ± 1.50 | 2.80 ± 0.93 | 1.20 ± 0.31 | 0.60 ± 0.22 | 0.20 ± 0.02 | 7.87 | < 0.001 |
| | NO$_{3}$ | 4.10 ± 2.95 | 3.70 ± 1.60 | 2.90 ± 0.76 | 3.20 ± 0.53 | 1.80 ± 0.21 | 0.95 | 0.44 |
| | TN | 9.20 ± 1.10$^{a}$ | 7.50 ± 2.27$^{ab}$ | 5.70 ± 0.93$^{ab}$ | 5.00 ± 0.67$^{ab}$ | 3.50 ± 0.62$^{b}$ | 2.34 | 0.07 |
| | TP | 1.80 ± 0.73$^{a}$ | 0.50 ± 0.19$^{b}$ | 0.50 ± 0.21$^{b}$ | 0.20 ± 0.06$^{b}$ | 0.10 ± 0.03$^{b}$ | 6.97 | 0.001 |
| **Sampling sites** | | **6** | **32** | **21** | **3** | **2** | | |
| TDI | EC | 595.30 ± 174.74$^{a}$ | 384.90 ± 37.79$^{b}$ | 244.10 ± 33.895$^{b}$ | 101.50 ± 25.02$^{b}$ | 95.00 ± 26.00$^{b}$ | 4.91 | 0.002 |
| | DO | 4.20 ± 1.15$^{a}$ | 5.40 ± 0.31$^{b}$ | 6.40 ± 0.41$^{b}$ | 7.10 ± 1.61$^{b}$ | 6.90 ± 1.80$^{b}$ | 2.35 | 0.06 |
| | COD | 44.20 ± 3.48$^{a}$ | 29.50 ± 3.80$^{ab}$ | 22.70 ± 3.88$^{ab}$ | 16.90 ± 3.74$^{b}$ | 20. 50 ± 1.03$^{b}$ | 1.90 | 0.122 |
| | COD$_{Mn}$ | 5.60 ± 0.83 | 5.80 ± 0.89 | 6.20 ± 1.35 | 3.30 ± 0.09 | 3.90 ± 0.18 | 0.27 | 0.90 |
| | BOD$_{5}$ | 6.70 ± 1.78$^{a}$ | 6.30 ± 0.85$^{a}$ | 4.20 ± 1.37$^{a}$ | 4.40 ± 2.26$^{a}$ | 3.20 ± 1.00$^{a}$ | 0.70 | 0.60 |
| | NH$_{3}$ | 2.40 ± 0.60$^{a}$ | 1.20 ± 0.29$^{b}$ | 0.30 ± 0.07$^{b}$ | 0.10 ± 0.02$^{b}$ | 0.20 ± 0.05$^{b}$ | 4.11 | 0.005 |
| | NO$_{3}$ | 2.80 ± 1.01 | 3.20 ± 0.52 | 2.70 ± 0.54 | 1.40 ± 0.13 | 1.50 ± 0.27 | 0.55 | 0.70 |
| | TN | 7.90 ± 0.60$^{ab}$ | 5.40 ± 0.73$^{bc}$ | 4.90 ± 0.66$^{bc}$ | 1.60 ± 0.14$^{a}$ | 2.80 ± 1.27$^{c}$ | 2.02 | 0.10 |
| | TP | 1.50 ± 0.45$^{a}$ | 0.20 ± 0.06$^{b}$ | 0.20 ± 0.04$^{b}$ | 0.00 ± 0.02$^{b}$ | 0.00 ± 0.03$^{b}$ | 12.45 | < 0.001 |
| **Sampling sites** | | **9** | **26** | **21** | **3** | **5** | | |
| CEE | EC | 704.90 ± 127.69$^{a}$ | 346.50 ± 29.61$^{b}$ | 237.10 ± 24.25$^{b}$ | 229.30 ± 44.88$^{b}$ | 97.70 ± 15.12$^{b}$ | 12.87 | < 0.001 |
| | DO | 3.20 ± 0.85$^{a}$ | 5.60 ± 0.28$^{b}$ | 6.70 ± 0.40$^{b}$ | 7.40 ± 0.54$^{b}$ | 6.00 ± 0.69$^{b}$ | 7.07 | < 0.001 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | COD | 45.50 ± 10.11$^{a}$ | 28.40 ± 3.54$^{ab}$ | 23.10 ± 3.09$^{ab}$ | 16.10 ± 4.86$^{b}$ | 18.80 ± 2.06$^{b}$ | 3.08 | 0.02 |
| | COD$_{Mn}$ | 7.50 ± 1.32 | 5.10 ± 0.80 | 6.50 ± 1.52 | 3.50 ± 0.17 | 3.60 ± 0.18 | 0.86 | 0.49 |
| | BOD$_5$ | 7.80 ± 1.49$^{a}$ | 6.60 ± 1.33$^{a}$ | 4.00 ± 0.61$^{a}$ | 1.00 ± 0.97$^{a}$ | 3.90 ± 1.31$^{a}$ | 1.94 | 0.11 |
| | NH$_3$ | 3.60 ± 0.71$^{a}$ | 0.70 ± 0.09$^{b}$ | 0.30 ± 0.08$^{b}$ | 0.30 ± 0.19$^{b}$ | 0.10 ± 0.02$^{b}$ | 26.47 | < 0.001 |
| | NO$_3$ | 3.00 ± 1.21 | 3.50 ± 0.57 | 2.20 ± 0.23 | 4.40 ± 3.01 | 1.50 ± 0.10 | 1.37 | 0.25 |
| | TN | 8.20 ± 1.42$^{ab}$ | 5.20 ± 0.68$^{abc}$ | 4.10 ± 0.50$^{bc}$ | 8.70 ± 3.53$^{a}$ | 2.10 ± 0.49$^{c}$ | 4.48 | 0.003 |
| | TP | 1.30 ± 0.34$^{a}$ | 0.20 ± 0.03$^{b}$ | 0.20 ± 0.05$^{b}$ | 0.10 ± 0.02$^{b}$ | 0.00 ± 0.02$^{b}$ | 15.39 | < 0.001 |
| **Sampling sites** | | **3** | **11** | **15** | **34** | **1** | | |
| ROTT | EC | 696.30 ± 214.85$^{a}$ | 505.50 ± 103.31$^{ab}$ | 436.50 ± 50.11$^{bc}$ | 211.50 ± 17.67$^{c}$ | 121.0 | 11.68 | < 0.001 |
| | DO | 4.20 ± 1.77$^{a}$ | 4.40 ± 0.66$^{ab}$ | 5.00 ± 0.54$^{ab}$ | 6.60 ± 0.26$^{b}$ | 5.1 | 5.85 | 0.001 |
| | COD | 45.70 ± 3.75$^{a}$ | 37.10 ± 6.11$^{ab}$ | 33.60 ± 7.23$^{ab}$ | 20.80 ± 2.01$^{b}$ | 19.5 | 4.04 | 0.01 |
| | COD$_{Mn}$ | 6.30 ± 0.74 | 5.30 ± 0.58 | 6.50 ± 1.56 | 5.50 ± 0.96 | 4.1 | 0.18 | 0.91 |
| | BOD$_5$ | 5.50 ± 1.29$^{a}$ | 8.90 ± 2.05$^{a}$ | 7.30 ± 1.74$^{a}$ | 3.50 ± 0.51$^{a}$ | 4.2 | 4.43 | 0.007 |
| | NH$_3$ | 2.40 ± 0.96$^{a}$ | 2.50 ± 0.59$^{a}$ | 1.00 ± 0.36$^{b}$ | 0.30 ± 0.04$^{b}$ | 0.2 | 13.56 | <0.001 |
| | NO$_3$ | 2.70 ± 2.03 | 3.40 ± 0.91 | 3.00 ± 0.72 | 2.70 ± 0.43 | 1.8 | 0.16 | 0.92 |
| | TN | 8.00 ± 1.34$^{a}$ | 7.10 ± 1.22$^{a}$ | 5.20 ± 0.92$^{a}$ | 4.30 ± 0.58$^{a}$ | 4.1 | 2.47 | 0.07 |
| | TP | 2.20 ± 0.58$^{a}$ | 0.50 ± 0.14$^{b}$ | 0.20 ± 0.09$^{b}$ | 0.10 ± 0.03$^{b}$ | 0.1 | 36.46 | <0.001 |

The second main type of distribution was identified as that of increasing health categories with the average values of chemical parameters also gradually changing in a negative direction. With the multiple comparison K-W test, the variations were significant between the very good category and the poor and critical categories and between the very good and good categories with the critical category. This kind of variation was identified as Type II. Five chemical parameters showed the same variation trends, for example, NH3 for SHE (Fig. 5A) and EC for CEE (Fig. 5B).

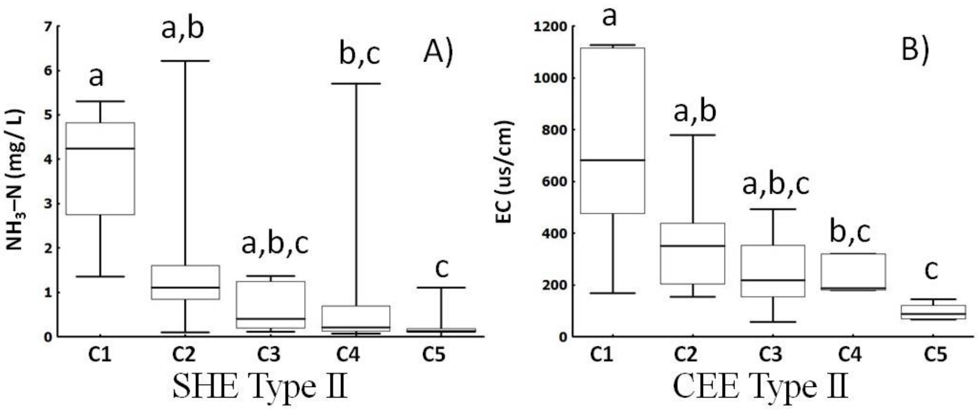

**Fig. 5. Box plots for select indices within Type II. Different letters above the boxes indicate significant differences (P< 0.05) based on the multiple comparison K-W test.**

## 4. DISCUSSION

Diatom-based indices have successfully been used as bioassessment indicators of freshwater ecosystems to reflect different kinds of human activities [11,43]. Our study confirmed that eight diatom-based indices could be used to effectively evaluate the main water quality degradation gradients (Table 3). Since these diatom-based indices were mainly developed based on the taxa found in numerous European countries, some studies have questioned the suitability and applicability of diatom-based indices in other countries, such as in China [44]. However, these indices have been successfully applied for river health assessment in several non-European countries, including Canada [45], Argentina [46], Brazil [47] and South Africa [48]. Other studies conducted in Chinese rivers, including subtropical rivers located at southern [22,29] and central China [18], in addition to our research results of temperate Taizi River in northeastern China, recommended the broad applicability of diatom-based indices in China. As the best what we know, these diatom-based indices were used for the river health assessment at the first time at temperate rivers in the Northeastern China.

This study revealed the potentially high usefulness for integrating the eight indices into the river health assessment of Taizi River and other streams in northeastern China. The linear regression analyses revealed the high explanation of the main stressor gradient (PCA Factor 1) to eight indices with relatively high $R^2$ (0.32–0.48, P<0.001). In our study, concentrations of the most tested chemical parameters were relatively high. For example, high concentrations of COD (maximum 110.5 mg/L), $COD_{Mn}$ (maximum 29.25 mg/L) and $BOD_5$ (maximum 28.5 mg/L), as well as of TN (0.68–17.75 mg/L) and TP (0–3.03 mg/L) reflected relatively high levels of organic pollution and nutrient enrichment in the Taizi River (Table 1). Both diatom-based indices and communities have proven useful to assess organic pollution and eutrophication over the long term, either synthetically or separately [49-51]. The negative direction of those chemical parameters in the first axis of the PCA ordination biplot (Fig. 2) showed that synthetic contamination was common in the research area. Thus, the significant regression results supported the usefulness of the diatom-based indices for river health assessment. Diatom-based indices are usually developed to assess specific contaminants for example TDI was developed specifically to evaluate eutrophication. However, the results of our study showed that these indices could reflect general contamination and evaluate river health condition [25,28]. Meanwhile, the relatively high, positive inter-correlation coefficients (0.666–0.951) revealed similar variation trends to human stressors among the eight indices (Table 2).

The general patterns (Type I and Type II) were mainly identified from the box plots and multiple comparisons (Fig. 4, Fig. 5 and Table 4). The major types and sub-types identified must show consistent increasing or decreasing trends with increasing diatom-based index classes [52]. The variation trends and significance of the eight indices were not consistent across the different chemical parameters. In light of the general patterns identified (both Type I and Type II), BDI and SHE were recommended as the most suitable indices for river health assessment in the Taizi River. SPI, DES and ROTT were also identified as suitable indices for the Taizi River. Among those indices, the usefulness of BDI has been reported for other freshwater ecosystems in China [18,22,30,31]. As a result, BDI may be recommended as a national bio-indicator for river health assessment in China. SHE, SPI, DES and ROTT may be recommended as local bio-indicators for river health assessment in northeastern China.

Because the variation trends were not consistent between the water quality classes and diatom-based indices, our results recommended that integrated river health assessment use both biological and chemical parameters. Water quality standards serve as a major basis for the management and protection of freshwater ecosystems. The Chinese government manages its water quality mainly based on ambient water quality standards. Currently, water quality grades are determined based on the chemical parameter with the worst contamination level. However, since these standards are mainly derived from environmental quality standards or criteria from developed countries that consider both ecosystem and human health, the current water quality assessment may not match well with the diatom-based indices [32]. The results also supported the notion that integrated river health assessment should include both biological and chemical information for better river management [53].

From the results it was apparent that several indices were widely applicable for river health assessment in the Taizi River. However, the community composition of benthic diatoms shows high variation and succession in different seasons, which directly affect the final index scores and river health assessment [18]. Meanwhile, variations in other natural conditions (e.g., altitude, forest cover) or human activities (e.g., watershed land uses, heavy metal contamination) may also affect community compositions of benthic diatoms and subsequently the final scores of diatom-based indices in the Taizi River. Thus, continual evaluation of diatom-based indices is necessary for river health assessment on either the national or local scale.

## 5. CONCLUSION

Even though diatom-based indices calculated from the OMNIDIA program were developed based on taxa from European rivers, our results confirmed that the eight selected indices were suitable for river health assessment in temperate regions of China. In this study, the eight tested indices all showed significantly high correlations with the main water contamination gradient (PCA Factor 1) according to linear regression models. BDI and SHE were recommended as the most suitable indicators, and SPI, DES, and ROTT were also suitable for river health assessment in the Taizi River. We also recommended BDI as a suitable indicator for the national river health assessment in China. Considering that the health categories of diatom-based indices were not consistent with Chinese water quality classes, we emphasized the importance of a river health assessment that integrates both chemical and biological parameters.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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