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RESEARCH ARTICLE

Physicochemical and biological status of Aghlagan river, Iran: effects of seasonal changes and point source pollution

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Abstract

Although the macroinvertebrates have been widely used as bio-indicator for river water quality assessment in developed countries, its application is new in Iran and data on the health status of the most ecologically important rivers in Iran is scarce. The present study aimed at monitoring and assessing the ecological quality of Aghlagan river, northwest of Iran, using integrated physicochemical-biological approaches. A total of 14,423 samplings were carried out from the headwater to downstream sites at four stations (S1, 2, 3, 4) by a Surber sampler (30 cm × 30 cm) from June 2018 to April 2019. The results obtained from macroinvertebrate biotic index revealed that the genera of Gammarus (Amphipoda) and Baetis (Ephemeroptera) were the most abundant in all seasons. The PAST software was applied to analyze the diversity indices (Shannon-Weiner diversity, Evenness, and Simpson indices). Based on the cluster analysis, S3 established the least similarity to other stations. The average frequency of each macroinvertebrate species was determined by one-factor analysis of similarities (ANOSIM). In accordance with canonical correspondence analysis (CCA), temperature and phosphate were found as the dominant factors effecting the macroinvertebrate assemblage and distribution. Moreover, the results obtained from the biological indices concluded very good quality of S4 by Helsinhoff and EPT indices and fair quality using BMWP index. The data on the macrobenthos assemblage and dynamics in the Aghlagan river across a hydraulic gradient provided useful information on water management efforts that assist us to find sustainable solutions for the enhanced quality of the river by balancing environmental and human values.

Keywords Biological indices · Freshwater · Macroinvertebrates · Organic pollution · Seasonal effect

Abbreviations

ANOSIM	Analysis of Similarity
APHA	American Public Health Association
AusRivAS	Australian River Assessment System
BEAST	BEnthic Assessment of SedimenT
BIBI	Benthic Index for Biotic Integrity
BMWP	Biological Monitoring Working Party
CCA	Canonical correspondence analysis
DO	Dissolved oxygen
EC	Electrical conductivity

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EPT	Ephemeroptera, Plecoptera, and Trichoptera
HFBI	Hilsenhoff Family Biotic Index
IBI	Index of Biotic Integrity
MDO	Mean dissolved oxygen
PAST	Paleontological Statistics
PIBI	Periphyton Index for Biotic Integrity
RIVPACS	River Invertebrate Prediction And Classification Scheme
S	Station
SIGNAL	Stream Invertebrate Grade Number Average Level
SIMPER	Similarity percentage
TEM	Temperature

Introduction

Understanding of river health condition and water quality arises through the measurement and response of aquatic flora and fauna, as many of these organisms are sensitive to physical and chemical changes in an aquatic ecosystem (Bayrami et al. 2017; Sun et al. 2019). Traditionally, river monitoring has focused on water chemistry and hydrological characteristics. More recently, biological indices have been developed to understand water quality, river health, and waterbody classification (Aguiar et al. 2015). These indices aim to provide simplified measures of diverse communities, thus allowing easy understanding of the outputs for non-specialist, water resource managers (Morgan et al. 2007). Such indices are a powerful tool for detecting environmental changes in river ecosystems (Atazadeh et al. 2007). Therefore, the development of ecological response (incorporating biological indices) has great potential to improve water quality and stream condition.

River is a dynamic ecosystem of aquatic species such as macroinvertebrate, which is affecting by various anthropogenic activities such as agriculture, aquaculture, dam, dumping, domestic runoff, and industrial exploitation (Wang et al. 2020). This encouraged the development of a wide range of methods for monitoring the water quality using physical and chemical parameters and/or design indices of biotic integrity (IBI) to assess the type of contamination. Aquatic macroinvertebrates play a vital ecological role in freshwater bodies and in the food web (Abidi et al. 2018; Thorp and Rogers 2011). The significance of macroinvertebrates as biomonitoring tool can be explained based on a number of factors: (i) most macroinvertebrates have limited migration patterns or a sessile mode of life, thus representing the status of the sampling area; (ii) their lifespan is relatively short hence, the changes in the composition of their community can be used for rapid detection of the environmental changes; (iii) they are sensitive to pollutants and critical conditions such as the lack of dissolved oxygen and fluctuation of the temperature. The macroinvertebrates fauna is influenced by several hydrological, climatic, physicochemical, (e.g. pH and electrical conductivity) (Florencio et al. 2014), and biological (e.g. nutrient concentration and organic pollution factors) (Lancaster and Downes 2018). Furthermore, rapid population growth has brought about an increased contamination of aquatic environment. In some instances, it results in a disruption in stream substrate and changes in the distribution of macroinvertebrates wherein the sensitive species are replaced by resistant ones (Eady et al. 2014; Arimoro and Keke 2017). In this context, implementation of ecological management tools and controlling the rivers health condition by biological indices is of importance in developing countries. Though, checking the physiochemical parameters have been a conventional approach for decades, biological index attracts more attention owing to be affordable and highly efficient (Lenat and Barbour 1994).

Over the past couple of decades, macroinvertebrates were widely used as bio-indicator in the northern American countries, European ecoregions, and some countries in Northeast Asia (Beyene et al. 2009; Borisov et al. 2016; Lydy et al. 2000). In Iran, however, biological monitoring using macroinvertebrates is new and the studies on river pollution status using bio-markers are very few (Aazami et al. 2015; Shokri et al. 2014). In the present study, for the first time, the health condition of Aghlagan river, which is the main source of drinking water and ecologically important river in the northwest of Iran, is assessed throughout a year (2018–2019) using macroinvertebrates as bio-indicator. The following objectives were set and assessed during the four seasons: (i) studies on the physicochemical properties of the river; (ii) identification of macroinvertebrates of this river at family level; (iii) investigating the status of the point source contamination such as the establishment of fish farming and their impact on the distribution of macroinvertebrates; (iv) developing monitoring system and formulating protection guidelines for this ecosystem.

Material and methods

The study region

The Aghlagan (Bulakhlar) river is the primary tributary of the Balekhluchai river in Nir country, situated in the Ardabil province in Iran. This river is one of the main sources of drinking water in Nir, Sarien and Ardabil. It originates from Sabalan and Bozgoush mountains in northwest of Iran. Discarding municipal, industrial, and agricultural wastewater plus constructing fish farms alongside the river are the main sources of pollution in this river (see Fig. 1).

The sampling was performed at four sites along the longitudinal river gradient during the June 2018 to April 2019. Two

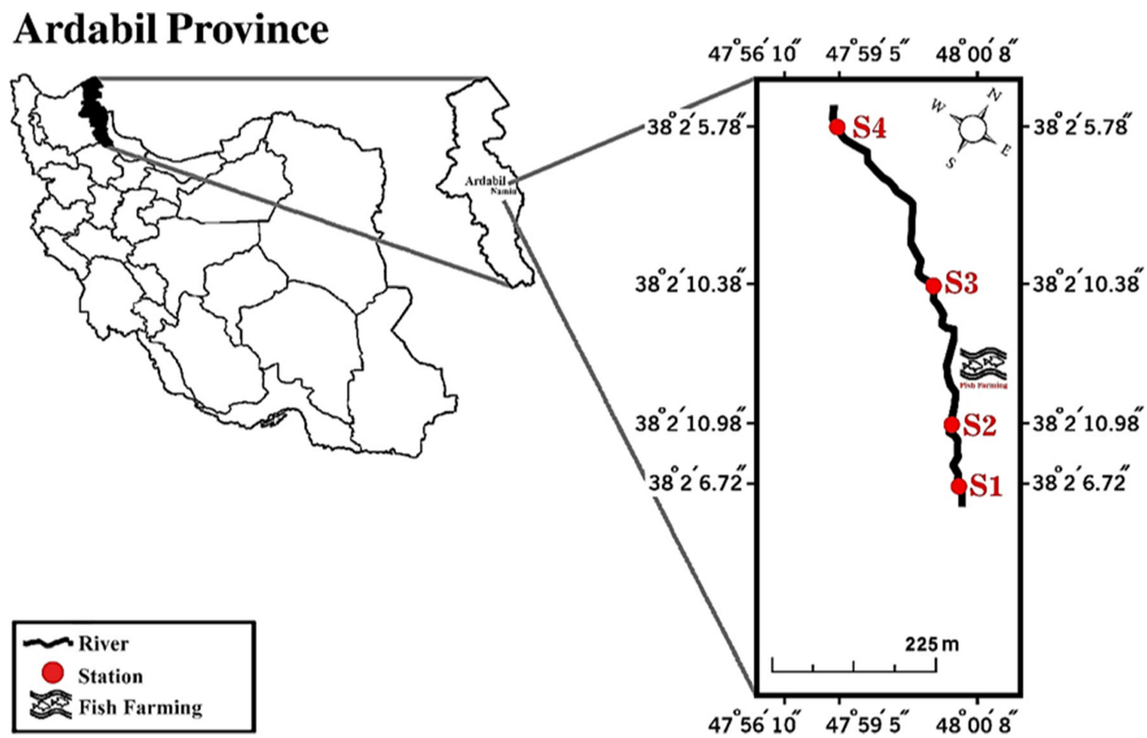


Fig. 1 Map of study area showing the four sampling stations on the Aghlagan river (shown by thick line) and the fish farming location (S1–S2: upstream and S3–S4: downstream)

sites were located upstream before fish farming operation (S1 and S2) and another two sites were located downstream and after the fish farming (S3 and S4). The detailed characteristics of the sampling stations are given in Table 1. The location of S2 and S3 was chosen before and after the fish farm (~ 1 km) to assess the effect of the point source pollution. On the other hand, the distance between the S3 to S4 was about two-times longer in order to assess the capacity of river self-purification. The quantitative sampling was carried out using a Surber (0.3 m × 0.3 m, 0.09 m^2) for the edge and riffle mesohabitat. The protocols used for the rapid bioassessment of rivers were followed (Barbour et al. 1999) and repeated three-times. The collected material was sieved through 500 mm mesh screens and gathered in plastic jars in the field. The collected water samples were preserved with 5% buffered formalin until laboratory analyses and counting (APHA 1999). All the macroinvertebrate in the sediment and water samples were identified to the lowest possible taxonomic level (family level) with a dissecting a microscope according to (Pescador et al. 2004; Tachet et al. 2000).

The temperature (T, °C), pH, electrical conductivity (EC, µs/cm), and dissolved oxygen (DO, mg/L) of water were determined in situ by a portable multi-parameter meter (model WTW, made in Germany). The water samples were collected from each station to measure the concentrations of nitrate (NO_3^-, mg/L), ammonium (NH_4^+, mg/L), and phosphate (PO_4^{3-}, mg/L) in the laboratory following American Public Health Association (APHA 1999) procedures.

Biotic index

There are a number of biological indices that are derived from multivariate analysis techniques such as RIVPACS (River Invertebrate Prediction And Classification Scheme), which was developed in UK (Wright et al. 1993, 1998), and BEAST (BEnthic Assessment of SedimenT) that was

Table 1 Depiction of the sampling stations

Station no.	Coordinates	Altitude (FT)	Substrate
1	38° 2′ 6.72″ N; 47° 59′ 2″ E	5382.8	Large cobbles-clay and silt
2	38° 2′ 10.98″ N; 47° 59′ 5.58″ E	5370.9	Clay and silt
3	38° 2′ 10.38″ N; 47° 59′ 9.88″ E	5337.3	Large cobbles-clay and silt
4	38° 2′ 5.78″ N; 47° 59′ 31.51″ E	5287.6	Large cobbles-clay and silt

developed in Canada (Reynoldson et al. 1997a, b), AusRivAS (Australian River Assessment System) (Parsons et al. 2002), and SIGNAL (Stream Invertebrate Grade Number Average Level), both of which were developed in Australia (Chessman et al. 1997). Multi-metric techniques (biotic integrity indices) are used as an alternative approach as they maintain an integrated balance in adaptive biological systems between elements (e.g. species, genus, and assemblage) and processes (e.g. nutrient and energy dynamic, biotic interaction, and meta-population process) in natural habitats (Karr 1996). The concept of biotic integrity has been developed for fish (Index of Biotic Integrity; IBI) in shallow rivers in USA (Karr 1981). According to Gordon et al. (2004), within the biotic integrity index minimal disturbance to the system has negligible impact on the biological integrity of the system. There are several well-known biological indices based on biotic integrity including: IBI (abovementioned); BIBI (Benthic Index for Biotic Integrity) based on macroinvertebrates (Kerans and Karr 1994); PIBI (Periphyton Index for Biotic Integrity) employing algal periphyton (Hill et al. 2000). In the present study, three biotic indices were selected based on their current or potential use in biological monitoring systems in Iran and the fact that they were developed outside of Iran. The biotic indices were the following: (1) BMWP-Biological Monitoring Work Party Scoring System (Walley and Hawkes 1996, 1997), (2) HFBI-Hilsenhoff Family Biotic Index (Hilsenhoff 1988), and (3) EPT index of Ephemeroptera, Plecoptera, and Trichoptera (Lenat 1988).

Data analysis

The data analysis was carried out using PAST software (version 3.25). Before statistical analysis, Shapiro–Wilk test was applied to set the normality of data and the significance level to $p < 0.05$. For total macroinvertebrate, univariate diversity techniques vis. Shannon-Weaver's diversity index ($\hat{H}$), Dominance (D), evenness index (E), and Simpson (1-D) were used in this study. Multivariate technique (ANOSIM, SIMPER, and CLUSTER) was used to investigate differences between stations. The macroinvertebrate communities were compared in various seasons using one-factor analysis of similarities (ANOSIM) and the SIMPER test (square root, Bray-Curtis similarity) were used to determine the average contribution rate of each macroinvertebrate species to similarity in the different seasons (Liao et al. 2016). The CLUSTER analysis (Bray-Curtis similarity matrix, PAST 3.5) was performed using the group—abundance linking considering the similarity between the stations in different seasons. Finally, canonical correspondence analysis (CCA) via CANOCO 5 was used to examine the correlations between macroinvertebrate species and environmental variables (Ter Braak and Smilauer 2013). The one-way analysis of variance test (one-way ANOVA) was used to assess the status of physicochemical parameters and biological indicators. A Tukey post hoc test was applied to indicate the differences between the physicochemical parameters and biotic indexes.

Results

Macroinvertebrates sampling-physical and chemical characteristics-univariate analysis

Macroinvertebrates

During the survey, 14,423 macroinvertebrate individuals related to 10 families were collected from the selected four sites in total (Table 2). Overall individuals collected during the four seasons were 3858, 3993, 3156, and 3416 in autumn, summer, winter, and spring, respectively. The most abundant family was Gammaridae (12,396 individuals), thereupon Baetidae (2644 individuals), Heptogeniidae (670 individuals), Chironomidae (760 individuals), Hydropsichidae (493 individuals), and Simuliidae (607 individuals). The biomass of

Table 2 A list of macroinvertebrates recorded in four stations on Aghlagan river

Phylum	Class	Order	Family	S1	S2	S3	S4
Arthropoda	Crustaceae	Amphipoda	Gammaridae	*	*	*	*
Arthropoda	Insecta	Ephemeroptera	Baetidae	*	*	*	*
Arthropoda	Insecta	Ephemeroptera	Heptageniidae	*	*	*	*
Arthropoda	Insecta	Trichoptera	Hydropsychidae	*	*	*	*
Arthropoda	Insecta	Diptera	Chironomidae		*	*	
Arthropoda	Insecta	Diptera	Simuliidae		*	*	
Arthropoda	Insecta	Diptera	Tabanidae		*	*	
Annelida	Oligochaeta				*	*	
Annelida	Hirudinea			*	*	*	*
Mollusca	Gastropoda				*		*

Table 3 Characteristics of the sampling sites in Aghlagan river

Season	Parameter	Mean ± SD	*p* value	*F* value
Summer	NO_3^-	3.29 ± 1.60	0.08	3.25
	PO_4^-	1.96 ± 1.14	0.08	6.94
	O_2	5.67 ± 0.47	0.08	46.80
	TEM	14.87 ± 0.24	0.97	46.80
	FLOW	4.23 ± 0.01	0.00	3.69
Autumn	NO_3^-	4.65 ± 0.02	0.01	46.80
	PO_4^-	1.93 ± 0.01	0.28	9.83
	O_2	10.71 ± 0.50	0.03	4.49
	TEM	12.93 ± 0.48	0.06	1.37
	FLOW	4.39 ± 0.005	0.03	7.93
Winter	NO_3^-	4.43 ± 1.04	0.00	9.83
	PO_4^-	0.79 ± 0.49	0.03	7.87
	O_2	10.97 ± 1.45	0.09	65.09
	TEM	11.18 ± 1.68	0.00	90.18
	FLOW	4.39 ± 0.005	0.03	7.93
Spring	NO_3^-	3.68 ± 0.77	0.00	1.76
	PO_4^-	2.69 ± 0.60	0.01	3.42
	O_2	10.32 ± 0.51	0.00	10.37
	TEM	15.92 ± 0.09	0.01	46.12
	FLOW	3.08 ± 0.82	0.02	260.76

the macroinvertebrates in summer, autumn, winter, and spring were respectively 1565.33, 2508.28, 851.21, and 2541 mg/m^2. The status of the four dominant species was reviewed at the selected four sites in all seasons. At the third station (after the fish farm): Chironomidae and Simuliidae families from the Insecta class, Gammaridae family from the Crustaceae class, and Hirudinidae family from the Hirudinea class were more abundant when compared to those of other sampling sites.

The Amphipoda order was highly frequent at S3 in the autumn and was the least at S1 in the spring. This class was observed almost in all seasons and all the studied points. The inter-station study showed no significant differences between the stations ($p > 0.05$), however, the survey on Amphipoda order showed a significant alteration in the population size of this order between autumn and other seasons ($p < 0.05$). On the other hand, the occurrence of Ephemeroptera order was high at S4 in the winter and least at S2 in the spring. In the inter-station survey on this order, its population size in S4 was significantly higher than the other stations ($p < 0.05$). In the case of the Diptera order, it was the most common order at S3 in the summer and was less frequent at S1 in the spring. The inter-station studies on this order showed that the S3 population was significantly prevailed over other stations ($p < 0.05$). The survey on the Tricoptera order demonstrated its topmost and bottommost frequency at S4 and S1 in the spring, respectively. In the inter-station study, no significant differences were observed for the population size of this order among the stations ($p > 0.05$).

Physical-chemical properties of Aghlagan river

The changes in a number of physical-chemical variables were determined in each season and sampling date. The obtained values of physical and chemical variables are shown in Table 3. In all seasons, the water temperature varied between 10 °C and 15.92 °C. The maximum water temperature was recorded at S4 (15.92 °C) in both summer and spring. The mean dissolved oxygen (M_{DO}) concentration varied by changing the seasons and ranged between 5.6 to 10.97 mg/L. The highest concentration of nitrate was measured at S2 and S3 during the spring and autumn, respectively. Although, a high average of phosphate was recorded at S3 and S4 during the spring month, the effect of the point source (fish farming) on nitrate concentration at S3 and nonpoint source at S2 was noticeable and statistically significant ($p < 0.05$).

Table 4 Results of similarity percentages analysis (SIMPER) by Bray-Curtis method at four sampling sites

Season	Average Dissimilarity (%)				
	Station	S1	S2	S3	S4
Summer	S1		72.36	38.66	73.94
	S2	72.36		55.75	93.22
	S3	38.66	55.75		53.94
	S4	89.92	47.48	53.94	
Autumn	S1		35.98	30.94	31.76
	S2	35.98		25.38	25.46
	S3	30.94	38.25		23.02
	S4	73.62	55.66	23.02	
Winter	S1		87.82	52.22	73.62
	S2	87.82		65.97	55.66
	S3	52.22	65.97		39.85
	S4	73.62	55.66	39.8	
Spring	S1		88.85	89.36	89.92
	S2	88.85		30.47	47.48
	S3	89.36	30.47		53.94
	S4	89.92	47.48	53.94	

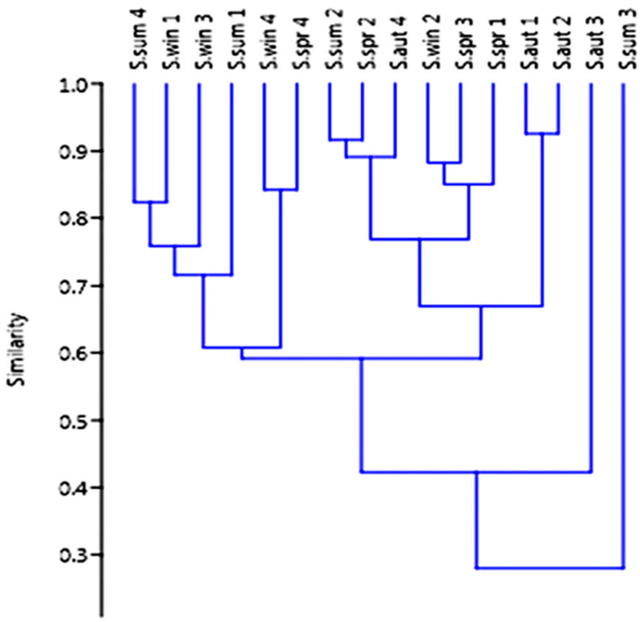


Fig. 2 Cluster analysis of the macroinvertebrates' community collected in Aghlagan river during four seasons in 2018–2019

Multivariate analysis and biological index

The significance of the differences between the macroinvertebrates' communities among the stations 1 to 4 was computed by one-way ANOSIM analysis. The results showed that the second station had a statistically significant difference with the other stations in the frequency of macroinvertebrate (p = 0.002, r = 0.16). Moreover, the SIMPER analysis was used to determine the taxa in charge of the observed differences among the groups of samples in four seasons. The results of SIMPER analysis disclosed that the Gammaridae family was the major taxon with highest contribution to the differences among family groups. Table 4 summarizes the community dissimilarity indexes at four seasons.

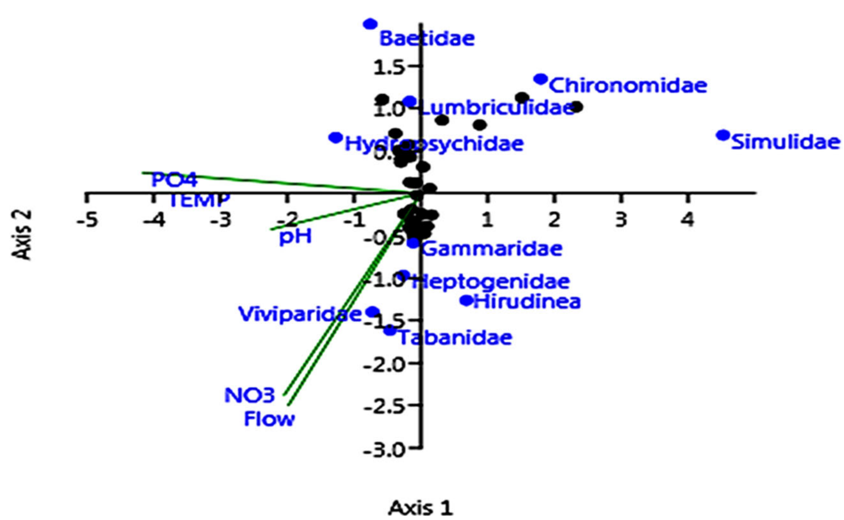
Table 5 Eigenvalues for CCA axes

Parameter	Axis 1	Axis 2	Axis 3	Axis 4	Axis 5
Eigenvalues	0.11	0.06	0.01	0.009	9.80
%	54.66	33.49	7.04	4.81	0.00

On the other hand, the Classical hierarchical cluster analysis based on a Bray-Curtis similarity index was used to compare the biomass similarity at the studied stations and in all seasons. The outcomes of this analysis revealed that the third station had the least similarity (30%) to other stations in summer and autumn (Fig. 2). Subsequently, the relationship between the macroinvertebrates and environmental factors were evaluated by canonical correspondence analysis (CCA). The results of CCA analysis showed that the temperature and phosphate concentration were the main environmental factors that affected the distribution of macroinvertebrates (Fig. 3). The first two axes of CCA explained 54.66% variance. The score of axis two (33.49% variance, eigenvalue equal to 0.069) sites was positively correlated with phosphate (r = 0.19, $p < 0.05$) and temperature (r = 0.14, $p < 0.05$) and negatively correlated with pH ($r = -0.02$, $p < 0.05$), nitrate ($r = -0.32$, $p < 0.05$), and flow ($r = -0.33$, $p < 0.05$). Data on the characteristic values of each ordination axis along with the correlation coefficients among the macroinvertebrates' functional groups and environmental factors are provided in Table 5.

The results of Shannon-Weaver diversity (H), Dominance (D), evenness (E), and Simpson (1-D) indices are shown in Fig. 4. The Shannon-Weaver index (H) showed maximum values in S2 and S4 (1.35 and 1.32) and minimum values in S1 and S3 (1.06 and 1.21). The Simpson index (1-D) showed

Fig. 3 CCA ordination diagrams of macroinvertebrates and environmental factors in Aghlagan river



maximum values in S2 and S4 (0.73 and 0.71) while minimum values in S1 and S3 (0.59 and 0.65). The evenness index (E) showed the maximum values in S2 and S4 (0.96 and 0.93), while minimum values were in S1 and S3 (0.72 and 0.84), and the results of this index were similar to the previous indices. On the contrary, the Dominance diversity index (D) showed significant variations among the studied stations being highest in S1 and S3 (0.40 and 0.34) and lowest in S2 and S4 (0.26 and 0.28).

The mean values of Hilsenhoff Family Biotic Index (HFBI) ranged from 4.07 to 5.29. The lowest value was at S4 in the autumn and the highest value was at S3 in the summer (Table 6). The differences among sampling sites were significant ($p < 0.05$). Mean values for biological monitoring working party (BMWP) index varied between 21 and 36.01. The differences among the mean values of BMWP were significant ($p < 0.05$) with lowest and highest values at S1 in spring and at S3 in winter, respectively (Table 6). The mean values of EPT (Ephemeroptera, Plecoptera, and Trichoptera) index were recorded from 10 to 385. Both the highest and the lowest values were recorded in spring at stations 4 and 1, respectively.

Within the same column, there are significant differences ($p < 0.05$) between the values indicated with different letters (a, b, c).

Discussion

Biological indices can be used in conjunction with multivariate statistical analysis to understand the sensitivity of the aquatic biota and to determine the factors drive the control of their responses (Pastorino et al. 2020). Eutrophication of rivers resulted from the speeded-up anthropogenic pressure on the aquatic environment is a serious ecological problem in most developing countries including Africa, e.g. Ethiopia (Devi et al. 2008), and Kenya (Ndiritu et al. 2003), from Asia, Iran (Sharifinia et al. 2012) and India (Selvanayagam and Abril 2015) and some other countries (Blettler et al. 2019). In Iran, the pollution from point and non-point origins has disrupted the ecosystem of most rivers. Therefore, the use of macroinvertebrate is the best biological method to assess the status of river ecosystems, which can provide good management scenarios after bioassay. In the present study, the

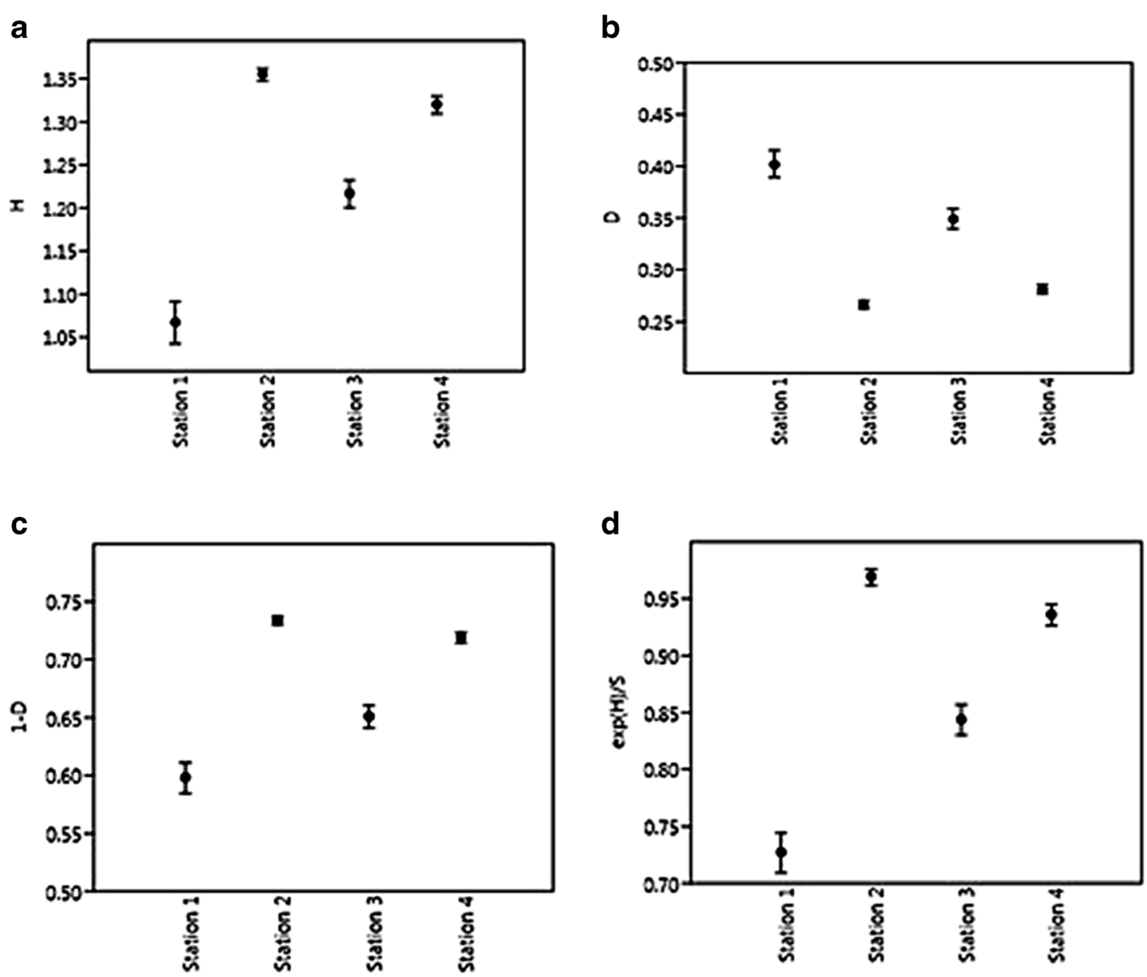


Fig. 4 Taxonomic diversity (**a** Shannon-Weaver, **b** Dominance, **c** Simpson, **d** evenness index) at the four sampling sites in Aghlagan river

Table 6 Mean ± SE of macroinvertebrate metrics per site and season in the Aghlagan river (2018–2019)

Season	Station	EPT	HFBI	BMWP
Summer	S1	81 ± 28.28[a]	4.43 ± 0.34[a]	28 ± 7.07[a]
	S2	21 ± 1.41[a]	4.16 ± 0.04[a]	26 ± 1.41[b]
	S3	137 ± 1.07[b]	5.29 ± 0.4[b]	28.5 ± 3.54[a]
	S4	120 ± 20.51[c]	4.16 ± 0.02[a]	26.5 ± 0.71[b]
Autumn	S1	37.50 ± 15.96[a]	4.12 ± 0.06[a]	31 ± 0.00[a]
	S2	16.50 ± 14.85[b]	4.18 ± 0.05[a]	35.50 ± 0.07[b]
	S3	40 ± 2.83[c]	4.16 ± 0.01[a]	31 ± 0.00[a]
	S4	40.5 ± 6.36[c]	4.07 ± 0.05[a]	35.10 ± 0.09[b]
Winter	S1	136.5 ± 68.59[a]	4.15 ± 0.08[a]	29.50 ± 4.95[a]
	S2	62.5 ± 37.48[b]	4.12 ± 0.01[a]	34.88 ± 0.00[b]
	S3	85 ± 37.48[c]	4.66 ± 0.43[b]	36.01 ± 0.00[c]
	S4	348 ± 128.69[d]	4.09 ± 0.01[a]	35.68 ± 0.00[c]
Spring	S1	10 ± 11.31[a]	4.96 ± 0.26[a]	21 ± 1.41[a]
	S2	51.5 ± 19.09[b]	4.10 ± 0.05[b]	22.50 ± 0.71[b]
	S3	39.50 ± 0.71[c]	4.32 ± 0.05[b]	22 ± 0.12[b]
	S4	385.5 ± 121.51[d]	4.18 ± 0.16[b]	29.50 ± 2.12[c]

results of chemical variables revealed high concentrations of nitrate at S2 and S3 and high average values of phosphate at S3 and S4. The values obtained for the average of nitrate, phosphate, and dissolved oxygen clearly indicated that the pollution at S3 was mainly originated from fish farm. The alteration in physicochemical parameters such as increased nitrate, phosphate, and ammonia along with oxygen depletion are the detrimental effects of fish farming on river and stream ecosystems (Barik et al. 2019). In this study, macroinvertebrates' resistant to organic contamination increased at S3 (located after fish farming) when compared to other stations. The effluent from fish farming could influence the arrangement and distribution of benthic populations and consequently, the tolerant taxa such as Diptera was increased, while the population of the sensitive taxa such as Ephemeroptera and Tricoptera was decreased. These results are consistent with the findings on the effects of fish culture reported by other researchers (Fries and Bowles 2002; Roberts et al. 2009). Although, the Gammaridae was constantly present at the four studied stations, it was the highly abundant family at S3 in all seasons. As reported, the Cructacea are more tolerant than insects to organic contamination (Soucek and Dickinson 2012).

Relationship between macroinvertebrates' communities and environmental factors

According to the CCA, temperature and phosphate were the main environmental factors that affected the distribution of macroinvertebrates. In terms of the spatial arrangement of the macroinvertebrates' communities, the highest diversity of macroinvertebrates was observed in autumn and winter and the lowest diversity was in spring. The high diversity in cold seasons can be explained based on the lower and stable temperature of the river environment. On the other hand, the lower species diversity of the macroinvertebrates in warm seasons could be due to the reduced gametogenesis and reproduction, decreased dissolved oxygen, and increased hydrogen sulfide deposition (Patang et al. 2018; Saravanakumar et al. 2007). Moreover, the results of this study revealed that the determined concentrations of phosphate have had positive correlation with the distribution of macroinvertebrates. However, this relation can have negative effects on the diversity of macroinvertebrates if the concentrations were at higher ranges (Struijs et al. 2011). It is noteworthy that the effects of chemical parameters on the structure and distribution of macroinvertebrates are not generally counted as significant because their effect on the river ecosystem is considered temporary (O'Neill et al. 2016).

Diversity and biotic index

The Shannon-Weaver index was proposed by Shannon and Weaver in 1949 for assessing the biodiversity of an area (Almeida et al. 2013). The classification of this index was defined based on the biodiversity of macroinvertebrates in 1968 wherein the low value of this index shows the organic contamination of the area. In this context, Shannon-Weaver index is related to the pollution level of the water as follows: (i) the index value bellow 1 means very less diversity level or highly polluted water; (ii) the value between 1 and 2 indicates less diversity or moderate pollution level; (iii) the index value between 2 and 3 shows moderate diversity or light pollution level; and finally (iv) the values between 3 and 4 points the high level of diversity when the water quality is classified as slight (Kumar and Sharma 2014). According to the result, the highest Shannon-Weaver index values were observed in the sampling site of S2 and S4, which could be resulted from the rich and undisturbed habitat structure of the area. On the contrary, the lowest values were obtained from the S1 and S3 stations mainly due to the high level of organic contamination in S3 caused by the wastewater of the fish farm and the bioturbation of the bottom (Sharifinia et al. 2012). In line with the results of this work, the study conducted by Kumar and Khan, on the distribution and diversity of macroinvertebrates in Pondicherry mangroves, the Dominance index was positively correlated with Berger-Parker and Fisher alpha indices and negatively correlated with evenness (E), species richness, and equitability (J) indexes. Conclusively, the results of diversity indexes in S3 indicated the moderately pollution of the site and the macroinvertebrate community has been under stress due to the anthropogenic factors (Pesce et al. 2020).

The EPT taxa are widely applied to assess anthropogenic impacts on aquatic ecosystems (Couceiro et al. 2012; Moya 2007). The Plecoptera families disappear as a consequence of the vanishing of plant habitats and disruption of the river bed. The response to human activities is much faster by Plecoptera order compared with Ephemeroptera and Trichoptera families wherein they are rapidly diminished and replaced by more resistant species (Ab Hamid and Md Rawi 2017). In the present study, no families of Plecoptera order were identified in Aghlagan river. This implies the habitat disturbance, which could be resulted from the attendance of a large number of tourists in the area and the effluent of the fish farm. Based on Lenat and Barbour (1994), the downstream reaches have normally higher EPT index values than headwater streams that indicates the small validity of EPT index (Lenat and Barbour 1994). The results of this study were in compliance with Lenat and Barbour report wherein the S1 (headwater) and S4 (downstream) stations had respectively the smallest and highest EPT index in all seasons. The Hilsenhoff Biotic index estimates the overall tolerance to low dissolved oxygen or high organic contamination. The sensitivity of the organisms to organic pollution is classified to a tolerance number ranged from zero to ten wherein zero designated for most sensitive organisms and ten is given to the most tolerant one (Pinto et al. 2004). The results of Hilsenhoff Biotic index over Aghlagan river showed "very good" water quality in the most station.

The Biological Monitoring Working Party (BMWP) family level was developed in the Freshwater Ecology Institute (Great Britain) as a part of RIVPACS system—a basis for evaluating the health condition of running waterbodies in Great Britain and Australia. The results of BMWP index showed that all stations were in proper water quality category with the exception of S3 in the spring. It is worth mentioning that the application of one biotic index cannot accurately impart the health status of an aquatic ecosystem. Herein, the BMWP biotic index showed lower water quality than the Hilsenhoff index for all the stations, which is consistent with the literature (Lydy et al. 2000). A summary of the health status of Aghlagan river is given in Table 7.

Table 7 Summary of the health status of Aghlagan river using various indices over a year (2018–2019)

Season	Station	EPT	HFBI	BMWP
Summer	1	Excellent	Good	Poor
	2	Good	Very good	Poor
	3	Excellent	Fair	Poor
	4	Excellent	Very good	Poor
Autumn	1	Excellent	Very good	Poor
	2	Good-fair	Very good	Poor
	3	Excellent	Very good	Poor
	4	Excellent	Very good	Poor
Winter	1	Excellent	Very good	Poor
	2	Excellent	Very good	Poor
	3	Excellent	Good	Poor
	4	Excellent	Very good	Poor
Spring	1	Fair	Good	Poor
	2	Excellent	Very good	Poor
	3	Excellent	Good	Poor
	4	Excellent	Good	Poor

Conclusion

In this study, the water quality of Aghlagan river in Iran was assessed with reference to a number of physicochemical properties (temperature, pH, DO, EC, NO_3^-, NH_4^+, and PO_4^{3-} concentrations) and biological indices based on the macroinvertebrate fauna from June 2018 to April 2019. The most abundant family was Gammaridae, followed by thereupon Baetidae, Chironomidae, Heptogeniidae, Simuliidae, and Hydropsichidae families. Nonetheless, no families of Plecoptera order, which are highly sensitive species to pollution and habitat disruption, were identified in Aghlagan river. In this case, the large number of tourists in the area and the effluent of the fish farm could be appointed as the habitat disturbance and pollution sources. This was consistent with the Shannon-Weaver index results, which were the least for S3 following the high organic load through fish farm wastewater and the bottom sediments bioturbation. Moreover, the highest diversity of macroinvertebrates was observed in autumn and winter owing to the lower and stable temperature of the river environment. On the contrary, the reduced gametogenesis and reproduction, decreased dissolved oxygen, and increased hydrogen sulfide deposition could be of the main reasons for the lowest diversity in spring. The high concentrations of nitrate and phosphate, and relatively low dissolved oxygen clearly signified that S3 pollution was derived from fish farm. As per CCA analysis, temperature and phosphate were also found as the key environmental factors that affected the distribution of macroinvertebrates. The data on the macrobenthos assemblage and dynamics in the Aghlagan river across a hydraulic gradient could be useful in water management efforts to find sustainable solutions in the river by balancing environmental and human values. Based on the reported results in this study, it is recommended to minimize the point source pollution by facilitating fish farms to more advanced discharge treatment systems. Further studies are required to address the operational rules of water management concerning economic and social objectives. This is because the water allocation in this river is multilateral in character and must support social and economic aspirations.

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Shima Rahim Pouran: Writing–review and editing, supervision, visualization.

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Compliance with ethical standards

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