

Sediment heavy metals and benthic diversities in Hun-Tai River, northeast of China

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Abstract In aquatic ecosystems, metal contamination in sediments has become a ubiquitous environmental problem, causing serious issues. Hun-Tai River, located in northeast of China, flows through an important heavy industry region and metropolitan area. This study examined the heavy metals (Cd, Cr, Cu, Fe, Mn, Pb, Ni, and Zn) of sediments and diversities (taxa richness, Shannon diversity, and evenness) of benthic assemblages (benthic algae and macroinvertebrate) in Hun-Tai River. The results clearly described the spatial patterns of metal contamination in terms of geo-accumulation index and contamination factor, as well as the spatial patterns of benthic diversities in terms of taxa richness, Shannon index, and evenness by kriging interpolation. The sediments were largely contaminated by Cd, followed by Cu, Fe, Zn, Mn, and Ni. Cd and Zn had similar spatial patterns and similar sources. Cu, Fe, Mn, and Ni showed similar spatial patterns and similar sources. The surface sediments were unpolluted by Cr and Pb. The metal mines and the heavy industry in the major cities were the potential pollution sources. Benthic algae and macroinvertebrate responded similarly to the heterogeneous environment and metal contamination, with high taxa richness and Shannon index in middle-upper reaches of Hun-

Tai River. Evenness showed complex spatial patterns. Under low contamination, both taxa richness, Shannon diversity, and evenness had a large variation range. However, under the moderate and high contamination, the taxa richness and Shannon diversity kept to a low level but the evenness had a high level. This study provided insights into the sediment heavy metal contamination in Hun-Tai River.

Keywords Contamination level · Benthic diversity · Heavy metals · Surface sediment · Spatial pattern

Introduction

Heavy metal pollution has become a ubiquitous environmental problem and is one of the most serious global issues (Barbieri et al. 2014; Cheng et al. 2014; Hasegawa et al. 2016), due to their acute and chronic toxicity, persistence, and bioaccumulation (Tam and Wong 2000; Cui et al. 2011; Li et al. 2015; Qin et al. 2015). In aquatic ecosystems, contamination of sediments by heavy metals is one of the major threats (Vishnivetskaya et al. 2011; Skordas et al. 2015; Ma et al. 2016).

Heavy metals in sediment can be introduced from a variety of natural and anthropogenic sources. Generally, in uncontaminated aquatic ecosystems, the heavy metal concentrations are mostly controlled by geogenic origins including geologic weathering and atmospheric deposition (Négre et al. 1993; Zvinowanda et al. 2009; Varol and Sen 2012). The most significant anthropogenic sources of heavy metals are related with mining; agricultural practices; municipal, residential, and industrial wastes; and dry and wet deposition (Alexakis et al. 2012; Batista et al. 2012; Li et al. 2014; Zhu et al. 2016). Consequently, sediments are essential repositories of heavy metals derived from the drainage basin and, thus, indicators of contamination (Bettinetti et al. 2003; Farkas et al. 2007; Fan 2014; Ioannides et al. 2015).

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Benthic assemblies, such as periphyton and macroinvertebrate, play central ecological roles in aquatic ecosystems and are among the most ubiquitous and diverse organisms in freshwaters (Voelz and McArthur 2000; Strayer 2006). Benthic organisms usually have poor locomotion ability and different sensitivities to various environmental disturbances and thus are largely influenced by heterogeneous habitats and human impacts (Maret et al. 2003; Brederveld et al. 2011). Acting as sinks of natural and anthropogenic fluxes of heavy metals, surface sediments also provide important habitats and food sources for benthic organisms, and the heavy metals in sediments pose a threat to benthic assemblages (Begum et al. 2009; Boamponsem et al. 2010; Bentum et al. 2011; Bere et al. 2016). It has been suggested that the total abundance, taxa richness, community structure, and growth and production of macroinvertebrates decreased with increasing heavy metal concentrations (Maret et al. 2003; Sola et al. 2004; Gray and Delaney 2008; Qu et al. 2010). The effects of heavy metals on macroinvertebrates are highly variable among taxa (Kiffney and Clements 1996; Qu et al. 2010) and among functional feeding groups (Clements et al. 2000; Watanabe et al. 2008).

The present work is a case study focusing on heavy metals in sediments and benthic diversities of a highly polluted river located in the northeast of China, the Hun-Tai River. Hun-Tai River originates from Changbai Mountain where it has high forest coverage and flows through agricultural, industrial, and metropolitan areas. Various heavy metals have been introduced into the river. The analysis of the distribution of heavy metals in sediments and benthic diversities throughout the study area could be used for the assessment of heavy metal pollution and provide important information for environment protection. The quantification of toxicity of contaminated sediment can be assessed with the help of the geo-accumulation index (I_{geo}) and the contamination factor (CF) (Sekabira et al. 2010; Garcia-Ordiales et al. 2016). The benthic diversity was assessed with taxa richness, Shannon index, and evenness. The objectives

of our study were (1) to determine the concentrations and spatial contamination patterns of heavy metals (As, Cd, Cr, Cu, Fe, Mn, Ni, Pb, and Zn) in sediments of Hun River, (2) to characterize the spatial patterns of benthic diversities in terms of benthic algae and macroinvertebrate, and (3) to identify the relationships between metals contamination and benthic diversities.

Materials and methods

Study area

The Hun-Tai River, located in northeast China, belongs to the Liao River system and is consisted by the Hun River (415 km in length) and its main tributary, the Taizi River (413 km in length) (Fig. 1). The catchment area is 11,400 km², the average annual temperature is 9 °C, and the average annual precipitation is 686.4 mm. Hun-Tai River drains across an important heavy industry region, where it is the economic center of northeast China. Our study area covered the main metropolitan area of Liaoning Province, including the cities of Shenyang, Anshan, Fushun, Benxi, Liaoyang, Qingyuan, and Xinbin (Fig. 1). This area contains a variety of industries, such as equipment manufacturing, metal refining, and petrochemical industries, which strongly rely on the abundant local coal, iron, petroleum, and other mineral resources (Liu et al. 2011). Shenyang is the largest city in Liaoning Province and has an integrated industrial system, including equipment manufacture, medicine, chemical, steel manufacture, nonferrous metal processing, etc. Fushun is the main coal industry city in Liaoning Province. Benxi is famous for its coal and steel industries. Anshan is a famous steel industry city. Liaoyang is an industry base for petrochemical and chemical fiber industries. Except for the heavy industry, Liaoning Province is one of the main areas of mineral resources in China with more than 120 kinds of mineral resources (MLR 2013). In our study area, the coal mines are mainly located in

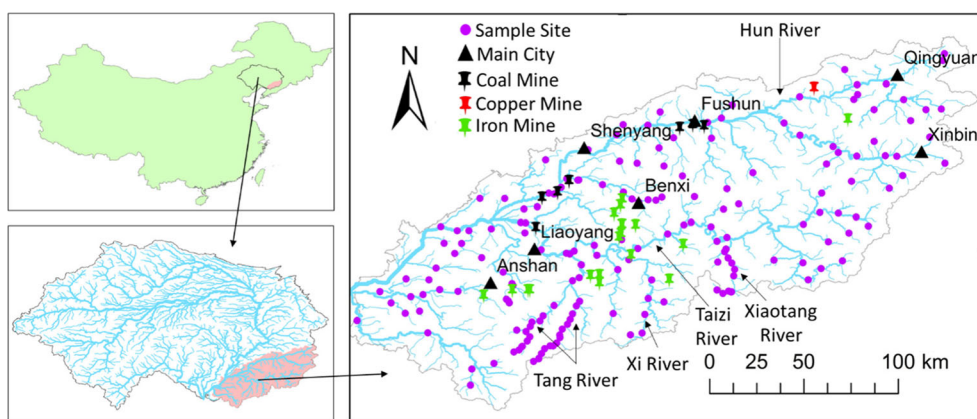


Fig. 1 The study area and sample sites. The total number site is 184 ($n_{total} = 184$), within which surface sediments were collected from 163 sites ($n_{sediment} = 163$), benthic samples were collected from 142 sites ($n_{benthic} = 142$), and 132 sample sites have both sediment and benthic

samples ($n_{both} = 132$). Purple dot represents the sample site; black triangle represents the main city; pushpin represents the main mine of coal (black), copper (red), and iron (green)

Shenyang, Liaoyang, Fushun, and Benxi. The iron mines are mainly located in Anshan, Benxi, Liaoyang, and Fushun. The main copper mine is Hongtoushan which is located between Fushun and Qingyuan. A large amount of pollutants has been discharged into Hun-Tai River via industrial wastewater, as well as municipal sewage (Guo and He 2013; Liu et al. 2015).

Field sampling

In total, 184 sampling sites were selected in Hun-Tai River, within which surface sediments were collected from 163 sites during the spring period (May and June) from 2009 to 2010 (Fig. 1). In 2009 spring, we collected the samples from the Taizi River watershed. In 2010 spring, we collected the samples from the Hun River watershed. The sampling time was concentrated within the spring season to avoid the influence of flooding during the summer-autumn period. The surface sediment samples were collected using a stainless steel container to a depth of approximately 5 cm. Sediment samples were kept in polyethylene bags and transported using an iced incubator. All samples were stored in a refrigerator at 4 °C for further analysis in the laboratory.

Meanwhile, periphyton and macroinvertebrates were collected from 142 sites by using quantitative methods. Nine stones were randomly picked (diameter ~ 25 cm) within a 300-m section of the sampling site. By using a 3.5-cm-diameter PVC pipe with a robber corer and a toothbrush, benthic algae were brushed thoroughly and rinsed with distilled water. Each sample and rinsed water was combined and preserved using a 4% formalin solution in a 50-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. By using the classic manuals of Hu et al. (1980) and Zhu and Chen (2000), most of the benthic algae were identified to the species level. Benthic macroinvertebrates were collected using a Surber net with three replicates (30 × 30 cm² and 500 μm mesh size). The substrates would be kicked or disturbed by using a shovel into the net. The sample is then transferred from the net to a 10-l plastic container. After rinsing all the samples through a 40-mm mesh with a stainless steel frame, all the samples were put into a 500-ml jar and preserved with 70% alcohol. In the lab, macroinvertebrates were identified to the lowest level, mainly to the genus level according to the manuals (Merritt and Cummins 1996; Morse et al. 1999). Finally, 132 sample sites have both sediment and benthic samples.

Sample analyses

Sediment samples were freeze dried, powdered, and sieved through a 100-μm-mesh nylon sieve. Sediment

subsamples of 0.1 g were collected and digested in Teflon vessels with 10 ml HNO₃ in a microwave oven (MARSX-press, CEM). Each sample solution was adjusted to 50 ml with double deionized water and then filtered through a 0.45-μm membrane filter for analysis. The sediment extracts were analyzed for Fe, Mn, Zn, Cu, Cd, Cr, Ni, and Pb using inductively coupled plasma-mass spectrometry (ICP-MS) (Agilent 7500CX).

Based on the laboratory quality assurance and quality control methods, the analytical data quality was guaranteed through the following processes: (I) use of standard operating procedures, (II) analysis of reagent blanks, (III) recovery of spiked samples, and (IV) implementation of triplicates. In order to evaluate the precision and recovery of the analysis procedure, a stream sediment reference material (GBW07310) was used following the course of analysis. The results showed that the analysis procedure is repeatable and reliable, with recoveries for all metals ranging from 96.1 to 105.4%. The relative standard deviation ranged from 5 to 10% for three replicates in the analysis. The results were expressed as the average values of the triplicates.

Statistical analyses

Monitoring spatial distribution and quantifying the degree of heavy metal enrichment are important for understanding and assessing anthropogenic inputs of metals into the aquatic ecosystem. In this study, a possible sediment enrichment of metals was evaluated by calculating the geo-accumulation index (I_{geo}) and the contamination factor (CF).

The I_{geo} , which was introduced by Muller (1969), can be used to determine the degree of heavy metal pollution in sediment. The formula to calculate the I_{geo} value is expressed as

$$I_{geo} = \log_2 \left(\frac{C_n}{1.5B_n} \right) \quad (1)$$

where C_n is the measured concentration of metal (n) and B_n is the geochemical background concentration of metal (n). The constant factor 1.5 is a background matrix correction that was used to minimize the effect of possible variations in the background values due to lithogenic effects and weathering (Müller 1986; Hasegawa et al. 2016). The sediment quality can be graded into seven classes (Müller 1969; Müller 1986): class 0, unpolluted ($I_{geo} \leq 0$); class 1, unpolluted to moderately polluted ($0 < I_{geo} \leq 1$); class 2, moderately polluted ($1 < I_{geo} \leq 2$); class 3, moderately to strongly polluted ($2 < I_{geo} \leq 3$); class 4, strongly polluted ($3 < I_{geo} \leq 4$); class 5, strongly to extremely polluted ($4 < I_{geo} \leq 5$); and class 6, extremely polluted ($5 < I_{geo}$).

The CF is another parameter which can be used to evaluate metal contamination in the environment. CF

Table 1 Summary statistics of sediment metal concentrations in Hun-Tai River (mg/kg by dry weight)

	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Minimum	0.00	8	7	8045	155	5	0.00	9
Maximum	5.06	76	254	88,370	3942	46	21.06	609
Mean	0.29	30	34	28,983	551	23	1.44	71
S.D.	0.47	11	31	12,455	394	8	3.54	92
CV (%)	165.65	38.33	89.43	42.97	71.36	36.29	245.55	130.20

can be calculated from the following equation given by Tomlinson et al. (1980):

$$CF = \frac{C_n}{B_n} \tag{2}$$

where C_n is the measured concentration of metal (n) and B_n is the geochemical background concentration of metal (n). CF was classified into four groups (Hakanson 1980; Tomlinson et al. 1980): $CF \leq 1$ indicates low contamination factor, $1 < CF \leq 3$ indicates moderate contamination factor, $3 < CF \leq 6$ indicates considerable contamination factor, and $CF > 6$ indicates very high contamination factor.

For diversity metrics, Shannon index (H) and evenness index (J) were calculated as follows:

$$H = - \sum_{i=1}^S P_i \ln P_i \tag{3}$$

$$J = \frac{- \sum_{i=1}^S P_i \ln P_i}{\ln S} \tag{4}$$

where S is the number of taxa found in each site and P_i is the proportional density represented by the i^{th} taxa.

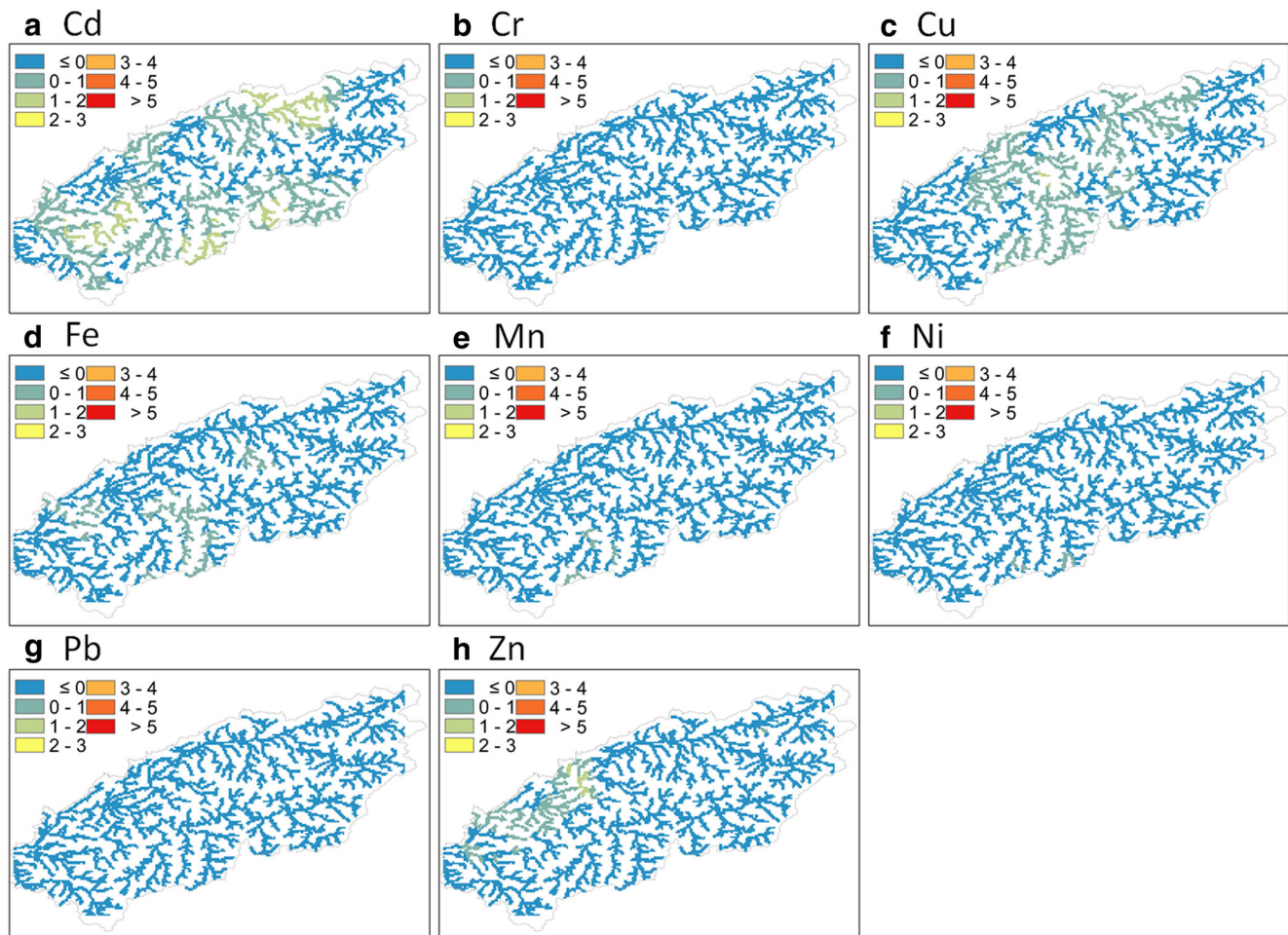


Fig. 2 Spatial patterns of sediment metal pollutions in terms of geo-accumulation index (I_{geo}) in Hun-Tai River

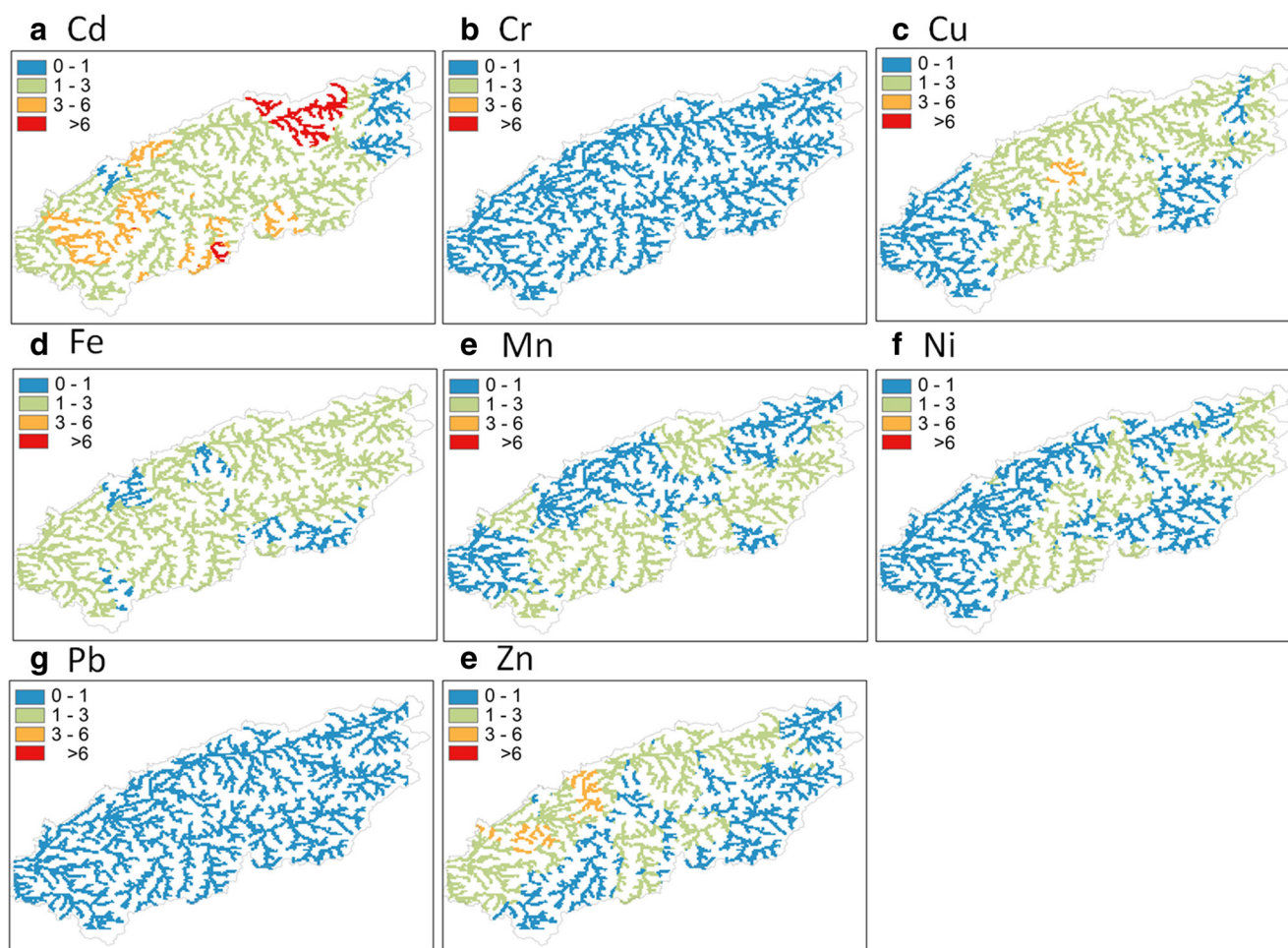


Fig. 3 Spatial patterns of sediment metal pollutions in terms of contamination factor (CF) in Hun-Tai River

The study of the spatial distribution metals in stream sediments and benthic biodiversity is considerably important for the assessment contamination. For mapping the spatial patterns of I_{geo} and CF for each metal and the diversities of benthic algae and macroinvertebrates, kriging interpolation

was applied by ArcGIS 10.3. The correlation analyses among different metals, between metal contamination indexes and benthic diversities, and between diversities of benthic algae and diversity between macroinvertebrate were conducted by SPSS 20.0.

Table 2 Pearson correlation coefficients of the metal concentrations in the sediments

Metal	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Cd	1.000	0.028	0.428**	0.080	0.064	0.064	0.111	0.480**
Cr		1.000	0.293**	0.305**	0.163*	0.645**	-0.127	0.146
Cu			1.000	0.292**	0.237**	0.438**	-0.058	0.404**
Fe				1.000	0.428**	0.204**	0.235**	0.321**
Mn					1.000	0.197*	0.137	-0.048
Ni						1.000	-0.085	-0.039
Pb							1.000	0.094
Zn								1.000

Levels of significance: * $P < 0.05$; ** $P < 0.01$

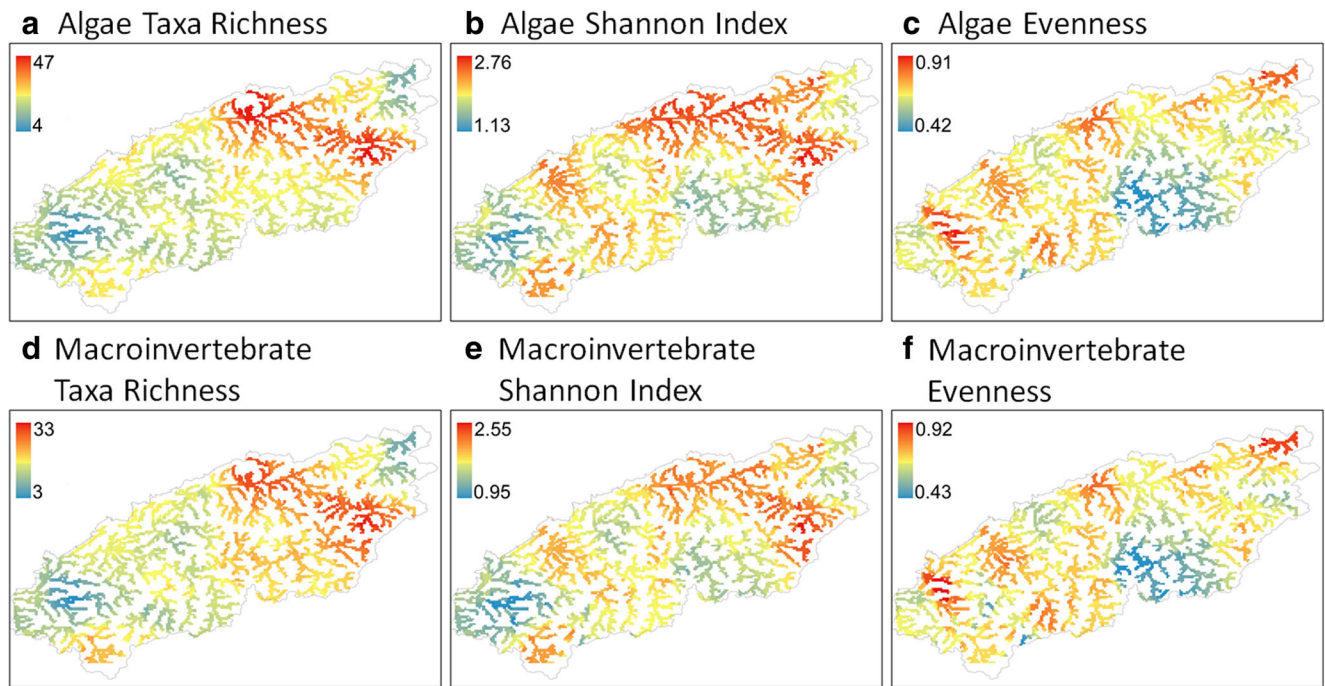


Fig. 4 The spatial patterns of diversities (taxa richness, Shannon diversity, and evenness) of benthic assemblages (benthic algae and macroinvertebrate) in Hun-Tai River

Results and discussion

Spatial patterns of sediment metal contamination

Sediment metal concentrations in Hun-Tai River were summarized in Table 1. Across the whole watershed, Cd, Pb, and Zn showed high variations with a coefficient of variation (CV) higher than 100% (Table 1). The degree of anthropogenic metal contamination in sediments of Hun-Tai River was assessed using I_{geo} and CF. From the pollution map of sediment metals made by kriging interpolation, we can clearly know the spatial pollution pattern of each individual metals in the study area (Figs. 2 and 3). In terms of I_{geo} , for Cd, 53% of the sediments had an $I_{geo} > 0$ and about 20% had an $I_{geo} > 1$, which are located in the middle-upper Hun River and upstream of Xi River (Fig. 2a), indicating that the sediments were moderately contaminated by Cd. For Cr and Pb, all of

the area had an $I_{geo} < 0$ (Fig. 2b, g), which means the sediments were not contaminated by Cr and Pb. For Cu, around 40% of the sediments had an $I_{geo} > 0$, which are located in the middle-upper reaches of Hun River and downstream of Taizi River. For Mn, Ni, and Zn, more than 78% of the sediments had an $I_{geo} < 0$ (Fig. 2c–f, h), indicating very slight contamination by these metals.

However, in terms of CF (Fig. 3), for Cd, 20% of the sediments were highly contaminated ($CF > 3$) which are located in the middle-upper and lower reaches of Hun River, and 51% of the sediments were moderately contaminated ($1 < CF \leq 3$) (Fig. 3a). For Cu, more than 70% of the sediments showed moderate or even high contamination (Fig. 3b). For Fe, 65% of the sediments were moderately or highly contaminated, except the downstream part of Hun River and the upstream part of Taizi River, where the sediments were lowly contaminated (Fig. 3c). Mn and Ni exhibited similar spatial contamination

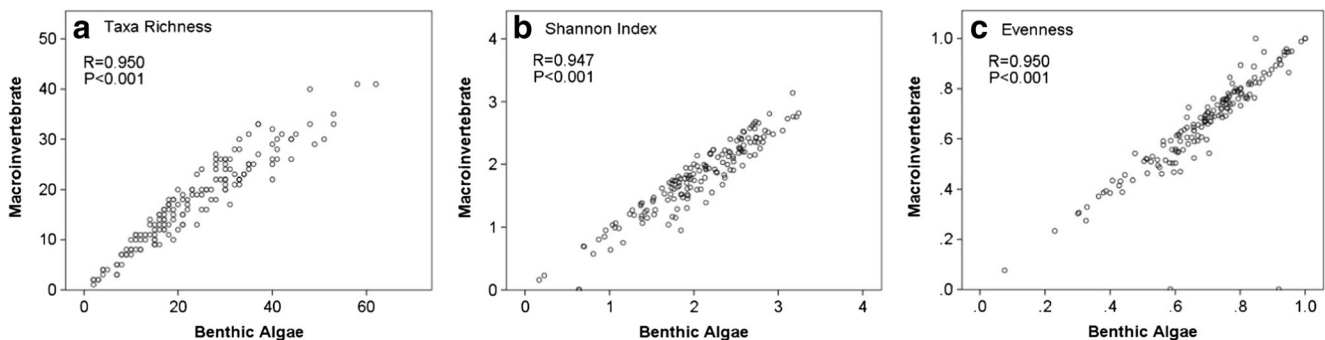


Fig. 5 The relationships between benthic algae diversities and macroinvertebrate diversities in terms of **a** taxa richness, **b** Shannon index, and **c** evenness

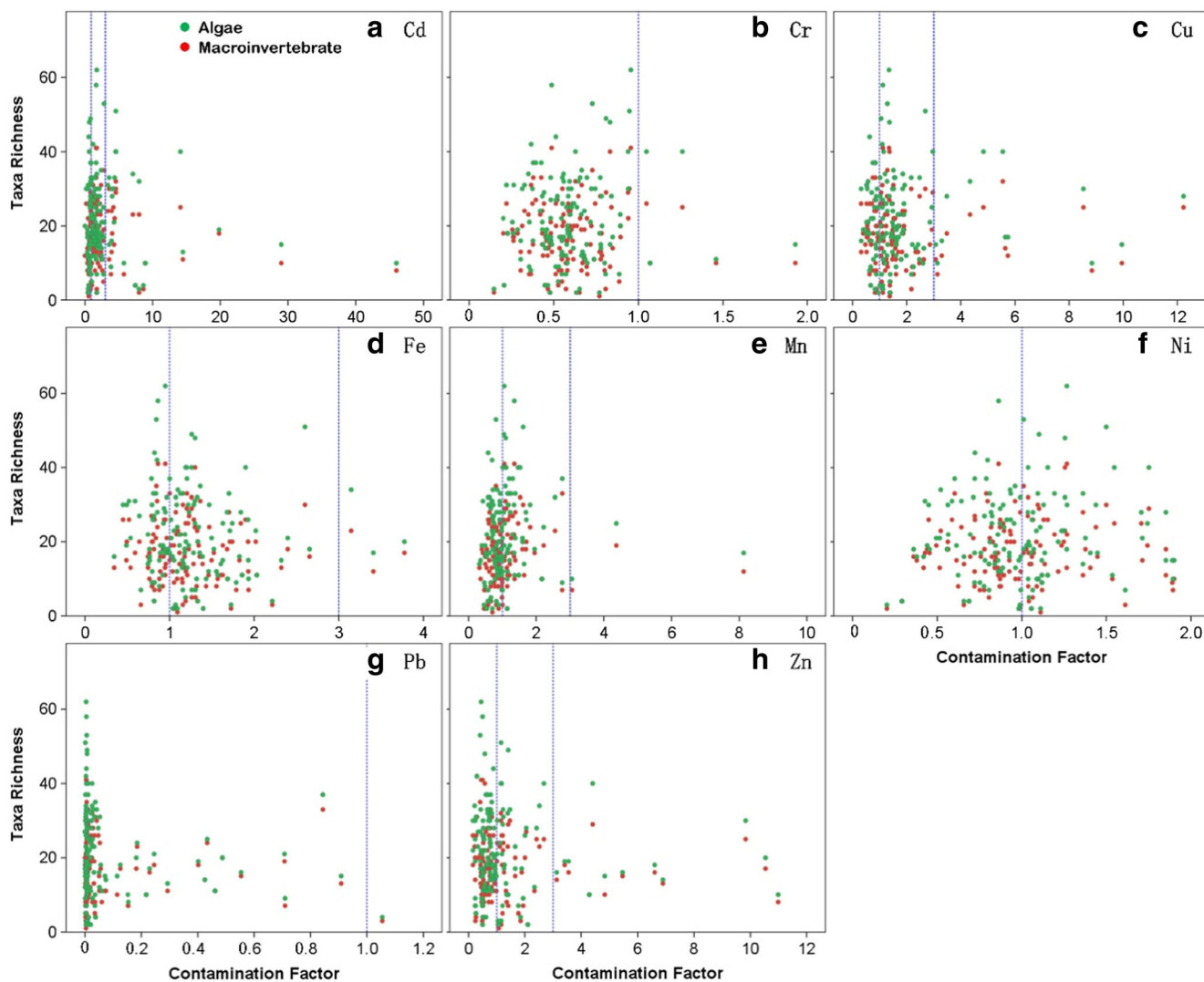


Fig. 6 The relationships between contamination factor and benthic taxa richness. The *green dots* represent benthic algae and the *red dots* represent macroinvertebrate. The *vertical dot lines* show CF = 1 and CF = 3, respectively

patterns with around 40% of the sediments moderately contaminated (Fig. 3e, f). For Zn, 29% of the sediments were moderately contaminated and 8% were highly contaminated (Fig. 3h), which are located in the middle and downstream parts of Hun River and Taizi River. Cr and Pb still showed unpolluted in almost the whole study area (Fig. 3b, g), which were similar to the results of I_{geo} . In general, the pollution patterns were more discriminated in terms of the CF than I_{geo} . In the following discussion, we mainly used the results of CF.

Controlled by numerous factors, such as the heavy metal contents in rocks and parent materials, soil formation processes, human contaminations, and other anthropogenic activities, heavy metals in sediments often exhibit high variations, different spatial patterns, and complex interrelationships (Zhao et al. 2009; Yi et al. 2011). The interelement relationships were analyzed by Pearson correlation analysis (Table 2) to assess possible similar metal sources (Garcia-Ordiales et al. 2014). The metals with a significant positive correlation may have similar

sources (Sekabira et al. 2010; Guo et al. 2013; Lim et al. 2013; Yan et al. 2014). In the sediments of Hun River, Cd and Zn showed a significant correlation ($R = 0.48$, $P < 0.01$). This could be explained by that Cd and Zn are congeners and always symbiose in nature. In the industrial process, Cd is a by-product of zinc refining. Thus, Cd and Zn are usually explored and processed together. Meanwhile, there were significant correlations among the concentrations of Cu, Fe, Mn, and Ni, which have shown similar spatial patterns, indicating that they may originate from similar pollution sources. Liaoning Province has very rich mineral resources with more than 120 kinds of mineral resources which have been discovered (MLR 2013). In our study area, iron is a widely distributed mine and is mainly located in Anshan, Benxi, Liaoyang, and Fushun (Fig. 1), consistent with the spatial pollution pattern of Fe (Fig. 3d). The iron mining and steel industries were the potential pollution sources of Fe, Zn, Mn, and Cu (Ni et al. 2007). This might explain that Zn had similar spatial pollution

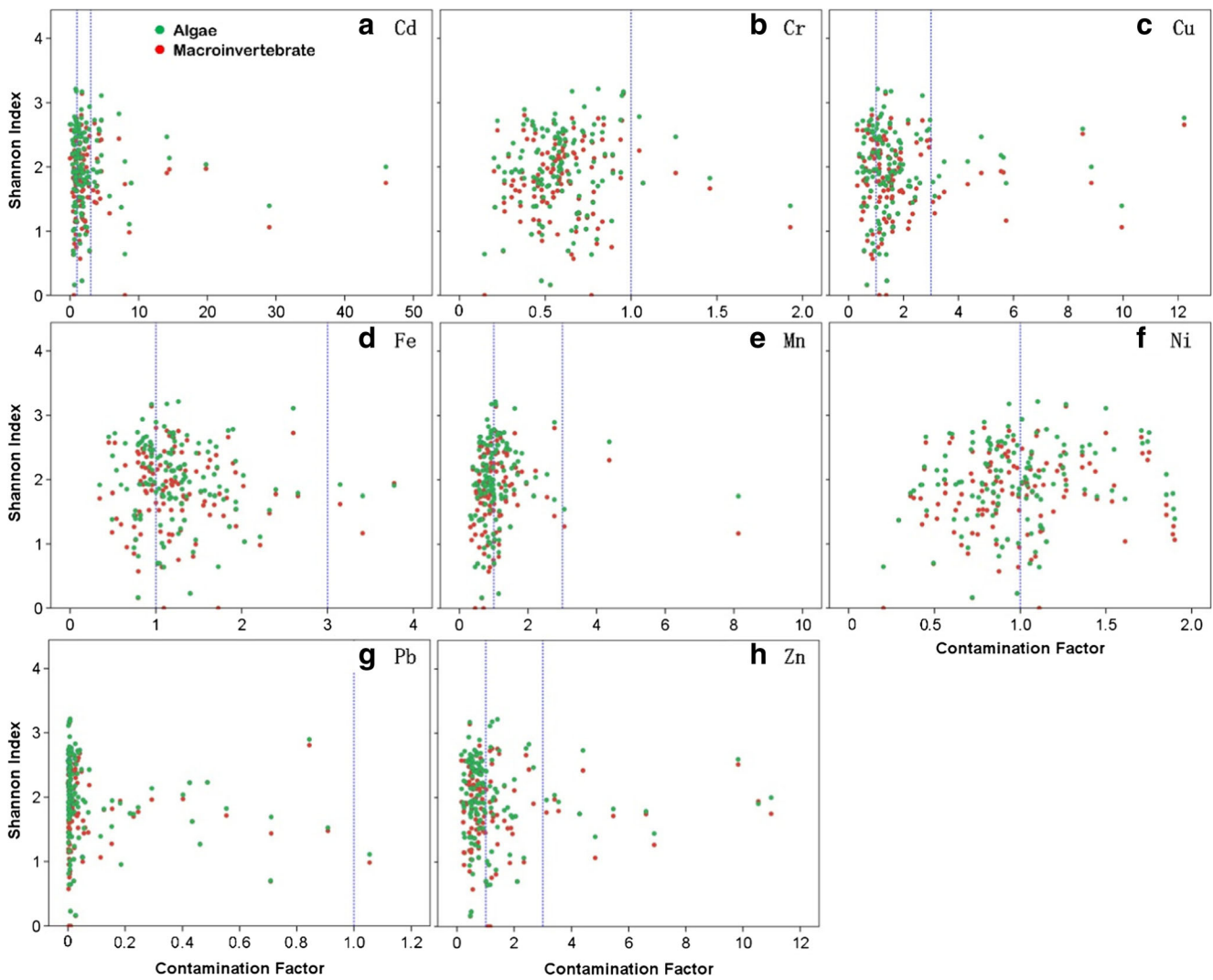


Fig. 7 The relationships between contamination factor and benthic Shannon diversity. The *green dots* represent benthic algae and the *red dots* represent macroinvertebrate. The *vertical dot lines* show CF = 1 and CF = 3, respectively

patterns with Fe, and the concentrations of Cu and Mn had significant correlations with Fe. In our study area, the main copper mine is Hongtoushan, which is located between Fushun and Qingyuan (Fig. 1). It has been studied that in the soil around this copper mine, the main heavy metal pollutions were Cd, Zn, and Cu (Shi et al. 2008), which was consistent with our study (Fig. 3a). Besides zinc mining and copper mining, Cd is widely used in nonferrous metal smelting as well as the electroplating industry, chemical industry, electronics industry, etc. The wide pollution sources explained the spatial pattern of Cd pollution in sediments (Fig. 3a). The major cities in this area have diversiform heavy industries, including equipment manufacture, medicine, chemical, steel manufacture, nonferrous metal processing, coal, petrochemical and chemical fiber, etc. The industrial wastewater and municipal sewage were another main potential pollution of various heavy metals (Guo et al. 2013; Liu et al. 2015).

Spatial patterns of benthic diversity

The spatial patterns of benthic diversity were shown by kriging interpolation using different metrics (taxa richness, Shannon index, and evenness, Fig. 4). Both benthic algae and macroinvertebrate have higher taxa richness in the middle-upper reaches than downstream (Fig. 4a, d). Shannon index also showed similar patterns with taxa richness and have high values in the middle-upper and middle-lower reaches (Fig. 4b, e). However, the evenness showed very complex spatial patterns (Fig. 4c, f). The linear regression showed that macroinvertebrate and benthic algae have significantly strong relationships with each other in terms of taxa richness, Shannon index, and evenness (Fig. 5), indicating that they responded similarly to the heterogeneous environment and metal contamination. According to the distribution map of main cities and mines in the study area (Fig. 1), the main cities,

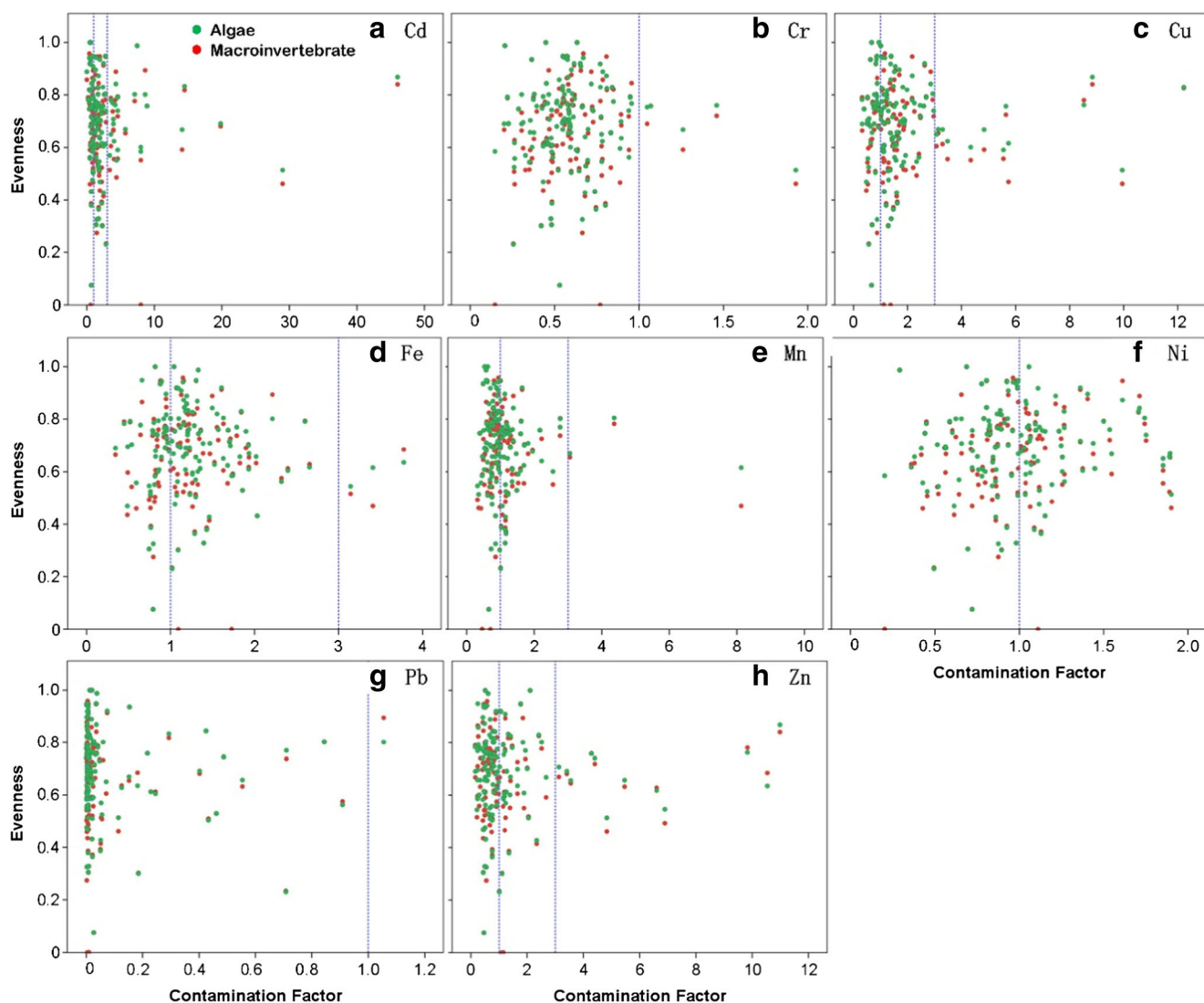


Fig. 8 The relationships between contamination factor and benthic evenness. The *green dots* represent benthic algae and the *red dots* represent macroinvertebrate. The *vertical dot lines* show $CF = 1$ and $CF = 3$, respectively

coal mines, and iron mines are mainly distributed in the middle and downstream parts of Hun-Tai River, consistent with the spatial patterns of benthic diversities (Fig. 4). However, there was no remarkable consistency when comparing the spatial patterns of metal pollutions in sediments (Fig. 3) and the spatial patterns of benthic diversities (Fig. 4). It suggested that the synthetic influences of pollutions from industries and mining affect the diversity patterns of benthic assemblages.

The relationships between sediment metal pollution and benthic diversities

The relationships between sediment metals and benthic diversities showed that, due to their sensitivities to heterogeneous habitats and human impacts, the benthic taxa richness (Fig. 6), Shannon diversity (Fig. 7), and evenness (Fig. 8) have a large variation range from very low to very high levels when the

sediment metals were unpolluted or lowly polluted. In this study, for most of the metals, when the sediments show moderate contamination ($1 < CF \leq 3$) or high contamination ($CF > 3$), the benthic taxa richness (Fig. 6) and Shannon diversity (Fig. 7) kept to a low level. While for evenness (Fig. 8), even when the contaminations were high, the evenness still had a high level. This suggested that in the highly contaminated environment, only certain taxa have high relative abundance while others have low relative abundance. It has already been known that heavy metal pollution can alter benthic diversity as well as community structure (Lefcort et al. 2010; Hamidian et al. 2016). Benthic organisms have different sensitivities to various environmental disturbances/pollutions and have poor locomotion ability, and thus, some species may dominate in a polluted environment due to their high tolerance (Whitton et al. 1981; Costello and Burton 2014; Podda et al. 2014; Plachno et al. 2015). For example, *spirogyra*, *euglenophytes*,

and *chironomids* have shown high tolerance to various kinds of metal contamination (Albergoni et al. 1980; Plachno et al. 2015; Hamidian et al. 2016). In highly contaminated sites, these highly tolerant benthic organisms can aggregate metals within their organs and potentially threaten the health of many organisms at high trophic levels (Klavins et al. 1998; Demirak et al. 2006; Hamidian et al. 2016). While Ephemeroptera, Plecoptera, and Trichoptera (EPT) are the most sensitive stream insect families (Cain et al. 1993; Pollard and Yuan 2006; Lefcort et al. 2010; Costello et al. 2014). Heavy metals mainly affected the sensitive taxa, and correspondingly, a shift in community composition from sensitive to tolerant taxa can occur when aquatic ecosystems are contaminated by heavy metal (Beltman et al. 1999; Qu et al. 2010).

Conclusions

In this study, various analysis methods have been employed for understanding the metal contaminations in sediments and benthic diversities of Hun-Tai River. The geo-accumulation index and the contamination factor have been employed for evaluation of sediment contamination in Hun-Tai River, and kriging interpolation was applied for mapping the spatial patterns of contamination. Our results demonstrated that the CF was more discriminated than I_{geo} in contamination assessment, although they have similar results. The sediments were largely contaminated by Cd, followed by Cu, Fe, Zn, Mn, and Ni. Cr and Pb were unpolluted in almost the whole study area. Cd and Zn had similar spatial patterns and might have similar sources. Cu, Fe, Mn, and Ni showed similar spatial patterns and similar sources. The metal mines and the heavy industry in the major cities were the potential pollution sources. Benthic algae and macroinvertebrate responded similarly to the heterogeneous environment and metal contamination. The Shannon index and taxa richness had similar spatial patterns with high values in the middle-upper reaches of Hun River. Evenness showed complex spatial patterns. The benthic taxa richness and Shannon diversity had a large variation range when the sediment metals were unpolluted or low polluted and kept to a low level when the sediments show moderate or even high contamination. The evenness had a large variation range when the contaminations were low, while when the contaminations were high, the evenness had a high level.

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