Impact factor analysis of aquatic species diversity in the Huai River Basin, China

Hao Chen, Qi-ting Zuo and Yong-yong Zhang

ABSTRACT

Water pollution has been a significant issue in the Huai River Basin (HRB) of China since the late 1970s. From December 2012, five experiments were carried out along the main streams of the HRB. The monitoring indices contained physicochemical variables, habitat environmental indicators and the community structure of phytoplankton, zooplankton and zoobenthos. The correlations between species diversity and physicochemical variables were analyzed using cluster analysis, correlation analysis method and redundancy analysis method. Results indicated that the species diversities of the Shaying River's upstream and Huai River's mainstream were better than the Shaying River's midstream and downstream. All the sections were divided into five clusters, and different clusters were affected by different physicochemical factors. Dissolved oxygen (DO), habitat quality index (HQI) and chemical oxygen demand (COD_{Cr}) were the main factors affecting the species diversity of the Shaying River's upstream; total phosphorus (TP), total nitrogen (TN), ammonia nitrogen (NH_4-N), COD_{Cr} and permanganate index (COD_{Mn}) had a great influence on the Shaying River's midstream and downstream; DO, water temperature (WT), HQI and COD_{Cr} were the main factors affecting the Huai River's mainstream. These results provide valuable information for policy decision makers and stakeholders on water quality assessment, water ecosystem restoration, and sustainable watershed management in the HRB.

Key words | cluster analysis, correlation analysis, Huai River Basin (HRB), redundancy analysis, species diversity

Hao Chen
Key Laboratory of Water Cycle and Related Land Surface Processes,
Institute of Geographic Sciences and Natural Resources Research,
Chinese Academy of Sciences,
Beijing 100101,
China
and
School of Water Conservancy,
North China University of Water Resources and Electric Power,
Zhengzhou 450045,
China

Qi-ting Zuo
School of Water Conservancy & Environment,
Zhengzhou University,
Zhengzhou 450001,
China
and
Center for Water Science Research,
Zhengzhou University,
Zhengzhou 450001,
China

Yong-yong Zhang (corresponding author)
Key Laboratory of Water Cycle and Related Land Surface Processes, Institute of Geographic Sciences and Natural Resources Research,
Chinese Academy of Sciences,
Beijing 100101,
China
E-mail: *zhangyy003@igsnrr.ac.cn*

INTRODUCTION

With the rapid growth of populations and the development of economies, mankind has carried out large-scale sluice and dam construction to develop water conservancy. However, this also has a variety of impacts on ecosystems, some of which are very sensitive, lasting and irreversible, the most important factor affected being ecological diversity. Globally, biodiversity ecosystems are under serious threat as a result of rapid population growth and increased human activities (Hill *et al.* 2015).

The most unusual feature of life on Earth is its diversity (Cardinale *et al.* 2012). The biological characteristics of a freshwater ecosystem are defined by the interrelationships between the entire complex of plants and animals living in water bodies and the physical and chemical conditions of organisms and water bodies and their catchment areas. These conditions are mainly influenced by climate, geographical location and type of water bodies (Kumar *et al.* 2015). Species are an important part of the ecosystem, and they provide the basic transfer of energy flow and information in material circulation and the ecosystem. When ecosystems lose some species, this can lead to imbalance in ecosystem functions, and even affect the survival and development of humankind. Biodiversity is crucial in ecosystem structure and processes, and the diversity of tribute species, which has complex ecosystem functions, is an important

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embodiment of biodiversity, and can reflect the function of the ecosystem to some extent.

Diversity indices are classic scalar ecological indicators. The application of these indices is common in ecological analysis. It is often found that species diversity indicates the status of the ecosystem or community (Diserud *et al.* 2002), and the quality of the living environment (Forio *et al.* 2017). Moreover, high species diversity contributes to the stability and health of the ecosystem (Sankaran *et al.* 1999; Moore 2005). In addition to some books, the methodology of diversity measurement in ecology is also discussed (Pielou 1975; Magurran 2004). Meanwhile, some articles analyze the influencing factors of biodiversity (Janse *et al.* 2015). Some people believe that habitat loss, pollution, and overexploitation are usually studied and managed in isolation, although the view of a single source of pressure is becoming increasingly apparent when ecosystems and species are threatened by a variety of common sources of stress (Darling *et al.* 2010). Therefore, species diversity will be affected by different impact factors in different watersheds.

The Huai River Basin (HRB) is a typical watershed in China, with a large population, many sluices and dams, serious pollution and so on. Over the past half-century, the HRB has been severely affected by human activities, especially the construction of dams and weirs and the discharge of pollutants (Zhang *et al.* 2013). Excessive sluice and dam construction hinder the flow of rivers, greatly affect the spatial and temporal distribution of pollutants, and endanger aquatic ecosystems. The long-term mismanagement of dams and sluices, combined with excessive pollution discharge, has led to severe degradation and extreme instability of ecosystems in many middle and lower river reaches (Zhang *et al.* 2015). Meanwhile, due to the downstream river flood control system, there are embankments on both sides of the Huai River, which further exacerbates the destruction of riparian habitats and seriously threatens the biodiversity of the lower reaches of the Huai River. Consequently, species diversity needs to be assessed to understand the environment and to rebuild or restore healthy aquatic ecosystems in the HRB.

However, because species diversity is influenced by different factors in different regions, the relationship between species diversity and impact factors cannot be fixed and applied to different periods and regions. Currently, these relationships are still at the exploration stage, and only to be given more attention in the HRB, along with the implementation of ecological conservation in China. The specific objectives of this paper were as follows: (i) to analyze the temporal–spatial variations of species diversity; (ii) to detect the relationship between species diversity and impact factors. These results provide valuable information for policy decision-makers and stakeholders in river ecosystem health assessment, water ecosystem restoration, and sustainable watershed management in the HRB.

MATERIALS AND METHODS

Study area

The Huai River (30°55′–36°36′N; 111°55′–121°25′E) is the sixth largest river in China and has many tributaries. The Shaying River is the largest and most seriously polluted tributary of the Huai River (Zuo & Li 2013). We examined the phytoplankton, zooplankton and zoobenthos communities and water quality conditions of the Shaying River and upper reaches of Bengbu Sluice in the Huai River (32°57′–33°42′N; 112°41′–117°17′E). The schematic of the study area and ten sections is shown in Figure 1.

Aquatic organism sampling and identification

To acquire experimental data, we carried out five experiments, namely, the first experiment (December 2012), second experiment (July 2013), third experiment (December 2013), fourth experiment (July 2014) and fifth experiment (December 2014), respectively. The sample collection, concentration and preservation methodologies for phytoplankton, zooplankton and zoobenthos are provided in *Water and Waste Water Monitoring Analysis Method* (4th edition) (State Environmental Protection Administration Water and the Waste Water Monitoring Analysis Method Editorial Committee 2002).

Phytoplankton sampling and identification

Water samples were collected in the 1,000 mL plastic bottles at about 0–2 m below the water surface, and

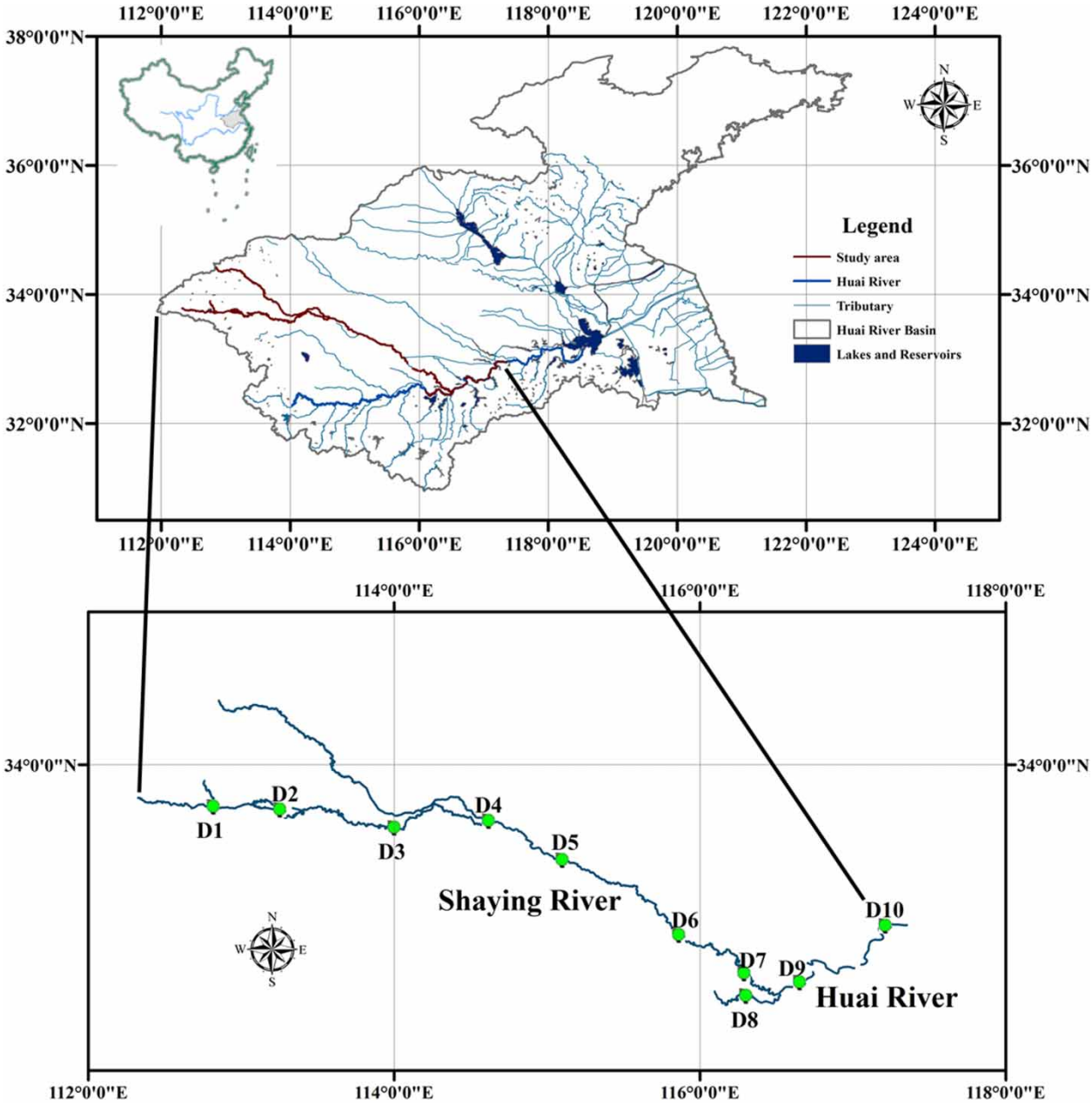

Figure 1 | Schematic diagram of the study area. D1~D3 sections are Shaying River upstream; D4~D7 sections are Shaying River midstream and downstream; D8~D10 sections are Huai River mainstream.

immediately preserved with a 1.5% concentration Lugol's solution. Then, a 24-hour sedimentation method was used to concentrate the sample to 30 mL, and 1 mL formaldehyde solution was added. The changes in total density and species number of phytoplankton are shown in Table 1.

Zooplankton sampling and identification

Water samples were collected in organic glass products about 0–2 m below the water surface, and a water specimen (50–100 mL) was obtained by 25 biological net (200 mesh) filtering water (50–100 L), then

Table 1 | Total density and species number changes of phytoplankton in the HRB

	Density (10^4 cell L^{-1})		Species number	
Sampling sections	Range	Mean ± SD	Range	Mean ± SD
D1	3.0–46.3	20.9 ± 14.7	5–23	12.4 ± 5.9
D2	18.3–588.8	199.0 ± 220.4	6–24	15.0 ± 6.6
D3	12.5–364.5	127.7 ± 123.3	13–26	19.0 ± 4.7
D4	23.7–894.7	434.3 ± 297.2	12–32	23.8 ± 7.4
D5	12.7–1,152.2	345.1 ± 414.1	17–33	25.0 ± 6.7
D6	43.3–280.4	161.5 ± 98.0	11–30	18.8 ± 6.5
D7	19.8–250.8	110.5 ± 80.9	12–29	19.6 ± 6.7
D8	1.6–109.2	35.9 ± 37.9	5–13	7.8 ± 2.7
D9	9.2–109.0	48.4 ± 40.4	4–21	12.8 ± 6.4
D10	13.2–44.7	22.5 ± 11.4	5–28	10.8 ± 8.6

samples were preserved with formaldehyde solution, about 5% of the sample amount. The changes of total density and species number of zooplankton are shown in Table 2.

Zoobenthos sampling and identification

A clam bucket collector or D-frame net was used to collect sediment samples. The samples were washed with water by a 60 mesh filter, and then the zoobenthos was picked out and fixed in 75% alcohol solution. The changes of total density and species number of zoobenthos are shown in Table 3.

Table 2 | Total density and species number changes of zooplankton in the HRB

	Density (cell L^{-1})		Species number	
Sampling sections	Range	Mean ± SD	Range	Mean ± SD
D1	1–6.5	3.2 ± 2.0	1–7	4.2 ± 2.3
D2	3–35.1	13.6 ± 12.5	4–13	7.2 ± 3.1
D3	6.5–30	17.2 ± 8.7	7–10	8.6 ± 1.4
D4	0.6–97.5	39.9 ± 37.7	1–14	8.0 ± 4.9
D5	2–376.8	91.9 ± 144.6	3–14	6.4 ± 4.1
D6	3–418.8	90.7 ± 164.1	4–12	7.2 ± 2.8
D7	2–109.8	32.8 ± 41.4	4–17	8.6 ± 4.7
D8	1–28.5	10.3 ± 9.6	2–12	6.6 ± 3.4
D9	0–54.6	21.0 ± 21.8	0–15	7.0 ± 5.3
D10	3–48.6	16.6 ± 16.5	3–15	8.0 ± 4.0

Table 3 | Total density and species number changes of zoobenthos in the HRB

	Density (individual m^{-2})		Species number	
Sampling sections	Range	Mean ± SD	Range	Mean ± SD
D1	42.8–216.8	100.1 ± 62.9	13–20	15.6 ± 2.4
D2	16.4–262.8	110.3 ± 96.1	5–12	9.2 ± 2.5
D3	6.4–124.8	36.6 ± 44.8	2–10	6.6 ± 2.8
D4	10–68.8	29.8 ± 23.3	3–8	6.2 ± 1.7
D5	1.6–126.8	31.1 ± 48.3	2–9	4.2 ± 2.6
D6	0.8–12.4	7.4 ± 3.9	2–7	4.4 ± 1.7
D7	0.8–151.2	48.5 ± 59.3	2–8	4.8 ± 2.0
D8	0.8–16.8	8.7 ± 5.7	2–5	3.8 ± 1.2
D9	1.2–26.8	11.9 ± 9.5	2–6	3.8 ± 1.8
D10	0–42.4	17.7 ± 16.4	0–9	4.2 ± 3.1

Sampling and detection of physicochemical variables

We measured water physicochemical indices *in situ*: water temperature (WT), pH, dissolved oxygen (DO), oxidation–reduction potential (ORP), and electrical conductivity (EC), using HACH HQ 30d and Hydrolab DS5. Meanwhile, water samples were collected with 1,000 mL plastic bottles, and NH_4-N, COD_{Mn}, COD_{Cr}, total phosphorus (TP), and total nitrogen (TN) were quantified in the laboratory immediately. Sample collection and testing methodologies were as provided in *Water and Waste Water Monitoring Analysis Method* (4th edition). Water physicochemical indices are shown in Table 4.

Environmental indicators of river habitats

The habitat quality was evaluated from ten parameters including substrate, habitat complexity, velocity–depth combination, bank stability, channel alteration, stream flow conditions, vegetation diversity, water quality conditions, the intensity of human activities and riverside land use. The habitat quality index (HQI) was calculated by the accumulative sum method, and the full score of ten indicators was 200 points (An *et al.* 2002). The HQI values are shown in Table 5.

Table 4 | Selected physicochemical water indices of the HRB

		First experiment		Second experiment		Third experiment		Fourth experiment		Fifth experiment	
No.	**Factors**	**Range**	**Mean ± SD**	**Range**	**Mean ± SD**	**Range**	**Mean ± SD**	**Range**	**Mean ± SD**	**Range**	**Mean ± SD**
1	WT	6.2–9.2	8.0 ± 1.00	28.9–33.9	31.6 ± 1.70	9.8–11.5	10.7 ± 0.72	27.6–29.4	28.3 ± 0.69	6.5–10.1	8.7 ± 1.07
2	pH	7.9–8.6	8.2 ± 0.19	6.5–8.9	8.0 ± 0.68	8.1–8.5	8.3 ± 0.15	7.8–8.6	8.2 ± 0.29	7.2–8.9	8.4 ± 0.58
3	DO	9.7–12	11.2 ± 0.84	2.0–17.4	9.2 ± 4.32	8.6–13.4	10.7 ± 1.32	3.7–16.7	8.2 ± 3.56	9.0–18.4	11.4 ± 2.58
4	NH_4-N	0.10–2.86	0.75 ± 0.81	0.13–0.81	0.34 ± 0.20	0.20–2.35	0.64 ± 0.63	0.13–0.50	0.31 ± 0.11	0.05–0.72	0.41 ± 0.25
5	COD_{Mn}	0.80–4.32	2.97 ± 1.15	1.21–5.41	3.30 ± 1.03	0.97–5.29	3.11 ± 1.32	0.98–4.86	3.47 ± 1.15	0.82–5.30	3.30 ± 1.33
6	COD_{Cr}	10.0–23.1	14.3 ± 3.88	10.0–14.6	11.2 ± 1.80	10.0–16.3	11.2 ± 1.87	12.6–21.6	16.9 ± 2.85	10.0–19.4	13.5 ± 2.99
7	TP	0.026–0.299	0.13 ± 0.078	0.021–0.197	0.09 ± 0.052	0.016–0.251	0.09 ± 0.071	0.021–0.385	0.16 ± 0.129	0.026–0.428	0.16 ± 0.116
8	TN	0.50–8.96	3.76 ± 2.30	0.69–4.28	2.41 ± 1.30	0.93–8.82	3.61 ± 2.09	0.97–4.46	2.54 ± 1.26	0.79–8.79	4.09 ± 2.35
9	ORP	83.2–289.2	178.5 ± 66.8	119.9–166.2	141.1 ± 15.9	201.3–246.7	212.5 ± 12.8	108.6–430.1	279.6 ± 115.5	39.4–111.0	66.7 ± 20.6
10	EC	297–1,055	695 ± 256	159–988	574 ± 299	309–1,126	682 ± 247	344–1,358	787 ± 383	288–1,215	732 ± 345

Unit of water temperature (WT) is degrees Celsius, oxidation reduction potential (ORP) is mV, electrical conductivity (EC) is $\mu s\ cm^{-1}$, pH has no unit, and the rest are all in $mg\ L^{-1}$.

Table 5 | Changes of *HQI* values in the HRB

	***HQI* values**	
Sections	**Range**	**Mean ± SD**
D1	112–165	140.2 ± 18.2
D2	82–154	118.4 ± 26.8
D3	68–121	101.6 ± 19.2
D4	73–129	108.8 ± 20.6
D5	82–140	109.2 ± 18.9
D6	112–140	119.0 ± 10.6
D7	110–138	128.8 ± 10.4
D8	103–141	129.6 ± 14.7
D9	108–146	125.0 ± 14.5
D10	115–170	136.8 ± 18.4

Methods

Species diversity analysis method

The biodiversity index is used to indicate the changes of community structure and reflect the changes in ecological status. We employed the commonly used Shannon–Wiener index (H) to estimate aquatic community diversity (Shannon 1948):

$$H = -\sum_{i=1}^{S}[(N_i/N)\log_2(N_i/N)] \quad (1)$$

where H is species diversity; N_i refers to the number of i species (individual L^{-1}); N is the total number of all species in a site (individual L^{-1}) and S refers to the species type number in a section. The species diversity indices of phytoplankton, zooplankton and zoobenthos are calculated, respectively, and then the total species diversity index is obtained by the weighted sum method. By consulting the relevant literature (Liu *et al.* 2008), the weight of the zoobenthos, zooplankton and phytoplankton diversity index is 0.5, 0.3, 0.2, respectively. When zoobenthos is absent, the weight of the zooplankton and phytoplankton diversity index is adjusted to 0.6, 0.4, respectively.

Correlation analysis method

Cluster analysis (CA) was performed on the species diversity data to determine the variables that were correlated and to summarize the species diversity characteristics of sections in an ordination diagram. The CA standardized each variable using z-score standardization and used Ward's method to execute the standardized dataset (Salah *et al.* 2012). The species diversity index was determined by linkage distance, and the normality of data distribution and the accuracy of clustering were analyzed by the 1-sample Kolmogorov–Smirnov (K-S) test.

The 1-sample K-S test and correlation analysis (Pearson) were used with SPSS 19.0 software. The normal distribution of data was analyzed by the 1-sample K-S test method, and the difference of species diversity index and environmental factors was analyzed. The significance level was set to

0.05. Pearson correlation analysis was applied to analyze the correlation between environmental factors and species diversity.

Redundancy analysis method

Species diversity and environmental factors were used to estimate sorting axial gradient length by detrended correspondence analysis. The results showed that the lengths of gradient were 0.585 (first experiment), 0.348 (second experiment), 0.513 (third experiment), 0.464 (fourth experiment) and 0.674 (fifth experiment), and they were all less than 3, indicating a linear response of species diversity to the ecological gradient. The species were subjected to centralized and standardized processing during data processing, and the sample was not centralized. A total of 499 runs of the Monte Carlo Permutation Test was used to investigate the species. The analysis process was completed with Canoco 4.5 software.

RESULTS AND DISCUSSION

Regional distribution of species diversity

Shaying River upstream

As can be seen from Table 6, the species diversity of Shaying River upstream was better than that of the other sections, especially the D1 section, which was the best in most cases (80%), and this was basically consistent with the research results of Liu *et al.* (2008). The main reason was that it had significant advantages in terms of species size, spatial distribution, and habitat diversity conditions (Nakano & Nakamura 2008). The range of species diversity was 1.76–3.01, and the first experiment had the best species diversity (3.010). The worst species diversity (1.760) emerged in the fourth experiment, with only one zooplankton detected, resulting in zero zooplankton diversity index and affecting species diversity throughout the section. Based on the results of CA (as shown in Figure 2), the D1 and D3 sections were in the same cluster, which was because they were both river habitats, and the river morphology and riverbank type were relatively close. However, the D3 section was in Luohe City with significant human effects, so the species diversities were worse than in the D1 section.

Shaying River midstream and downstream

The species diversities were poor in each section of Shaying River midstream and downstream, which was mainly due to serious river channelization and poor river channel bending (Allan & Flecker 1993; Dong 2003). In the first experiment, the species diversity of the D5 section was the worst (1.53). In general, the change of species diversity in the D5 section was not very obvious, the range being 1.53–1.9, because the gates of the sluices were closed, and the river habitat conditions did not change significantly in each experiment, but a

Table 6 | Temporal and spatial variation of species diversity

Sections	First experiment	Second experiment	Third experiment	Fourth experiment	Fifth experiment
D1	3.01	2.86	2.96	1.76	2.51
D2	2.30	1.34	1.76	1.62	2.03
D3	2.91	1.83	2.63	1.38	2.50
D4	1.99	2.42	2.77	1.14	0.94
D5	1.53	1.89	1.90	1.59	1.71
D6	1.85	1.63	2.54	1.91	2.06
D7	1.99	2.42	1.83	1.50	2.16
D8	2.03	1.64	1.30	2.14	1.56
D9	1.89	1.98	1.93	1.50	1.73
D10	2.20	1.41	2.74	1.36	1.57

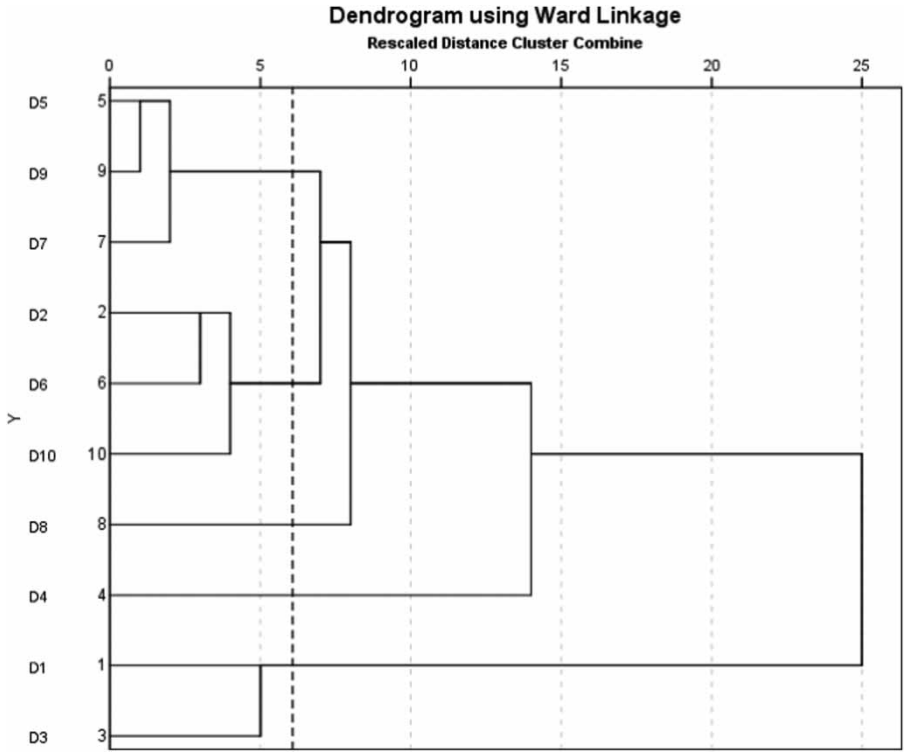

Figure 2 | Cluster analysis results for each section.

low flow was discharging (67 $m^3 s^{-1}$) in the third experiment, so it had better flow conditions. Compared with the D5 section, the species diversities of the D4 section were relatively higher, especially in the third experiment. There were many kinds of organisms (phytoplankton: 32 species, zooplankton: nine species, zoobenthos: seven species), and the species were well distributed.

Huai River mainstream

The species diversity of the three sections of the Huai River mainstream was worse than that of Shaying River upstream. The main reason was that the Huai River mainstream had embankments or slope protection, and the river channel was very straight, resulting in a reduction in the heterogeneity of the river habitat (Bis *et al.* 2000). The species diversity indices of the D8 and D10 sections varied sharply, in that the variation range of the D8 section was 1.3–2.14 and that of the D10 section was 1.36–2.74, and this was mainly because these sections were located downstream of the sluices and were affected by sluice regulation. But the changes of species diversity were not obvious in the D9 section (1.5–1.98), which is the mainstream section in the Huai River, and the influence of sluice and dam was bitty, which can provide more stable habitat conditions. In the fourth experiment. species diversity was the worst (only 1.5).

Impact factors of species diversity

Shaying River upstream

The ordering results of ecological and environmental factors are shown in Figure 3. In the fourth experiment, *H* and HQI present a significant positive correlation, as shown in Figure 3(d). The HQI of the fourth experiment was only 112, and *H* was also significantly affected by phytoplankton density and species number. However, the field experiment results had shown that the species number was less (nine species) and the density distribution was not uniform (the maximum density value accounts for 26% of the total density). In the three sections of Shaying River upstream, the species diversity in the D2 section was relatively poor, mainly because of the poor

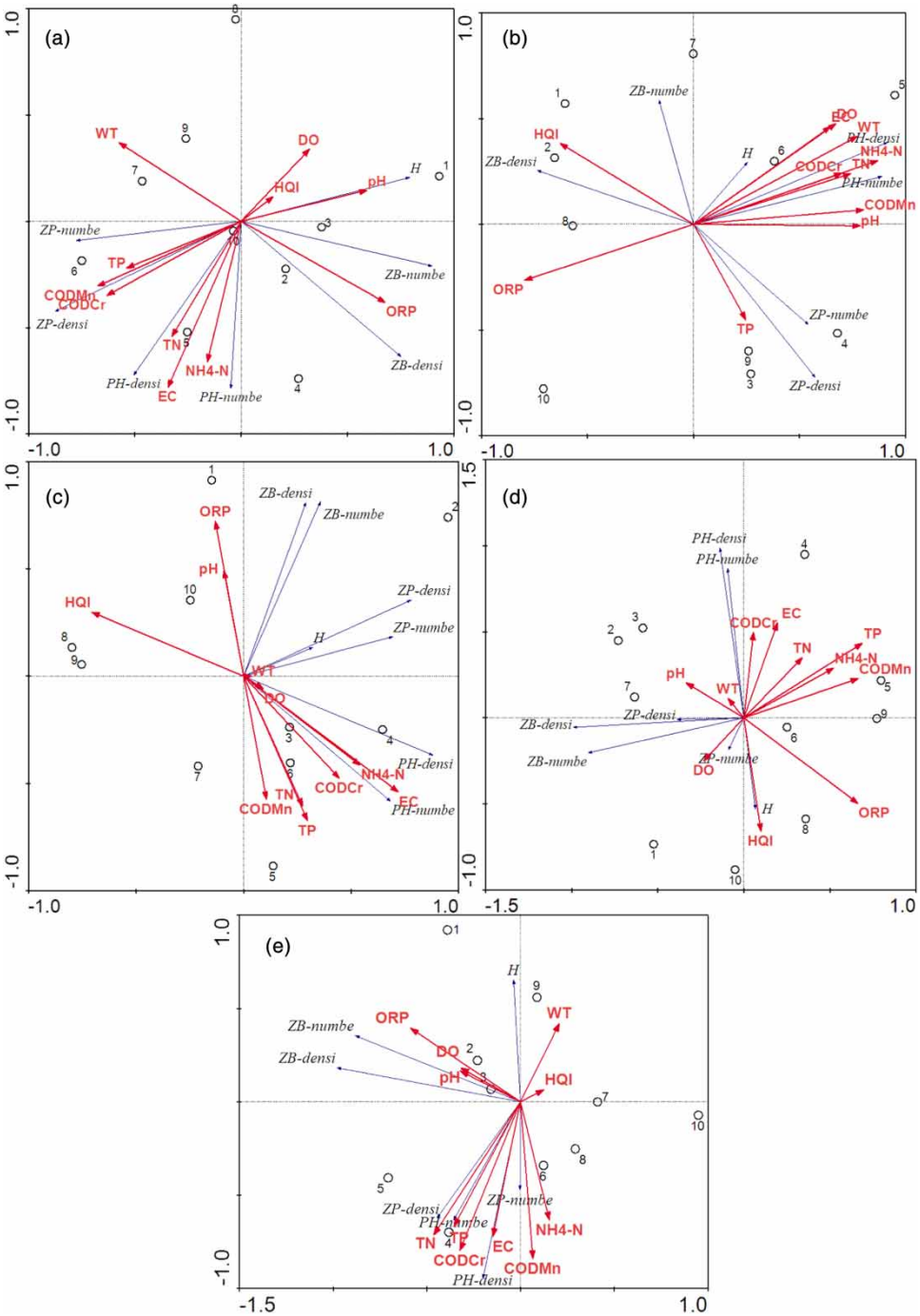

Figure 3 | The graph of RDA ordination for species diversity in the HRB in the five experiments: (a) first experiment; (b) second experiment; (c) third experiment; (d) fourth experiment; (e) fifth experiment. The hollow dot stands for section. The small solid arrow ray stands for the tendency of species diversity and ecological factors, and the large solid arrow line stands for environmental factors. The length of wire indicates the relationship among section, species distribution and environmental factors, and the direction of the arrow and angle with sorting axis indicates the correlation size of environmental factors and sorting axis, and the direction of the arrow indicates the increasing trend with environment variable values.

heterogeneity of habitats (narrow channel and fast water flow) and a large amount of aquaculture. Thus, these factors could have adverse affects on the survival of aquatic organisms, as in the second experiment where *H* and DO had a strong positive correlation, from Figure 3(b). However, DO concentration was too low (2 mg L^{-1}), which was not conducive to the

Table 7 | Effects of different physicochemical index on species diversity clustering results

Diversity index	NH_4-N	COD_{Mn}	COD_{Cr}	DO	TN	TP	pH	ORP	EC	WT	Chl a
H (5,9,7)	0.18	−0.13	−0.55*	0.52*	−0.20	−0.48*	0.09	−0.58*	−0.07	0.37	0.15
H (2,6,10)	0.33	−0.05	−0.07	0.35	0.29	−0.17	0.17	−0.02	0.17	−0.66**	0.10
H (8)	0.08	−0.41	0.82*	0.50	−0.73	0.69	−0.59	0.40	0.24	0.31	−0.22
H (4)	0.66	0.44	−0.80	0.27	0.08	−0.69	−0.38	0.44	−0.24	0.06	−0.06
H (1,3)	0.01	−0.55	−0.81**	0.55	0.15	0.02	−0.34	0.62	−0.22	−0.70*	−0.30

*Correlation is significant at the 0.05 level.
**Correlation is significant at the 0.01 level. *H* (number) stands for different clustering results.

survival of zooplankton and zoobenthos, so the zooplankton was only 3 individuals L^{-1}. Meanwhile, *H* had a positive correlation with species number and phytoplankton density. However, only six species of phytoplankton were detected in the second experiment, and the maximum density (*Phormidium tenus*) accounted for 56.3% of the total density values. The extremely uneven distribution of density will result in low species diversity. The Pearson correlation analysis method was used to analyze the impact factors on the species diversity indices, as shown in Table 7. COD_{Cr} is the main environmental impact factor in the D1 and D3 sections of Table 7, and the higher COD_{Cr} indicates that the organic pollution was more serious, possibly from pesticides, chemical plants, and organic fertilizer, which could cause persistent toxicity for aquatic organisms and the destruction of river ecosystems after the death of aquatic organisms. Therefore, we need to prevent organic pollutants from draining into these sections and improve the habitat environment of rivers.

Shaying River midstream and downstream

The D5 section was affected by TN, NH_4-N and EC, and these factors are negatively correlated with *H* in Figure 3(a). The concentrations of *TN* and NH_4-N in the first experiment were significantly higher than in the other experiments, and this led to poor species diversity in the D5 section of the first experiment. Based on the results in Table 7 and Figure 3, the diversity of the D5 section was mainly impacted by DO, COD_{Cr}, TP, TN and ORP, and this was basically consistent with the existing studies. For example, Zuo *et al.* (2019) found that DO, TP, TN and COD_{Mn} were the key influencing factors for phytoplankton diversity, zooplankton diversity and zoobenthos diversity, respectively. DO concentration was high (11.5 mg L^{-1}) in the third experiment, while COD_{Cr} and TP were relatively low, 11.6 mg L^{-1} and 0.166 mg L^{-1}, respectively. Meanwhile, *H* was positively correlated with zooplankton density and species number, and the distribution of zooplankton density was uniform, which led to better species diversity in the third experiment. Combined with the calculation results in Table 7 and Figure 3, comprehensive measures should be taken to improve species diversity in the middle reaches of Shaying River, such as restoring the natural condition of the river channel and reducing the pollutants in the water.

Huai River mainstream

The species diversity was the worst (only 1.5) for the D9 section in the fourth experiment (in Table 6). The main reasons were that *H* and HQI were positively correlated, but negatively correlated with the species number and density of phytoplankton and COD_{Cr} (in Figure 3), both HQI and COD_{Cr} were larger, and the distribution of density values of phytoplankton was very uneven. In addition, there was a significant negative correlation between species diversity and WT in the D10 section (Table 7), and this also led to better species diversity in winter than in summer, mainly due to the high temperature in summer, resulting in a large number of species, although the distribution of species was uneven. Combined with the calculation results in Table 7 and Figure 3, measures for controlling WT, improving river habitat environment and reducing the pollutant content should be adopted to improve species diversity in the Huai River mainstream.

CONCLUSIONS

We sampled phytoplankton, zooplankton, zoobenthos and water physicochemical variables to research the impact

factors for species diversity. This study can be regarded as a baseline survey for the future aquatic biological system and related ecological restoration in the HRB. Results showed the following:

(1) The species diversity of the Shaying River upstream and Huai River mainstream was better than Shaying River midstream and downstream. The species diversity of the D4 section was worst in the fifth experiment, and the species diversity of the D1 section was best in the first experiment.
(2) In the first experiment, *H* had the most obvious positive correlation with pH, HQI and DO, and was significantly negatively correlated with zooplankton density and species number; in the second experiment, *H* was most affected by EC, DO, phytoplankton density and species number; in the third experiment, *H* was affected by zooplankton density and species number, and negatively correlated with HQI; in the fourth experiment, *H* was most affected by HQI, and also greatly influenced by phytoplankton density and species number; in the fifth experiment, *H* was affected mostly by WT, and also significantly affected by the zooplankton species number and phytoplankton density values.

However, the relationships between diversity index and physicochemical variables are relatively complex, and thus, we will carry out more field experiments in the future, and collect more experimental data to analyze the relationships between them in more detail.

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