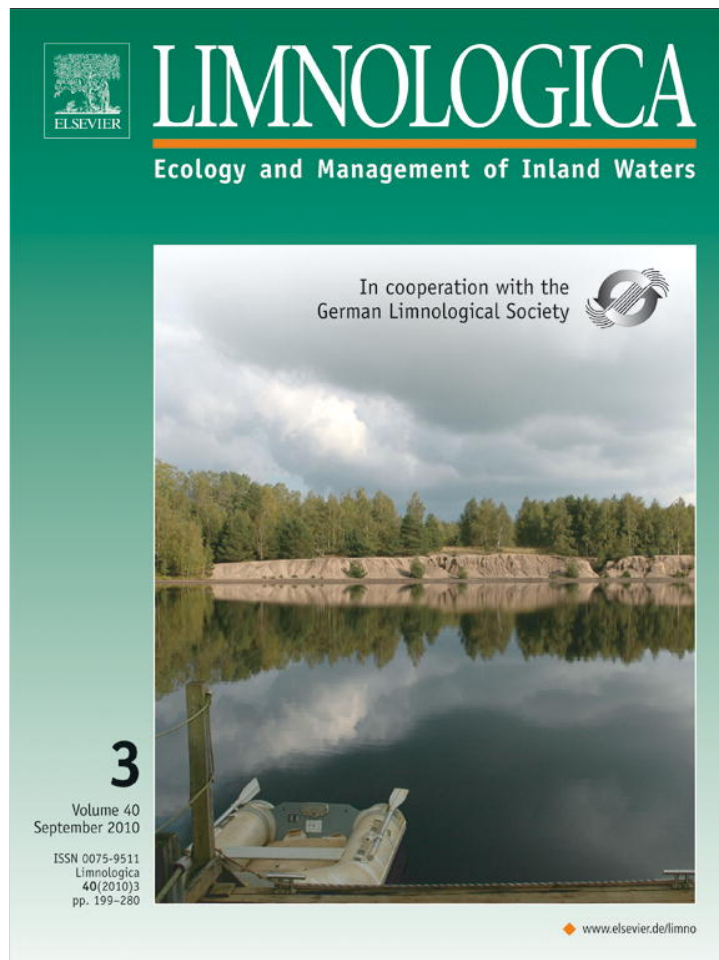


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Biological assessment of the Zayandeh Rud River, Iran, using benthic macroinvertebrates

Mohammad Nemati Varnosfaderany*, Esa Ebrahimi, Noorolla Mirghaffary, Alireza Safyanian

Department of Natural Resources, Isfahan University of Technology, Isfahan 84156-83111, Iran

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ABSTRACT

Benthic macroinvertebrate communities from the middle of Zayandeh Rud River were analyzed monthly during 1 year at 8 stations, in order to assess changes in their diversity and richness in relation to water quality. Two major groups of sites based on similarity between macroinvertebrate communities were identified by cluster analysis. The performances of the original and revised BMWP score systems were assessed by comparing the community structure indices of benthic macroinvertebrates along with physico-chemical parameters of the water. The biotic indices (BMWP, ASPT, revised BMWP and ASPT) showed better correlation with water quality parameters than that of the richness and diversity indices. The revised ASPT had the highest correlation with water quality parameters. It seems that the application of the revised BMWP score system could be useful for assessment of the water quality in Zayandeh Rud River.

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Introduction

The study of river benthic macroinvertebrates for biological monitoring techniques has been widely reported and described in the literature (Washington, 1984; Metcalfe, 1989; Rosenberg and Resh, 1993; Mandaville, 2002). Iliopoulou-Georgudaki et al. (2003) showed that the use of macroinvertebrates as bioindicators for the assessment of water quality has more advantages than those based on diatoms, fishes, riparian and aquatic vegetation. Benthic macroinvertebrates are often the taxa group of choice for biotic indices in river environments as they are found throughout the length of the river, have limited mobility and a relatively long lifespan. Most interestingly, freshwater macroinvertebrate species vary in their sensitivity to organic pollution (Rosenberg and Resh, 1993) and, as a result, their presence or absence can be used to make inferences about pollution loads. Biotic indices are numerical expressions combining a quantitative measure of species diversity with qualitative information on the ecological sensitivity of individual taxa (Czerniawska-Kusza, 2005). One of the most common biotic indices in use is the Biological Monitoring Working Party (BMWP) score system. This index allocates a single score to benthic macroinvertebrates at the family level that is representative of the family's tolerance to water pollution. The greater their tolerance to pollution, the lower the BMWP score

and vice versa (Armitage et al., 1983). This system reappraised at 1996 using the concept of average score per taxon (ASPT) and mathematical formulae based on 17,353 biological samples from England and Wales (Walley and Hawkes, 1996).

Generally, the use of indices requires prior modification according to environmental conditions or pollution types. The BMWP score system that developed for river pollution surveys in the UK (Armitage et al., 1983), have been successfully applied in other countries including Spain (Zamora-Muñoz et al., 1995), Italy (Solimini et al., 2000), Thailand (Mustow, 2002), Poland (Czerniawska-Kusza, 2005), Greece (Artemiadou and Lazaridou, 2005), Portugal (Faria et al., 2006), Brazil (Silveira et al., 2005), Malaysia (Azrina et al., 2006), Egypt (Fishar and Williams, 2008) and Hindu Kush-Himalaya region (Ofenböck et al., 2008).

Recent water-quality monitoring programs in Iran have been mainly based on the determination of physical and chemical parameters; in contrast, the biological assessment of rivers is very limited. While, the study of macroinvertebrates as an impact indicator can reveal occurrence of intermittent or unrecorded chemical pollution incidents (Rosenberg and Resh, 1993; Fishar and Williams, 2008).

The purpose of this study is to (1) present an overall view of the macroinvertebrate communities along the middle section of the Zayandeh Rud River, (2) determine the biological water quality based on benthic communities, (3) study the application of the BMWP and revised BMWP score systems in the river and (4) to compare the results of the BMWP and revised BMWP with four commonly used non-parametric community structure indices

* Corresponding author. Tel.: +98 311 3913565; fax: +98 311 3912840.
E-mail address: mnemati@na.iut.ac.ir (M.N. Varnosfaderany).

including taxa richness (TR), Margalef's index (R), Shannon–Wiener diversity index (H) and Simpson's diversity index (D).

Methods

Study area

The Zayandeh Rud basin is located in the central part of Iran, with geographical coordinates between $50^{\circ}24'–53^{\circ}24'$ East and $31^{\circ}11'–33^{\circ}42'$ North (Fig. 1). The area of the basin is about $42,000\text{ km}^2$, with an altitude ranging from 1466 to 3974 m and average annual rainfall of 130 mm (Salemi et al., 2000). The Zayandeh Rud River flow regime depends not only on climatic conditions but also is affected by hydroelectric power generation, as well as irrigation needs through the Chadegan Dam. Major land uses in the catchment basin include bare land (63.71%), outcrop (18.3%), agriculture (11%), range (6.17%) and urban development (0.82%), respectively (Iranian Ministry of Agriculture and Office of Statistics and Information, 1998). Eight stations were selected in the middle of the Zayandeh Rud River from Baghbahadoran to Zyar ($S_1–S_8$) along 132 km of river path (Fig. 1). The first station was located above Baghbahadoran city, where the water quality is acceptable for producing drinking water and about $12\text{ m}^3\text{ s}^{-1}$ of river flow is pumped to the Isfahan water treatment plant (Pourmoghadas, 2002). The visual common characteristics of the

substrates in the stations 1–6 were the presence of cobbles and pebbles, and sometimes sand and gravel; the two downstream stations consisted mainly of muddy type sediments. The S_7 is situated after the outfall of the urban wastewater treatment plant in the southern part of Isfahan (Fig. 1).

Water and benthic macroinvertebrates sampling

Water and benthic macroinvertebrate samples at each site were collected monthly from July 2006 to June 2007. At each site, water samples were collected from the top 30 cm of the water column at the middle of the river by means of an acid-washed plastic bucket, rinsed with water from the site. Samples were stored in the bottles (for chemical analysis) and sterile glass flasks (for bacteriological analysis), cooled, transported to the laboratory and processed within 12 h of collection. For each sample, water quality variables including pH, oxygen saturation, total dissolved solids (TDS), electric conductivity (EC), nitrate, phosphate, biological oxygen demand (BOD_5), and fecal coliforms were measured (APHA 1992).

A Surber sampler (catching area: 625 cm^2) was used for benthic invertebrate sampling in the six upstream stations that had shallow running water and substrates containing cobbles and pebbles. At the downstream stations (S_7 and S_8) with muddy and sandy substrates, a corer sampler (catching area: 81 cm^2) was

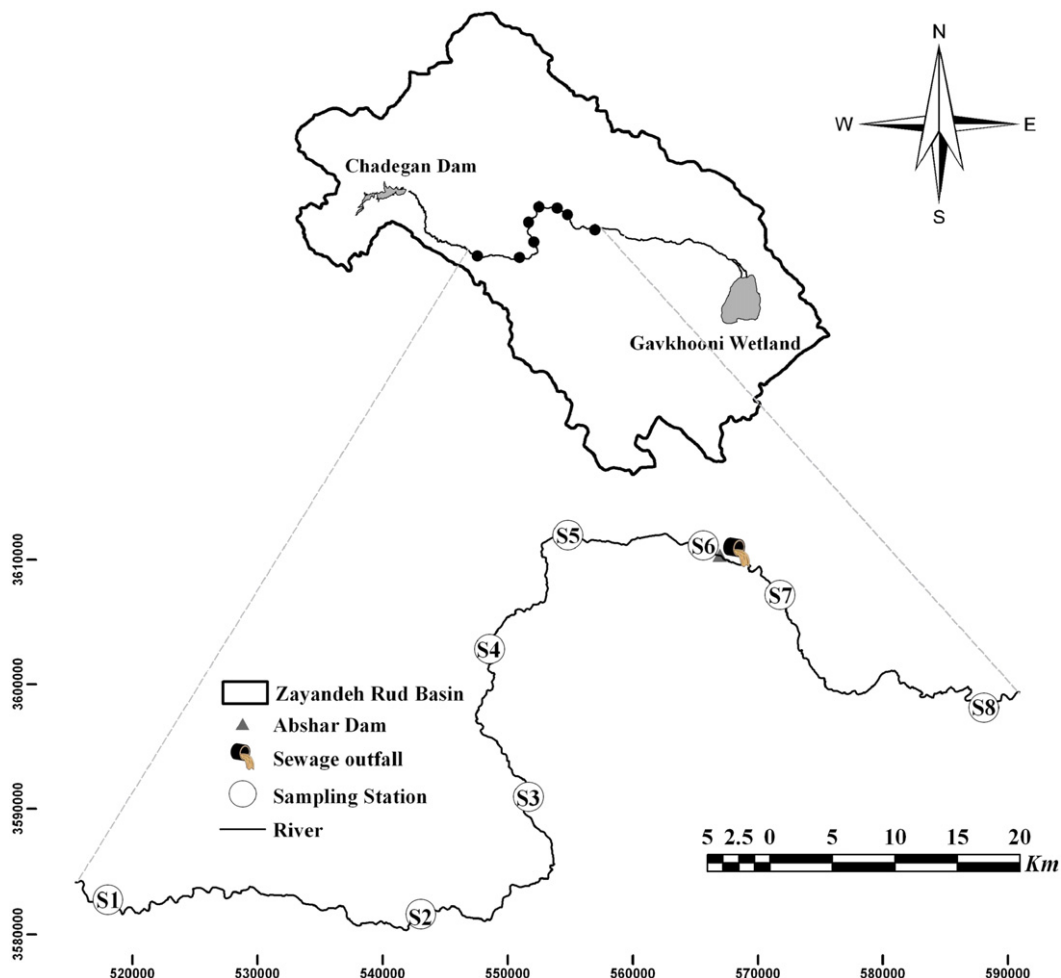


Fig. 1. Location of study area and sampling stations ($S_1–S_8$) (direction of river flow is west to east).

used for sampling. At each site, five samples were taken. Qualitative samples were also taken with a kick-sampling technique in order to capture taxa richness more completely. The content of each sample was washed in the field using a sieve with a mesh size of 0.4 mm. All captured organisms were placed in plastic bottles and preserved in 70% ethanol (Mandaville, 2002). The benthic macroinvertebrate identification was done to the lowest possible taxonomic level in the laboratory based on keys presented by Elliott et al. (1988), Hynes (1984), Milligan (1997), Pescador et al. (2004) and Timm (1999).

Data analysis

The degree of similarity between macroinvertebrate communities and the classification of sites was defined on the basis of Ward's method and a hierarchical cluster analysis (Bis et al., 2000).

The BMWP and revised BMWP in each site was monthly calculated by adding the individual scores of the families (Armitage et al., 1983; Walley and Hawkes, 1996).

The ASPT and revised ASPT were calculated by the ratio of BMWP and revised BMWP values to the number of families, respectively (Armitage et al., 1983; Walley and Hawkes, 1996).

The commonly used non-parametric community structure indices including taxa richness (TR), Margalef's index (R), Shannon–Wiener diversity index (H) and Simpson's diversity index (D) were calculated, based mostly on the genus (Washington, 1984).

The normality of data was tested using the Kolmogorov–Smirnov test. The t -test was used to compare the calculated values of BMWP and ASPT with the revised BMWP and ASPT. One-way analysis of variance (ANOVA) followed by Duncan multiple comparison tests were conducted to test the significant differences of biotic indices between sites. The correlations between biological indices and chemical variables were computed using the non-parametric Spearman's rank coefficient of correlation (Zar, 1999). All statistical analysis was performed using the SPSS software (version 10).

Results

Benthic macroinvertebrates

The examination of samples resulted in a total number of 42 families representing 5 classes and 16 orders of benthic macroinvertebrates (Table 1). The total number of identified families varied between 10 and 17 among the sites. The lowest number of families found during the monthly sampling was 4 at S_7 . The Plecoptera, Ephemeroptera and Trichoptera orders were absent at two downstream stations (S_7 and S_8).

Overall, the benthic macroinvertebrate communities of the Zayandeh Rud River were dominated by Chironomid larvae (30.51%), but the dominance structure differed among particular sites ($S_5=61.84\%$ to $S_7=0.1\%$). The second dominant taxa were Gammaridae (15.77%). They were absent at the two downstream stations (S_7 and S_8) and accounted for 63.88% of the samples taken

Table 1
Taxonomical list of benthic macroinvertebrates which were determined in Zayandeh Rud River and their BMWP^a and revised BMWP scores (Walley and Hawkes, 1996).

Taxon	BMWP score	Revised BMWP score	Taxon	BMWP score	Revised BMWP score
Plecoptera			Odonata		
Perlidae	10	12.5	Libellulidae	8	5
Perlodidae	10	10.7	Calopterygidae	8	6.4
Ephemeroptera			Platycnemididae	6	5.1
Heptagenia	10	9.8	Gomphidae	8	8 ^c
Potamanthidae	10	7.6	Hemiptera		
Baetidae	4	5.3	Corixidae	5	3.7
Leptophlebiidae	10	8.9	Amphipoda		
Caenidae	7	7.1	Gammaridae	6	4.5
Trichoptera			Bivalvia		
Polycentropodidae	7	8.6	Unionidae	6	5.2
Hydropsychidae	5	6.6	Sphaeriidae	3	3.6
Hydroptilidae	6	6.7	Gastropoda		
Rhyacophilidae	7	8.3	Lymnaeidae	3	3
Philopotamidae	8	10.6	Ancylidae	6	5.6
Psychomyiidae	8	6.9	Physidae	3	1.8
Diptera empididae	4 ^b	4 ^b	Hydrobiidae	3	3.9
Tabanidae	5 ^b	5 ^b	Valvatidae	3	2.8
Tipulidae	5	5.5	Hirudinea		
Simuliidae	5	5.8	Piscicolidae	4	5
Chironomidae	2	3.7	Glossiphoniidae	3	3.1
Coleoptera			Erpobdellidae	3	2.8
Elmidae	5	6.4	Oligochaeta	1	3.5
Dytiscidae	5	4.8	Tubificidae		
Hydrophilidae	5	5.1	Naididae		
			Lumbricidae		
			Lumbriculidae		
			Haplotaxidae		

^a Biological Monitoring working party.

^b Calculated scores according to reappraisal methodology.

^c Score was taken from BMWP original score.

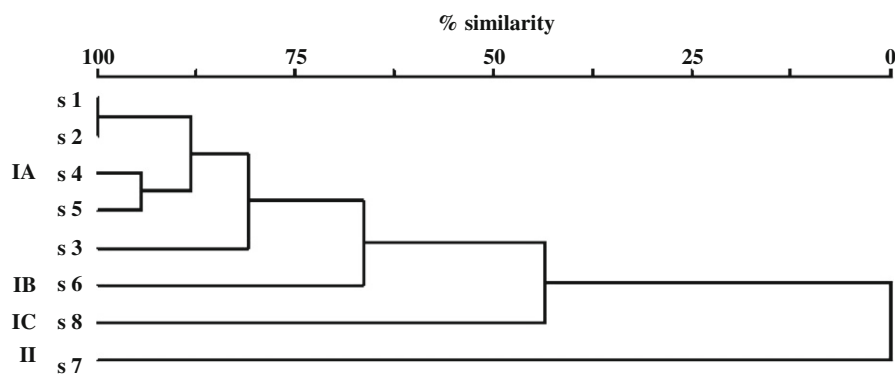


Fig. 2. The dendrogram of similarity of stations (S₁–S₈) in Zayandeh Rud River in respect of benthic macroinvertebrates communities. Two major groups of sites (I and II) presented. IA, IB and IC are the subgroups of I major group.

Table 2

Mean values (± standard deviation) of environmental and physico-chemical parameters, biotic indices and richness and diversity indices at the sampling sites of Zayandeh Rud River in the period of investigation.

Parameters/sites	S ₁	S ₂	S ₃	S ₄	S ₅	S ₆	S ₇	S ₈
Distance to Chadegan Dam (km)	106	137.58	158.51	174.15	188.45	200.38	208.83	236.77
Water temperature (°C)	11.4 ± 6.3	13.7 ± 7.3	14.1 ± 7.4	14.6 ± 8.3	13.9 ± 7.4	14.7 ± 8.2	15.1 ± 7.5	14.2 ± 8.9
Water flow (m ³ s ⁻¹)	60.6 ± 34	55.2 ± 33	45.2 ± 28	40.3 ± 24	30.7 ± 16	26.3 ± 13	17.4 ± 12	2.5 ± 5
pH	8 ± 0.3	8.1 ± 0.2	8.1 ± 0.2	8 ± 0.2	8 ± 0.2	8 ± 0.2	7.6 ± 0.3	7.6 ± 0.3
EC (μs cm ⁻¹)	217 ± 49	306 ± 137	314 ± 127	315 ± 135	326 ± 130	346 ± 145	363 ± 131	384 ± 130
TDS (mg l ⁻¹)	108 ± 24	153 ± 69	157 ± 63	157 ± 67	163 ± 65	173 ± 72	181 ± 66	191 ± 64
Oxygen saturation (%)	90.2 ± 9.1	93 ± 11.8	93.2 ± 15	92 ± 14.8	90.5 ± 12	81.4 ± 8.6	55.2 ± 9.2	38 ± 11.5
BOD ₅ (mg l ⁻¹)	15.3 ± 8.3	18.4 ± 7	17.4 ± 7.8	17.7 ± 6.7	18.8 ± 4.7	20.4 ± 4.8	21.6 ± 6.3	19.9 ± 6.8
NO ₃ ⁻ (mg l ⁻¹)	5.9 ± 3.1	9.3 ± 7.5	9.3 ± 7.8	9.4 ± 8.1	11 ± 9.8	10.8 ± 9.7	12 ± 10.3	16.8 ± 16
PO ₄ ⁻ (mg l ⁻¹)	0.3 ± 0.4	0.4 ± 0.6	0.4 ± 0.4	0.4 ± 0.7	0.3 ± 0.4	0.3 ± 0.4	1.1 ± 1.1	1.1 ± 0.9
Fecal coliform (MPN 100 ml ⁻¹)	599 ± 790	2764 ± 4877	1370 ± 3048	2342 ± 2966	2335 ± 1428	1154 ± 1038	30,750 ± 13,585	10,469 ± 6458
BMWP ^a	46.5 ± 9.9	44.9 ± 19.7	46.7 ± 15.5	44.4 ± 11.4	33.5 ± 14.9	37.8 ± 12.9	18.7 ± 7.2	20.8 ± 5.8
Revised BMWP	41.2 ± 11.8	37.0 ± 16.2	40.9 ± 15.6	37.9 ± 10.3	29.4 ± 16.4	36.4 ± 12.6	17.3 ± 7.2	16.0 ± 5.2
ASPT ^b	4.6 ± 0.8	4.2 ± 0.7	4.4 ± 0.8	4.7 ± 0.7	3.9 ± 0.9	4.0 ± 0.6	2.8 ± 0.3	3.0 ± 0.4
Revised ASPT	5.3 ± 0.6	4.9 ± 0.8	5.1 ± 0.7	5.3 ± 0.6	4.4 ± 0.6	4.2 ± 0.6	3.2 ± 0.3	3.6 ± 0.4
Shannon-wiener diversity index (H)	1.4 ± 0.2	1.6 ± 0.3	1.6 ± 0.3	1.4 ± 0.3	1.2 ± 0.5	1.1 ± 0.5	0.1 ± 0.1	1.5 ± 0.3
Simpson's diversity index (D)	0.7 ± 0.1	0.7 ± 0.1	0.7 ± 0.1	0.7 ± 0.1	0.5 ± 0.2	0.5 ± 0.2	0.03 ± 0.03	0.7 ± 0.1
Margalef's index (R)	1.7 ± 0.3	1.8 ± 0.5	1.7 ± 0.5	1.7 ± 0.4	1.7 ± 0.8	1.7 ± 0.3	0.7 ± 0.2	1.4 ± 0.3
Taxa richness (TR)	10.0 ± 1.7	10.2 ± 3.3	10.6 ± 2.8	9.8 ± 1.9	9.5 ± 3.8	11.2 ± 2.1	7.6 ± 2.0	7.8 ± 1.6

^a Biological monitoring working party.

^b Average score per taxon.

from S₆. Physidae had the third order of dominance (14.64%), but at the two downstream stations, the proportions were 98.46% and 15% at S₇ and S₈, respectively, whereas at six upstream stations, varied between 0% and 1.57%.

Seven families including Chironomidae, Erpobdellidae, Tubificidae, Naididae, Lumbricidae, Lumbriculidae and Elmidae were recorded at all of the sampling stations.

According to Wards similarity index, the most similarity was observed between the stations 1 and 2. The station 7 showed the most difference in the content of benthic macroinvertebrates, in terms of the numbers and taxa. The site classification based on the macroinvertebrate composition using cluster analysis is presented in Fig. 2. The dendrogram separates all sampling sites into two major groups. The first group consists of the six upstream stations and S₈ whereas the second group contains only S₇. The results of the cluster analysis allowed for further separation of three subgroups of sites in the first major group (IA: S₁–S₅, IB: S₆, IC: S₈).

Biological indices and physico-chemical parameters

A summary description of the calculated biotic indices, the community structure indices and the physico-chemical characteristics of the sampling sites are provided in Table 2. The different levels of water quality variables indicative of water pollution are significantly apparent from upstream to downstream stations.

The results of the *t*-test showed that BMWP values at all the stations were significantly lower ($t=20.385$, $df=95$, $P < 0.001$) than those obtained from revised BMWP values, the same for the ASPT and revised ASPT ($t=41.482$, $df=95$, $P < 0.001$).

As shown in Table 3, the calculated biotic indices (BMWP, ASPT, revised BMWP and ASPT) have positive correlation ($P < 0.001$) with percentage of oxygen saturation, water flow and pH; negative correlation ($P < 0.001$) with fecal coliform, EC and TDS. The biotic indices showed greater correlation with water quality parameters than that of the

Table 3
Spearman correlation coefficients between biological indices and water physico-chemical variables.

Water flow	Fecal coliforms	EC	TDS	pH	BOD ₅	PO ₄ -	NO ₃ -	Oxygen saturation	
0.43**	-0.49**	-0.32**	-0.31**	0.31**	-0.19	-0.03	-0.03	0.45**	BMWP
0.46**	-0.53**	-0.34**	-0.34**	0.33**	-0.22*	-0.04	-0.02	0.49**	Re-BMWP
0.52**	-0.54**	-0.37**	-0.37**	0.30**	-0.17	-0.17	-0.07	0.50**	ASPT
0.55**	-0.60**	-0.45**	-0.45**	0.36**	-0.18	-0.13	-0.08	0.54**	Re-ASPT
0.21**	-0.45**	-0.16	-0.16	0.20	-0.15	0.11	0.09	0.21**	H
0.38**	-0.46**	-0.18	-0.18	0.23*	-0.19	0.07	0.07	0.38**	R
0.20*	-0.46**	-0.15	-0.14	0.16	-0.12	0.06	0.06	0.20*	D
0.34**	-0.37**	-0.22*	-0.22*	0.25*	-0.11	0.05	-0.6	0.34**	TR

The variables include Biological Monitoring Working Party score (BMWP), revised BMWP score (Re-BMWP), average score per taxon (ASPT), revised ASPT (Re-ASPT), Shannon–Wiener diversity index (*H*), Simpson's diversity index (*D*), Margalef's index (*R*) and Taxa richness (TR).

* Correlation is significant at the 0.05 level.

** Correlation is significant at the 0.01 level.

richness and diversity indices. The revised ASPT had the highest correlation with water quality parameters. The values of revised ASPT showed a significant decrease from upstream to downstream in the river ($F=22.34$, $df=7$, $P < 0.001$). The Duncan test categorized the sites into three groups: S_1 – S_4 , S_5 & S_6 , and S_7 & S_8 .

According to the results of the revised ASPT at each station (Table 2), this part of the Zayandeh Rud River comprised from three water quality classes (Mandaville 2002). The scores of the four upstream stations (S_1 – S_4) were categorized as “good quality”; S_5 and S_6 “moderate pollution” and the 2 downstream stations (S_7 and S_8) “severe pollution”. Whereas, according to the Shannon–Wiener diversity index (Wilhm and Dorris, 1968), the four upstream stations and S_8 were categorized as “moderate”, S_5 and S_6 “moderate to substantial” pollution classes, while S_7 classified as the only “substantial” pollution class (Table 2). The other biological indices followed the same trend and indicated an overall increase of nutrient pollution, particularly along the downstream part of the river.

Discussion

Benthic macroinvertebrates

The numbers of Ephemeroptera taxa were highest at four upstream stations and decreased at S_5 and S_6 , while they were absent at the two downstream stations. This indicates the increase of water pollution from upstream to downstream as also mentioned by Merritt and Cummins (1978).

Chironomidae family was the main dominant taxa at the five up stream station. The Gamaridae family was the main dominant taxa followed by Chironomidae at S_6 . At S_8 , Chironomidae, Lumbricidae and Lumbriculidae families were the dominant taxa.

The seventh station was dominated by *Physa acuta* (Gastropoda) and *Oligochaeta*, which are known to be able to tolerate unfavorable conditions such as low dissolved oxygen and high pollutant concentrations (Zadory and Müller, 1981; Brinkhurst, 1967). The results of water quality parameters showed that the river was highly polluted at S_7 . The BOD₅ was 21.6 mg l⁻¹ and oxygen saturation was 55.2% at this station. Moreover, it had the highest amount of fecal coliform bacteria (Table 2). This station was located at 6 km downstream of the outfall of urban wastewater treatment plant in the southern part of Isfahan. The water was milky, smelly and contained a lot of suspended matter due to the sewage pollution. Water uptake from the river at the Abshar Dam (Fig. 1) for agricultural use lowers the flow volume before

the effluent discharge enters the river and causes the high pollution of the water.

Biotic indices

The biotic indices (BMWP and revised BMWP) were used at family level due to the lack of established taxonomic keys for Iranian macrobenthic invertebrates, especially to species level. The degree of tolerance to environmental conditions at the family level is related to the diversity of species as well as the tolerance range of individual species within the family. Therefore, the scores at the family level usually represent intermediate values of species tolerance (Armitage et al., 1983). In this regard, indices at the family level may under- or overestimate water quality more than those based on species level. However, the use of indices at the family level may be adequate in terms of cost-efficiency, because they are easy to calculate and require less taxonomic knowledge when taxonomic experts are not available (Rosenberg and Resh, 1993; Mustow, 2002; Czerniawska-Kusza, 2005).

In this study, the biotic system has been adapted as follows:

- I The Empididae and Tabanidae families which have not included in the original BMWP score list were present in the Zayandeh Rud River. Their scores were calculated based on reappraisal methodology developed by Walley and Hawkes (1996) and considering their scores in another modified version of BMWP (Alba-Tercedor and Sanchez-Ortega, 1988; Artemiadou and Lazaridou, 2005; Ofenböck et al., 2008).
- II For the Gomphidae which did not have revised scores (Walley and Hawkes 1996), the original scores was used (Armitage et al., 1983).

In the most of sampling sites, the values of revised BMWP and ASPT scores were generally higher than the BMWP and ASPT, although, the revised scores of BMWP for some families can be either greater or lower than original scores (Walley and Hawkes, 1996). This indicates that families present in the Zayandeh Rud River pertain mostly to the families that have greater revised scores than original scores.

Correlation of biological indices to water quality parameters

The correlation between water quality parameters and biotic indices are greater than that of community structure indices (Table 3). This reveals the more sensitivity of biotic indices to variation of water quality. Johnson et al. (2006) showed that the

stress-specific metrics (such as BMWP scoring system) are most robust than the community structure indices.

According to the greater correlation coefficient of revised BMWP and ASPT than original BMWP and ASPT with water quality parameters (Table 3), the revised system gives a better interpretation of biological quality in the Zayandeh Rud River. Walley and Hawkes (1996) indicated that revised BMWP score system is more reliable than original scores.

On the other hand, the revised ASPT had the higher correlation with water quality parameters than revised BMWP. This indicates the greater sensitivity of revised ASPT to the water quality variation, due to its less sensitivity to sampling techniques, seasonal changes and macroinvertebrate diversity (Armitage et al., 1983).

Water quality classification

When classifying the water quality according to biological assessment, one must be very cautious, because some classification systems are not universally applicable, such as the Wilhm and Dorris classification system that has been set for the Shannon diversity index of macroinvertebrates (Metcalf, 1989). Moreover, some of biological fluctuations exhibit the combination of natural and anthropogenic influences such as food availability, hydraulic conditions, substrate composition, nutrient loads and water quality variations (Rosenberg and Resh, 1993). The Shannon–Wiener diversity index for S_8 was higher than some of the upstream stations (S_1 , S_4 , S_5 and S_6), while water quality parameters reveal greater pollution at this station (Table 2). This implies the weakness of using diversity as a measure without taking into account the taxa tolerance. However, the biological classification of Zayandeh Rud River quality based on the revised ASPT showed a descendent trend along the river. Pourmoghadas (2002) reported that the Zayandeh Rud River was generally clean at the upstream part and was polluted due to urban waste waters and agricultural activities at the downstream part.

Conclusions

The presence of certain benthic macroinvertebrate taxa particularly in polluted and non-polluted parts of a river indicate that they could be used as potential bioindicators for river assessment. Since the knowledge on the life histories of the benthic macroinvertebrates from Iran is lacking up to now, the identified benthic macroinvertebrates in the Zayandeh Rud River is useful to develop further studies in Iran. Generally, more taxonomic work should be done for the identification of the organisms to species level.

Among the biological indices, the revised ASPT had the highest correlation with water quality parameters. The absence of pollution sensitive benthic macroinvertebrates and the presence of tolerant ones (e.g. *Physa acuta*) supported the classification of the downstream stations as “severely polluted” by using the revised ASPT, complemented by water quality parameters.

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