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Development and evaluation of a diatom-based index of biotic integrity (D-IBI) for rivers impacted by run-of-river dams

Naicheng Wu[a,b,*], Qinghua Cai[a,*], Nicola Fohrer[b]

[a] State Key Laboratory of Freshwater Ecology and Biotechnology, Institute of Hydrobiology, Chinese Academy of Sciences, Wuhan, Hubei 430072, PR China
[b] Department of Hydrology and Water Resource Management, Institute for the Conservation of Natural Resources, Kiel University, Kiel 24118, Germany

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ABSTRACT

Significant ecological effects of run-of-river dams on stream chemistry, benthic algae, phytoplankton, macroinvertebrates, macrophytes and fish have been found. However, the quantitative impacts and intensity of such impacts on aquatic ecosystems are still unclear and there was no assessment system to examine such impacts effectively. We investigated benthic diatom communities of 23 run-of-river dams in a Chinese river during the dry season with the goal of developing a diatom-based index of biotic integrity (D-IBI) that would be sensitive to such impacts. Four metrics were selected from 110 diatom attributes of training-site data set to construct the D-IBI based on: (1) significant differences between reference and impaired sites, (2) high separation power and (3) a low coefficient of variation (CV). We then calculated the total D-IBI scores by summing metrics for each site after transformation by 0–10 scaling system. The D-IBI and its metrics were tested using an independent testing-site data set from a tributary of the study catchment. The test results, calculated using the same criteria and scaling system as the training-site data set, indicated significant differences between reference and impaired sites. The developed D-IBI was powerful in terms of separation powers, % sites correctly classified, box-separation ratios, correlation index (CoI) and Cumulative_R^2. Then the ecological conditions of the sampling sites were evaluated based on the total D-IBI scores. Overall, the ecological conditions of references sites were "Good". D-IBI scores were significantly reduced at impaired sites, where affected by the dams, compared with references sites. The newly developed D-IBI effectively signaled impairment of flow regulations in the Xiangxi River of China, and it could be used in future studies measuring the long-term status of streams and the effectiveness of various remediation measures. However, further testing and assessment of their applicability in other regions impacted by small run-of-river dams are still needed.

1. Introduction

All parts of a river ecosystem are inter-connected and any disturbance to one part will create a greater or lesser impact on the whole system (Almeida et al., 2009; Wu et al., 2010a). Run-of-river dams can disrupt the river's natural course and flux of water, impede the cycling of organic matter, sediments and nutrients, and strongly alter the structure and dynamics of aquatic and riparian habitats and biota upstream and downstream by increasing the fragmentation of habitat with associated isolation of populations (Winston et al., 1991; Thomson et al., 2005; Wu et al., 2009). Ultimately, such dams disturb ecological connectivity, which underpins the transfer of materials and products (e.g., biota, nutrients, organic matter, or energy) of ecological functions and processes from up- to downstream. In aquatic ecosystems, the connectivity is mediated by flows and hydrological linkages (Jenkins and Boulton, 2003), which would likely have significant effects on the structure and function of river organisms (Kobayashi et al., 1998; Moreau et al., 1998; Tang et al., 2006). As a consequence, the impact of dam construction and flow diversion are receiving increasing attention (Benstead et al., 1999). Many studies have examined the impacts of such dams on stream chemistry (Velinsky et al., 2006), benthic algae (Thomson et al., 2005; Morley et al., 2008; Wu et al., 2009, 2010a,b), riverine phytoplankton (Wu et al., 2007), macroinvertebrates (Sharma et al., 2005; Fu et al., 2008a; Almeida et al., 2009), aquatic vegetation (Ot'ahel'ová and Valachovič, 2002) and fish (Cumming, 2004). For example, NH_4^+ was significantly influenced by dam removal (Velinsky et al., 2006), and Thomson et al. (2005) found that macroinvertebrate density and algal biomass declined significantly following dam removal. Furthermore, increased diatom species richness was also found after dam construction (Wu et al.,

* Corresponding authors at: State Key Laboratory of Freshwater Ecology and Biotechnology, Institute of Hydrobiology, Chinese Academy of Sciences, Wuhan, Hubei 430072, PR China.
E-mail addresses: wunaicheng2003@yahoo.com.cn (N. Wu), qhcai@ihb.ac.cn (Q. Cai).

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2010b). However, the quantitative impacts and intensity of run-of-river dams on aquatic ecosystems are still unclear and there was no assessment system to examine such impacts effectively.

Multi-metric indices are increasingly used to assess the ecological status of rivers as well as resource and ecosystem management because they are often more robust than their component metrics (Lacouture et al., 2006), and represent different taxonomic and functional groups within the assemblage, which respond differently to various stressors and can reflect ecological status in a comprehensive manner (Blanco et al., 2007; Zalack et al., 2010). As a multi-metric approach, index of biotic integrity (IBI), originally developed by Karr (1981), has become the most common indicator of aquatic conditions in use today. To date, many assessment methods based on IBI have been developed in several countries and regions for different impairments (e.g. Hill et al., 2000, 2003; Wang et al., 2005; Tang et al., 2006; Zhu and Chang, 2008; Rothrock et al., 2008; Kane et al., 2009; Bae et al., 2010; Li et al., 2010; Zalack et al., 2010; Wu et al., 2012). Nevertheless, to our knowledge, there was no IBI for assessing the impacts of run-of-river dams.

Benthic diatoms, the main primary producers of stream ecosystems, possess many attributes that make them ideal organisms in assessing ecological condition of lotic waters (Tang et al., 2006). For example, they are readily dispersed and can invade a variety of habitats; samples are easy to handle and create minimal impact to resident biota during collections; they are sensitive to subtle changes in environmental conditions and/or disturbances that may not visibly affect other communities or may only affect other communities at greater levels of disturbance (Lowe and Pan, 1996; Stevenson and Bahls, 1999; Stevenson and Pan, 1999; Hambrook, 2002). Therefore, diatoms are increasingly being used as bio-indicators for environmental monitoring (Stevenson and Smol, 2002; Wu et al., 2010b).

The Xiangxi River is the largest tributary near the Three-Gorge Dam (TGD) in Hubei Province, with a length of 94 km and a catchment area of 3099 km^2. Many run-of-river dams have been built within the watershed and the series of small cascade dams has become one of the main human disturbances. Former studies in the Xiangxi River suggested that the run-of-river dams had negative effects on aquatic organisms, especially in dry seasons or during long periods of drought (Zhou et al., 2008; Wu et al., 2010a). In this paper, a part of the series studies in the Xiangxi catchment (e.g. Wu et al., 2007, 2009, 2010a,b; Fu et al., 2008a,b; Zhou et al., 2008), we investigated 23 run-of-river dams during the dry season (October 2005) with the goal of developing a diatom-based index of biotic integrity (D-IBI) that would be sensitive to the impacts of run-of-river dams and examine ecological status of the study area by implementing the developed D-IBI.

2. Methods

2.1. Study area and site locations

Originating from Shennongjia Mountain (at 3150 m, the highest mountain in central China), the Xiangxi River has three main tributaries – Jiuchong, Gufu and Gaolan Rivers (Fig. 1B). Many small run-of-river dams have been constructed along this river, and in dry seasons much of the flow is abstracted and diverted through a special penstock to power plants, leaving the main channel with only a small part of the original flow.

We investigated 23 dams in October 2005, and at most, four sites were established at each river segment (Fig. 1C), and each of them was designated as either reference (Re) or impaired (Im) based on its habitat. Re was located approximately 50–100 m upstream of the dam where river channels were free of impacts from the dam. Im1 was in a pool below the dam, which was formed by overflow erosion in rainy seasons, and no velocity was detected during the sampling period. Im2 was upriver from the outlet of the small hydropower station (SHP), and flow was recovered after water gathering downstream from Im1. Im3 was at the outlet of the SHP, where powerful currents discharging from the outlet had formed a deep pool. Due to cascade construction of dams, sampling sites at upstream SHPs usually overlapped with the site of an adjacent dam (Fig. 1C). Three Re sites were not reachable because of road limitation. Therefore, a total of 71 sites were finally visited, which were divided into either training-site (development of D-IBI) or testing-site data set (testing D-IBI). Twenty-nine sites (Re, 9; Im, 20) located in Gaolan tributary were sorted as testing-site data set, while the other 42 sites (Re, 11; Im, 31) were used as training-site data set (Fig. 1B).

2.2. Field sampling and processing

Algae were collected from all available substrates and habitats. The objective was to collect a single composite sample that represented the benthic algae community in the reach. Three to five representative stones (diameter <25 cm) were collected from each section, and the surface area within a 2.7-cm diameter corer was brushed thoroughly and then rinsed with 350-ml distilled water. The sample was divided into two parts: one was preserved with 4% formalin for identification and the other was filtered through a Whatman GF/C filter for chlorophyll *a* (Chl-*a*) measurement. The filters were frozen in a dark container until further treatment. In the laboratory, extraction was performed by adding 90% acetone (lasted for 20–24 h), followed by centrifugation to produce a supernatant with minimal turbidity. Then, the absorbance of the processed samples was recorded at four different wavelengths (630, 645, 665 and 750 nm) on a spectrophotometer (Shimadzu UV-1601, Japan) for calculating Chl-*a* contents.

Identification of benthic algae involved two steps. First, we analyzed non-diatom algae with a 0.1-mL counting chamber and a microscope at 400× magnification. Second, we prepared permanent diatom slides after oxidizing the organic material with acid. We counted a minimum of 300 valves at 1000× magnification under oil immersion. Algae were identified to the lowest taxonomic level possible and their densities were expressed as ind./m^2.

2.3. Measurement of physicochemical factors

Values of pH, dissolved oxygen (DO), conductivity (COND), Chloride (Cl^-), calcium (Ca^{2+}), total dissolved solid (TDS), water temperature (WT), oxidation–reduction potential (ORP) and turbidity (TURB) were measured *in situ* with a Horiba W-23XD (multiprobe sonde). Water depth, channel width and current velocity (LJD-10 water current meter; Chongqing hydrological machines manufactory, Chongqing, China) were also measured at each site. We collected ~1 L of water in pre-cleaned plastic containers to measure chemical variables, including total phosphorus (T-P) and orthophosphate (PO_4-P). Samples were stored in the dark at 4 °C until the measurement in the laboratory, using the ammonium molybdate spectrophotometric method (at 700 nm; GB11893–89).

2.4. Development of D-IBI

Using 42 training sites (11 reference sites and 31 impaired sites), we developed the D-IBI based on the methods described by Wang et al. (2005), but with some modifications. We firstly compiled a large pool of attributes (totally 110 metrics) according to literatures (e.g. Menhinick, 1964; McNaughton, 1967; Berger and Parker, 1970; van Dam et al., 1994; Kelly and Whitton, 1995; Hill et al., 2000, 2003; Wang et al., 2005; Tang et al., 2006; Spatharis and Tsirtsisa, 2010), which were classified into 7 categories: biotic index, diversity index, evenness index, dominance index, growth

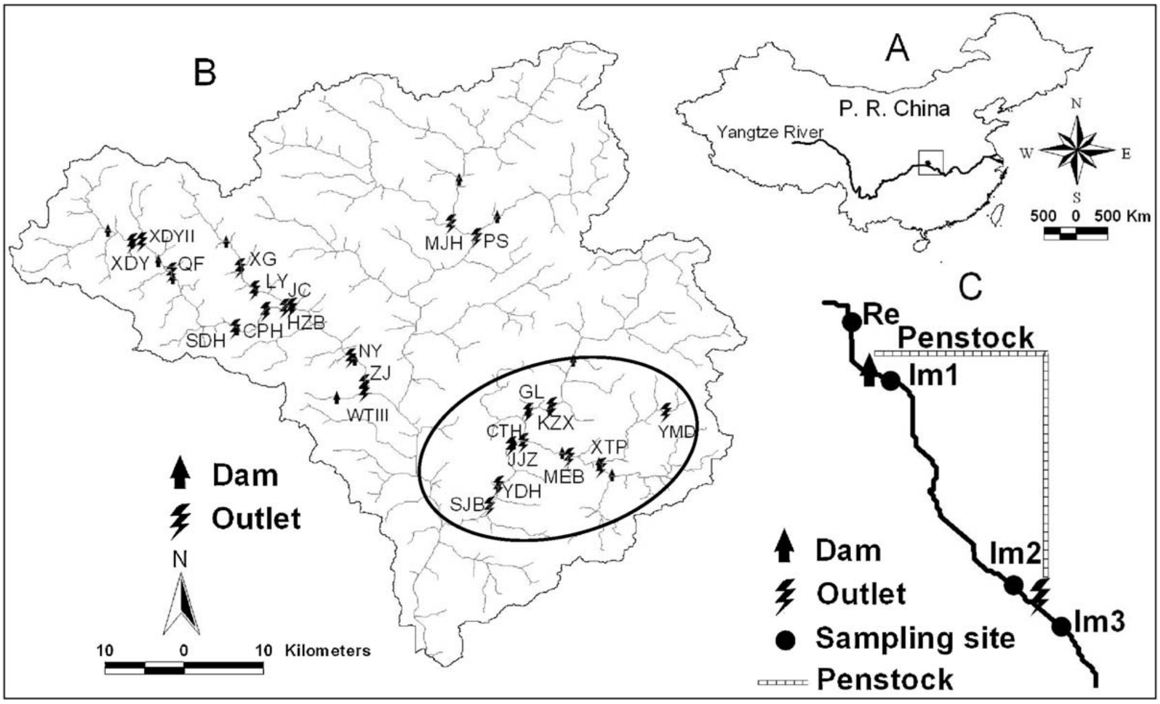

Fig. 1. Small run-of-river dams within the Xiangxi River watershed (B) in the People's Republic (P. R.) of China (A) and sketch map of sampling sites (C). Letters in B are names of small hydropower stations (SHPs); sites located in Gaolan tributary (circle of B) were sorted as testing-site data set.

form index, similarity index and taxonomic composition (Appendix A). Then metrics were selected based on the following analyses.

To begin, attributes with medians of 0 were eliminated because they would prevent identification of differences between these two groups (Wang et al., 2005). Their ability of remaining attributes to separate reference and impaired sites was evaluated by non-parametric Kruskal–Wallis test, since many metrics did not follow normal distributions (Kolmogorov–Smirnov test, $p < 0.05$) despite trying several transformations. We identified potential metrics when these tests showed significant differences ($p < 0.05$) between the two site groups. Then separation power and coefficient of variation (CV) were used to select the final metrics from the set of potential metrics. Separation power was defined as the degree of overlap between boxes (i.e., 25th and 75th percentiles) in box plots of the metric values for reference and impaired sites (Wang et al., 2005). We assigned a separation power of 3 when boxes did not overlap between the 2 site groups, a value of 2 when interquartile ranges overlapped but did not reach medians, a value of 1 when only one median was within the interquartile range of the other box, and a value of 0 when both medians were within the range of the other box. Final metrics were selected based on separation power $\geq$2 and CV < 1. When metrics were highly correlated ($r_s > 0.80$), we chose one based on a combination of highest separation power and lowest CV. At last, 0–10 scaling system was used to normalize the ranges of the final metrics as recommended by Hill et al. (2000) because of its fine scale. For metrics that decreased at impaired sites, the metric value for each site was divided by the 90th percentile of reference site values, and multiplied by 10. For metrics that increased at impaired sites, the metric value was divided by the 90th percentile of impaired site values, subtracted this value from 1, and multiplied the result by 10. The total D-IBI values were the sums of metric scores based on 0–10 scaling system.

2.5. Independent testing of metrics and D-IBI

The D-IBI and its metrics were tested using a different testing-site data set (29 sites) including 9 reference and 20 impaired sites (Fig. 1B). The calculation and scaling system of the final metrics for D-IBI were in the same manner as used for the training sites. We also employed separation power to examine the differences between reference and impaired sites. Three other methods were used to evaluate the D-IBI in the testing sites. First, a criterion was used to determine if D-IBI scores developed from training sites could distinguish reference from impaired conditions at testing-site data set (namely, % sites correctly classified). Its critical value was the average of the 75th percentile of IBI scores at impaired sites and the 25th percentile of IBI scores at reference sites from the training-site data set, and has been shown to be a better criterion than the other criterion (the 25th percentile of IBI scores at reference sites from the training-site data set) (Wang et al., 2005). Second, we used box-plot separation ratio to test the D-IBI, in which we divided the distance between boxes (defined by the difference between the 25th percentile of reference sites and the 75th percentile of all sites in the data set) by the median IBI score of reference sites. Third, we used 'correlation index' (CoI) and Cumulative_R^2 according to Blanco et al. (2007) and Wu et al. (2012):

$$\text{Cumulative\_}R^2 = S(r_s^2) \tag{1}$$

where R^2 = sum of r_s^2 with r_s = Spearman's correlation coefficient between D-IBI and a given environmental variable.

$$\text{CoI} = \frac{\text{Cumulative\_}R^2 S}{n^2} \tag{2}$$

where CoI is the correlation index for D-IBI, S is the number of r_s statistically significant at $p < 0.05$, and n is the number of environmental variables evaluated. CoI ranges from 0 to 1, while Cumulative_R^2 from 0 to n, indicating the theoretical minimum and maximum relationship between D-IBI and environmental variables, with high values indicating better relationship. The developed D-IBI was determined acceptable if there was no difference between values of above criteria at testing-site and training-site data set (examined by paired t-test). In our study, nonparametric Kruskal–Wallis tests, paired t-test, Spearman rank

Table 1
The summary statistics of environmental variables for reference and impaired sites in the training-site data set and results of non-parametric test between the reference and impaired sites.

Variables	Reference ($n=11$)			Impaired ($n=31$)			Kruskal–Wallis test	p-Values
	Percentile 25th	50th	75th	Percentile 25th	50th	75th		
PO_4-P (mg/L)	0.0037	0.0060	0.0113	0.0031	0.0064	0.0113	0.223	0.637
T-P (mg/L)	0.015	0.023	0.028	0.014	0.020	0.028	0.129	0.720
pH	7.39	7.73	7.91	7.36	7.58	7.84	0.344	0.557
COND (mS/cm)	190	233	294	203	235	284	0.118	0.731
TURB (NTU)	30.0	43.7	47.1	23.9	40.5	44.4	1.003	0.317
DO (mg/L)	9.54	10.12	10.62	9.45	9.87	10.67	0.361	0.548
WT (°C)	11.65	12.63	13.96	12.41	13.21	14.29	2.342	0.126
SAL	0.01	0.01	0.01	0.01	0.01	0.01	0.110	0.740
TDS (g/L)	0.12	0.15	0.19	0.13	0.15	0.18	0.074	0.786
ORP	−200	−190	−180	−213	−197	−175	0.090	0.764
Cl^- (mg/L)	7.25	8.82	13.60	6.73	8.04	11.80	0.361	0.548
Ca^{2+} (mg/L)	2.79	2.86	3.04	2.81	2.95	3.03	0.128	0.720
Velocity (m/s)	0.44	0.61	1.08	0.00	0.17	0.57	9.458	**0.002**
Width (m)	8.4	15.7	24.0	8.7	16.3	21.4	0.000	1.000
Depth (m)	0.30	0.37	0.51	0.40	0.58	0.78	6.055	**0.014**

Note: Values are in bold for significant difference between groups.
DO, dissolved oxygen; COND, conductivity; TDS, total dissolved solid; WT, water temperature; ORP, oxidation reduction potential; TURB, turbidity.

correlation tests and Kolmogorov–Smirnov tests were conducted by SPSS 11.5.

2.6. Application of the D-IBI

After testing the developed D-IBI, we applied it, in turn, for our study area to evaluate the ecological conditions of the sampling sites based on the total D-IBI scores. Four narrative classes were established according to the total D-IBI scores. Sites receiving a score in the 90th percentile of the sites were categorized as Excellent, 50th to 90th percentile as Good, 25th to 50th percentile as Moderate, and below the 25th percentile as Poor. The similar percentiles have been used in other studies as benchmarks for ecological assessment (Wang et al., 2005; Zalack et al., 2010).

3. Results

Sites varied widely in environmental variables. For example, pH ranged from 6.41 to 8.90 (mean: 7.87), COND ranged from 64.0 to 398.0 mS/cm (mean: 270.1 mS/cm), T-P ranged from 0.01 to 0.08 mg/L (mean: 0.03 mg/L), WT ranged from 10.58 to 19.77 °C (mean: 14.30 °C), while TURB ranged from 6.10 to 59.90 NTU (mean: 38.24 NTU). Stream depth ranged from 10 to 116 cm (mean: 48 cm), stream width ranged from 1.8 to 45.0 m (mean: 16.5 m), and velocity ranged from 0 to 1.80 m/s (mean: 0.43 m/s). Reference sites showed higher velocity, but lower water depth than those of impaired sites (Table 1). In all samples, the largest number of species belonged to Bacillariophyta (~80% of the total taxa), as was similarly the case in other studies of benthic algae in the Xiangxi River (Tang et al., 2004, 2006; Wu et al., 2009). We therefore focused on diatoms in subsequent analyses.

3.1. Development of the D-IBI

Totally 110 metrics belonging to 7 categories were calculated originally (Appendix A). Based on the selection methods, final four attributes – density of *Gomphonema parvulum* (Density-GoPa), chlorophyll *a* content (Chl-*a*), total diatom density (TDD) and diatom species richness (DSpR) – were selected. These metrics represented different aspects of benthic diatom communities, despite being significantly correlated (Table 2). All the metrics were greater at impaired sites and were normalized based on 0–10 scaling system (Table 3). Box plots of the total D-IBI scores showed good separation between reference and impaired sites (Fig. 2).

Table 2
Spearman rank correlations among four component metrics of the benthic diatom communities in training-site data set.

	Density-GoPa	Chl-*a*	TDD
Chl-*a*	0.479**		
TDD	0.560**	0.759**	
DSpR	0.567**	0.573**	0.763**

Chl-*a*, chlorophyll *a* contents; Density-GoPa, density of *Gomphonema parvulum*; TDD, total diatom density; DSpR, diatom species richness.
** $p < 0.01$.

Table 3
Four component diatom metrics of the diatom-based index of biotic integrity (D-IBI) and minimum and 90th percentile values of each metric used to calculate scores in the 0–10 scaling system. Values were obtained from the training-site data set.

	Minimum	90th percentile
Density-GoPa (ind/m^2)	1.62×10^7	7.02×10^8
Chl-*a* (mg/m^2)	2.92	39.91
TDD (ind/m^2)	6.16×10^8	1.62×10^{10}
DSpR	9	30

Note: Metrics abbreviations as in Table 2. Formula used for 0–10 scaling system was $(1 - X/\text{90th percentile of impaired sites}) \times 10$, X = value of the metric.

3.2. Testing the D-IBI

The D-IBI and its metrics were tested using 29 sites sampled within the Gaolan tributary. The application of the D-IBI, calculated using the same criteria and scaling systems as the training-site data set, indicated significant differences between reference and impaired sites (Kruskal–Wallis test, $p < 0.05$), and except for Chl-*a*, the separation powers were ≥2 (Figs. 3 and 4). The four metrics and D-IBI were highly correlated with environmental variables (Table 4). The critical value of % sites correctly classified was 27.8 under 0–10 scaling system. Twenty out of 29 (69%) sites were correctly classified, and only one reference site was classified as impaired site.

The box-plot separation ratio of testing-site was 0.07, which was lower than that of the training-site data set (0.16). Cumulative R^2 increased from training-site (0.65) to testing-site (1.00), while CoI remained constant at both training and testing-site (Table 5). Paired

Table 4
Spearman rank correlation coefficients (r_s) among metrics or diatom-based index of biotic integrity (D-IBI) and environmental variables using the testing data set.

Variables	Metrics or D-IBI				
	Density-GoPa (ind./m^2)	Chl-*a* (mg/m^2)	TDD (ind./m^2)	DSpR	D-IBI
PO_4-P (mg/L)	0.06	0.30	0.11	−0.29	0.04
T-P (mg/L)	0.11	0.32	0.05	−0.26	0.03
pH	0.12	0.29	0.12	−0.20	−0.02
COND (mS/cm)	0.12	0.22	−0.01	−0.18	0.04
TURB (NTU)	−0.17	−0.33*	−0.31	0.07	0.18
DO (mg/L)	0.04	0.17	0.07	−0.12	0.01
WT (°C)	0.18	0.21	0.14	0	−0.14
SAL	0.12	0.10	−0.08	−0.09	0.04
TDS (g/L)	0.12	0.23	−0.01	−0.17	0.04
ORP	0.03	0.01	−0.04	0.06	−0.03
Cl^- (mg/L)	0.15	0.10	0.29	0.21	−0.24
Ca^{2+} (mg/L)	0.39*	0.55**	0.42**	−0.05	−0.30
Velocity (m/s)	−0.33*	−0.31	−0.27	−0.31	0.36*
Width (m)	0.17	0.31	0.28	0.11	−0.24
Depth (m)	0.41*	0.36*	0.37*	0.57**	−0.54**

Note: Metrics abbreviations as in Table 2.
* $p < 0.05$.
** $p < 0.01$.

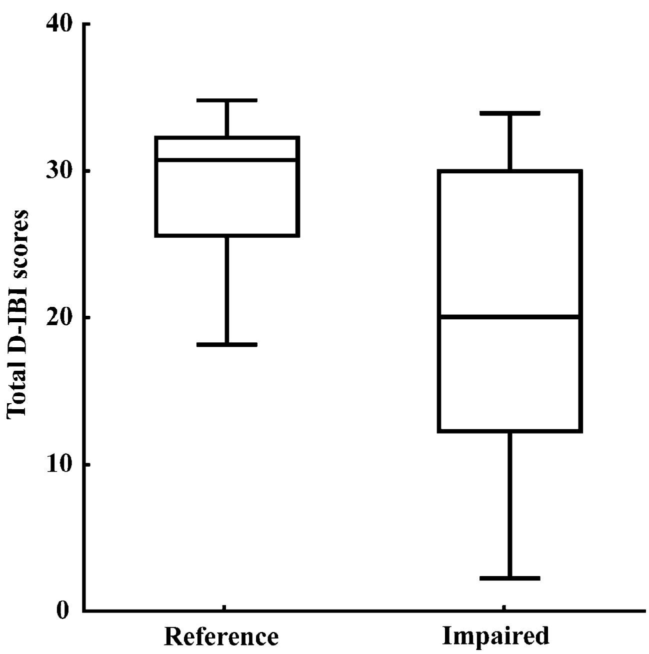

Fig. 2. Box plots of total D-IBI scores of reference (Re) and impaired sites (Im1–Im3) in the training-site data set under 0–10 scaling system. Boxes show interquartile ranges (25th and 75th percentiles), middle lines are medians, and whiskers are non-outlier ranges beyond the boxes.

t-test showed no difference between values of training-site and those of testing-site data set, in terms of separation powers, % sites correctly classified, box-separation ratios, CoI and Cumulative_R^2 (Table 5), indicating that the developed D-IBI was reliable.

Table 5
Separation powers, % sites correctly classified, box-separation ratios, CoI and Cumulative_R^2 of the diatom index of biotic integrity (D-IBI) based on the 0–10 scaling system from both training-site and testing-site data sets.

	Training-site	Testing-site
Separation powers	2	3
% sites correctly classified	0.67	0.69
Box-separation ratios	0.16	0.07
CoI	0.01	0.01
Cumulative_R^2	0.65	1.00
Paired *t*-test	$df = 4$, $t = -1.277$, $p = 0.271$	

3.3. Assessing dam impacts using the D-IBI

The total D-IBI scores for the whole Xiangxi catchment showed a wide range of values from 2.30 to 36.80 (40 max), and varied in the four different sampling sites (Fig. 5). The total D-IBI scores were classified as excellent, good, moderate, or poor based on being >33.9 (90th percentile) > 28.4 (50th percentile) > 18.9 (25th percentile) > 0, respectively. Box plots showed that median total D-IBI scores of reference sites (Re) were 31.7 (Fig. 5), which was the highest values among the four sites (Re, Im1–Im3). The lowest median values were observed at Im1 and Im3, which were 18.9 and 22.3, respectively. Median D-IBI score of Im2 was 28.4, higher than those of Im1 and Im3, but still lower than that of Re. Overall, the ecological condition of reference sites (Re) was "Good", while sites of Im1–Im3 were "Moderate" (Fig. 5). Due to the recovery of discharge, the D-IBI scores of Im2 were higher than those of Im1 and Im3, but still dramatically lower than those of Re (Fig. 5).

4. Discussion

After developing the D-IBI using training-site data set, it was shown to effectively discern reference sites from impaired sites in the testing-site data set. D-IBI scores were a good indicator of impairment caused by cascade run-of-river dams. Moreover, our D-IBI was powerful in terms of separation powers, % sites correctly classified, box-separation ratios, CoI and Cumulative_R^2 (Table 5).

4.1. Development and testing of the D-IBI

Managing stream and river ecosystems calls for an assessment of the ecosystem and a diagnosis of causes of degradation (Stevenson et al., 2010). Multi-metric indices of biotic integrity (IBI) based on diatoms have been constructed in several regions, depending upon their different impairment types (e.g. Hill et al., 2000, 2003; Kentucky Department for Environmental Protection Division, 2002; Wang et al., 2005; Tang et al., 2006; Zalack et al., 2010). However, all previously selected metrics were different among impairment types and study regions (Table 6), because an IBI should be built associated with special study goals (Stevenson et al., 2010). Even in the same area, Tang et al. (2006) and the present study for example, the metrics were also different for distinct anthropogenic stressors. Moreover, sites in different regions have not always been correctly identified as impacted or reference sites by certain indices. For instances, Hill et al. (2000) introduced

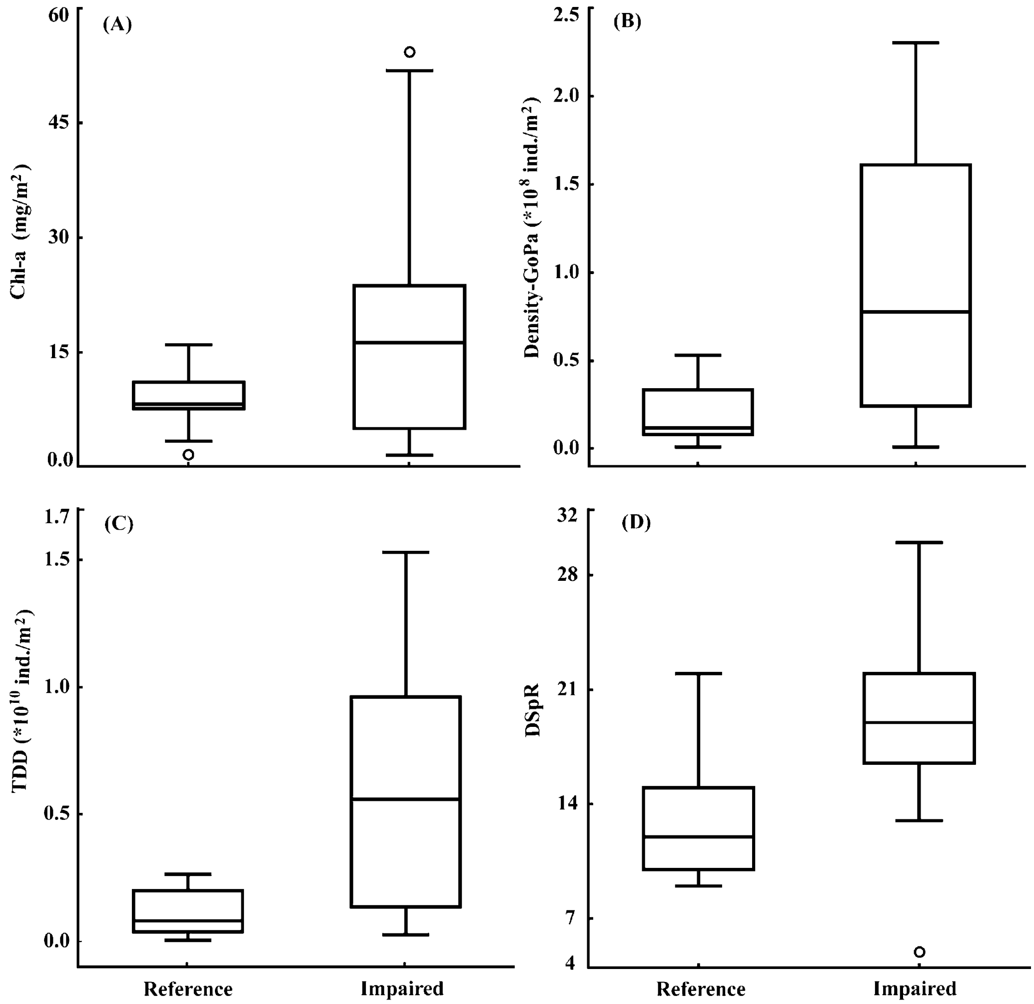

Fig. 3. Box plots of the four metrics of benthic diatom for reference and impaired sites in the testing-site data set. Boxes show interquartile ranges (25th and 75th percentiles), middle lines are medians, whiskers are non-outlier ranges beyond the boxes, and the dots are outliers. (A) chlorophyll *a* contents (Chl-*a*); (B) density of *Gomphonema parvulum* (Density-GoPa); (C) total diatom density (TDD); and (D) diatom species richness (DSpR).

the periphyton index of biotic integrity (P-BI), which was comprised of 10 metrics chosen to indicate various stressors, has not responded as predicted in all cases (Hill et al., 2000; Hamsher et al., 2004). This was not surprising since diatom communities showed great variability among different regions, which may result in the need to either regionally calibrate metrics or create and test different metrics that indicate stressors that may vary among regions. That was why the final four diatom metrics selected in the

Table 6
The comparisons of impairment type, study region and metrics selected among the present survey and previous studies.

	Impairment type	Study region	Metrics selected
Hill et al. (2000)	Water pollution	Mid-Appalachian streams, USA	Algal genera richness; the relative abundances of diatoms, Cyanobacteria, dominant diatom genus, acidophilic diatoms, eutraphentic diatoms, and motile diatoms; chlorophyll and biomass (ash-free dry mass) standing crops; alkaline phosphatase activity
Kentucky Department for Environmental Protection Division (2002)	Water pollution	Kentucky, USA	Total number of diatom taxa (TNDT); Shannon diversity (H′); pollution tolerance index (PTI); *Cymbella* group richness (CGR)
Hill et al. (2003)	Water pollution	Eastern United States	Species richness; species dominance; acidobiontic diatoms; eutraphentic diatoms; mobile diatoms; Chlorophyll; biomass and phosphatase
Wang et al. (2005)	Eutrophication	Interior Plateau Ecoregion, USA	KYDPTI; KY % no. sensitive species; No. of distinct reference species; average similarity to reference sites; % *Achnanthes*/(*Achnanthes* + *Navicula*); % no. of *Cymbella* species; % no. of *Navicula* species
Tang et al. (2006)	Water pollution	Xiangxi River, China	Acidobiontic algae (ACID); freshwater algae (FRESH); high oxygen requirement (HIGH-O); eutraphentic state (EUTRA); mobile taxa (MOBILE)
Zalack et al. (2010)	Acid mine drainage	Western Alleghany Plateau, USA	Species richness; % sensitive species; % tolerant species; no. of references species; % acidophilic species; % eutraphentic species; chlorophyll *a*; % similarity to reference sites; % *Cymbella* species
Present study	Flow regulation	Xiangxi River, China	The density of *Gomphonema parvulum* (Density-GoPa); chlorophyll *a* content (Chl-*a*); total diatom density (TDD); diatom species richness (DSpR)

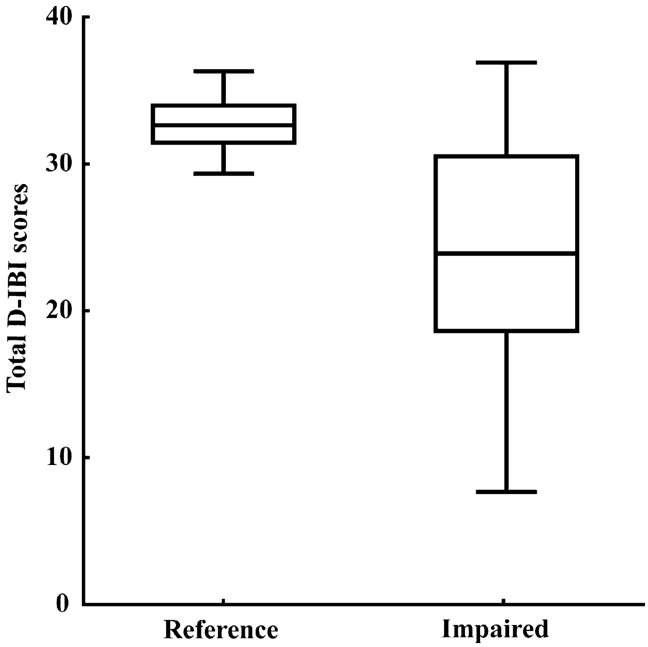

Fig. 4. Box plots of total D-IBI scores of reference (Re) and impaired sites (Im1–Im3) in the testing-site data set under 0–10 scaling system. Boxes show interquartile ranges (25th and 75th percentiles), middle lines are medians, and whiskers are non-outlier ranges beyond the boxes.

present study differed also from the previously reported research. Anyway, the regionally developed IBI should supply a quick assessment of the overall condition of a stream, and the individual metrics should provide insight into the causes of impairment (Hill et al., 2003).

McNair and Chow-Fraser (2003) concluded that chlorophyll *a* (Chl-*a*) was a good indicator of human-induced water-quality degradation and should be routinely monitored as part of an effective management program. However, Chl-*a* levels may change due to various pollutants, physical conditions and interactions of stressors (Zalack et al., 2010). Sites with above or below the median Chl-*a* content for a target stream received lowered scores (Hill et al., 2000), because excessive Chl-*a* can result in low dissolved oxygen problems leading to fish kills, loss of habitat for higher trophic levels and nuisance growth, while low Chl-*a* concentration may show the toxic effects of pollution (such as acid mine drainage) on diatom communities (Niyogi et al., 2002; Jia et al., 2009; Smucker and Vis, 2011). Conversely, moderate Chl-*a* levels serve as an important source of food and shelter for other stream organisms (Lamberti, 1996) and its utility in the D-IBI was an important measure of stream health. Diatom species richness (DSpR) was commonly used as metric in diatom-based environmental assessments (Bahls, 1993; Hill et al., 2001), in spite of the variable relationships with impairment (Stevenson and Pan, 1999; Smucker and Vis, 2009). The present study found that DSpR was greater in impaired than in reference sites and can also provide important information of stream health in a small area where a single source of impairment dominated (Zalack et al., 2010). Total diatom density (TDD), and Density-GoPa were employed because diatoms, which were the predominant group within periphyton (Tang et al., 2004; Wu et al., 2009), were a good indicator for both water quality and stressors (Reavie et al., 2010), while *Gomphonema parvulum* (GoPa), dominant species of the Xiangxi River (Wu et al., 2010b) and an indicator for higher nutrients (van Dam et al., 1994; Potapova and Charles, 2007), had a good discerning power.

In view of the habitat differences among groups, these four metrics were suitable for evaluating ecological conditions caused by run-of-river dams. Our conclusion was strongly supported by previous studies of Tang et al. (2006) and Li et al. (2010), who suggested that IBIs may be a suitable method for assessing river conditions within the Xiangxi River watershed. However, as an evaluation it is worth mentioning the weaknesses of the D-IBI and we must point out that (1) high correlations among the four metrics (Table 2) were found, which was caused probably by the dominance of a single stressor of human disturbances in the study region. Similar high correlations were found by Wang et al. (2005) and Wu et al. (2012); (2) the sensitivity of the D-IBI and its metrics can be further enhanced by expanding to a larger catchment or other regions impaired by run-of-river dams, but need further testing and assessment of their applicability; (3) future research should be addressed on other aquatic organisms and develop a more comprehensive IBI based on all potential metrics (e.g. fish, macroinvertebrate and plankton) for assessing flow regulations.

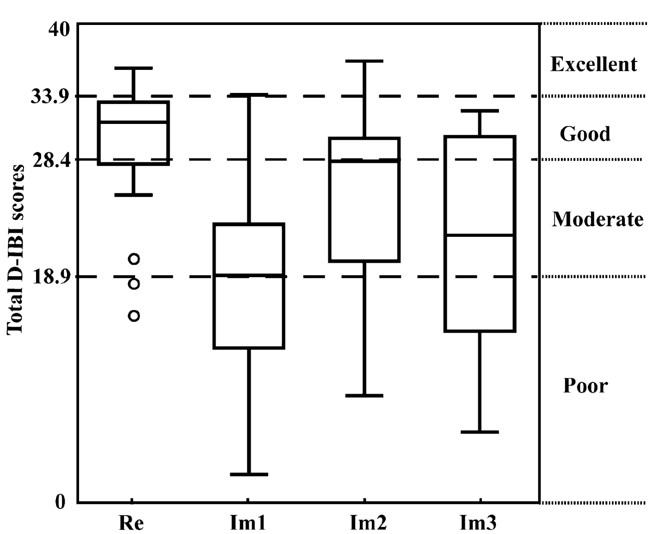

Fig. 5. Box plots of total D-IBI scores under 0–10 scaling system of four different habitats (Re, Im1, Im2 and Im3) in the whole Xiangxi catchment. Boxes show interquartile ranges (25th and 75th percentiles), middle lines are medians, whiskers are non-outlier ranges beyond the boxes, and the dots are outliers. Dashed lines indicate boundaries of different impairment categories marked in the right.

4.2. Total D-IBI scores of the four habitats

The establishment of the cascade run-of-river dams along the river made the habitats of lower reach (Im1–Im3) to have a significant different physical environment (Wu et al., 2010b), which was one of most significant factors influencing the structure, composition and diversity of stream faunal communities (Lammert and Allan, 1999; Downes et al., 2000; Wu et al., 2009, 2010a). Many stream organisms ranging from algae and aquatic plants to invertebrates and fish have close associations with the physical habitat, which is mainly determined by flow in streams (Bunn and Arthington, 2002). For instance, Pringle (1990) found important relationships between habitat type and nutrient supplies (e.g. more nutrients accumulating in pools) and stream algae. Also, Francoeur and Biggs (2006) demonstrated that increased velocity alone could remove benthic algal biomass and high suspended sediment concentrations further increased algal removal. In the Xiangxi River, because of the construction of run-of-river dams, the habitat (especially flow regime) of impaired sites (Im1, Im2 and Im3) changed significantly from reference sites (Re) within a very small distance, and was relatively stable during the sampling period, which was conducive to algal growth (Tang et al., 2004). Moreover, the decreasing biomass and density of benthic macroinvertebrates at impaired sites (Fu et al., 2008a), resulting in a lower grazing pressure, may be another reason increasing the four diatom metrics, which could potentially affect the D-IBI scores of Im1, Im2 and Im3. Those could be the possible mechanistic links between run-of-river dam construction and the final D-IBI scores of the four habitats (Re, Im1, Im2 and Im3).

4.3. Implications for watershed management

With the development of economy, there was always a conflict between the human demand and the ecological water requirement of river ecosystems. Our results indicated that the ecological condition of impacted sites was significantly declined from "Good" to "Moderate", which should be carefully considered by resource managers. Considering the huge numbers of run-of-river dams (>47) within the catchment, the overall influences of such dams on aquatic ecosystems were very severe. Although our survey provided with specific information and management targets, the minimal water requirement for the diatom community in the Xiangxi River was not obtained. Using weighted usable width method, Fu et al. (2008b) suggested the minimum ecological water requirement for the main group of the macroinvertebrates was 3.8 m^3/s in the Xiangxi River. At the same time, Li et al. (2009) provided an optimum instream environmental flow (2.639 m^3/s) by using weighted usable area method. Nevertheless, above values were theoretic and no evidence illustrated that such minimum discharges were really effective to improve the ecological condition of impacted sites. We thus proposed an 'Assessment–Management–Validation' method, which should put the minimal (or optimum) ecological water requirement into management practice and then use D-IBI to validate its effectiveness. We also expect that the D-IBI approach will ultimately improve communication to managers and legislators of the importance of protecting freshwater ecosystem functions.

5. Conclusion

Overall, the developed D-IBI effectively signaled impairment of flow regulation in the Xiangxi River of China, and it could be used in future studies measuring the long-term status of streams and the effectiveness of various remediation measures. However, further testing and assessment of their applicability are still needed, although the metrics selected would likely be useful in other regions impacted by small run-of-river dams. It is strongly suggested to continue this research on other aquatic organisms and explore the negative impacts of cascade run-of-river dams, which would help to develop a more comprehensive IBI based on all potential metrics (e.g. fish, macroinvertebrate and plankton). Furthermore, if ecologically acceptable flow had been ensured, such radical changes, particularly downstream of the dam, should not have occurred. Compared with reference sites, it can be concluded that additional abstractions from a watercourse give rise to adverse effects on aquatic ecosystem (as indicated by the D-IBI scores, Fig. 5). Therefore, we suggest that a minimal discharge should be preserved during the dry period and a basal flow kept for the maintenance of fluvial connectivity of the river water.

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Appendix A. Benthic diatom community metrics, taxonomic levels, their descriptions and references.

Metrics	Code	Taxonomic level	Descriptions and references
Biotic index			
Trophic diatom index	TDI	Species	Designed to detect eutrophication (Kelly and Whitton, 1995)
Pollution-tolerance index	PTI	Species	Weighted average of average abundance and tolerance value
pH index	PHI	Species	
pH-Acidobiontic	ACID	Species	
pH-Alkaliphilic	ALK	Species	
Salinity index	SalInd	Species	
Salinity-Fresh	FRESH	Species	
Salinity-Brackish	BRACK	Species	
Nitrogen uptake metabolism	NiUpMe	Species	
Nitrogen-autotrophic	AUTO-N	Species	Weighted average of average abundance and tolerance value (van Dam et al., 1994; Tang et al., 2006)
Nitrogen-heterotrophic	HETER-N	Species	
Oxygen requirements	OxyReq	Species	
High oxygen requirement	HIGH-O	Species	
Low oxygen requirement	LOW-O	Species	
Saprobity index	SapInd	Species	
Oligosaprobous	OLIG	Species	
Polysaprobous	SAPR	Species	
Diversity index			
Shannon diversity	H'	Species	Measure of evenness and richness
Margalef's diversity	M-diversity	Species	
Simpson's Dominance	SimDom1	Species	
Simpson's Diversity (1/Dominance)	SimDom2	Species	
Simpson's Diversity (1-Dominance)	SimDom3	Species	
Gleason diversity	GleDiv	Species	Spatharis and Tsirtsisa (2010)
Hill's N1	HillN1	Species	
Menhinick diversity	MenDiv	Species	Menhinick (1964)
Diatom species richness	DSpR	Species	Number of species in the count
No. of genera	NoGen	Genus	Number of genera in the count

Metrics	Code	Taxonomic level	Descriptions and references
Evenness index			
Evenness 1	E1	Species	Measure of species composition evenness (Spatharis and Tsirtsisa, 2010)
Evenness 2	E2	Species	
Evenness 3	E3	Species	
Evenness 4	E4	Species	
Evenness 5	E5	Species	
Dominance index			
Berger–Parker	B	Species	Berger and Parker (1970)
McNaughton	α	Species	McNaughton (1967)
Growth form			Wang et al. (2005)
% prostrate individuals	%ProInd	Genus	Relative abundance of prostrate genera
% erect individuals	%EreInd	Genus	Relative abundance of erect genera
% stalked individuals	%StaInd	Genus	Relative abundance of stalked genera
% unattached individuals	%UnaInd	Genus	Relative abundance of unattached genera
% mobile individuals	%MobInd	Genus	Relative abundance of motile genera
Similarity			
% reference species	%RefSpe	Species	% of species found in reference sites that occurred in impaired sites
Percent similarity to reference sites	%SimRef	Species	Bray–Curtis similarity to reference sites
No. of distinct reference species	NDiReSp	Species	No. of species found primarily in reference sites not in impaired sites
Taxonomic composition			
Chlorophyll *a*	Chl-*a*	–	The concentration of chlorophyll *a*
Total diatom density	TDD	Species	Total density of benthic diatoms
Dominant species	–	Species	The density of dominant species
% dominant species	–	Species	Relative abundance of dominant species
% NNS	–	Genus	The sum of the relative abundance of *Navicula*, *Nitzschia* and *Surirella* taxa
% *Ach*/(*Ach* + *Nav*)	–	Genus	Ratio of *Achnanthes* to *Achnanthes* and *Navicula* combined
% *Cym*/(*Cym* + *Nav*)	–	Genus	Ratio of *Cymbella* to *Cymbella* and *Navicula* combined
% (*genus*) individuals	–	Genus	Relative abundance of (*genus*)
No. of (*genus*) species	–	Genus	No. of (*genus*) species
% no. of (*genus*) species	–	Genus	% no. of (*genus*) species

Note: PTI = Kentucky Diatom Pollution Tolerance Index.

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