## Original Article

# Relationship between Biological and Qualitative Indices in Surface Waters Receiving the Effluent of Fish Farms in the Northwest of Iran

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### Abstract

**Background:** Water quality is usually measured using various indicators based on physical, chemical and biological parameters. By using the biological index that is based on the identification of the arthropod families, it is possible to make a logical judgment about the ecosystem condition. The aim of this study was measuring correlation coefficients between qualitative and biological Indices.

**Methods:** Water samples were collected 27 samples in northwest of Iran and aquatic insects' samples 54 in 2019. The NSFWQI and IRWQISC as the most important indices of physical and chemical quality of water ranged from 54.45–76.21 and from 41.32 to 77.40, respectively.

**Results:** A total of 2,953 aquatic insects were collected, and biological Index ranged from 6.26 to 3.38. It can be stated that increasing in the concentration of pollutants in the source and end of the river could lead to a sharp decrease in biological index. IRWQISC index, the effluent stations of fish farms can fit into 'fairly bad quality' and 'moderate quality' categories.

**Conclusion:** The linear regression analysis revealed a significant relationship between the Hilsenhoff biological Index and the physiochemical parameters of pH, DO (Dissolved Oxygen) and total dissolved solids. The activity of fish farms and discharging their effluents into water sources, can change the physical, chemical and biological parameters of receiving waters, therefore it is recommended that the location of these units be reviewed and also the appropriate treatment for such effluents should be considered, so that the health risks caused by them can be effectively reduced.

**Keywords:** Water quality index; Biological index; Aquatic insects; Water quality; Iran

## Introduction

Rivers and running waters have been always regarded as essential resources for human beings throughout history providing them with necessary water for drinking, farming and industrial purposes (1). One of the main reasons for the location and establishment of urban, agricultural and industrial centers is the presence of abundant water resources (2). These resources, however, are under grave threat by consumers (3). Hence, it is necessary to identify the resources of physiochemical and biological pollutants such as BOD (Biological oxygen demand), fecal coliform, nitrate, COD (Chemical oxygen demand), ammonium, phosphate, turbidity, TDS (Total dissolved solids), and etc. Raise awareness regarding their changing trends and devise effective strategies to hinder the deterioration of water quality and find ways to improve it (4–7). Several countries have provided their special guidelines to monitor the quality of water resources or have tried to follow the standard guidelines suggested by international organizations (8, 9). The water quality index is measured via physical, chemical and biological parameters (10, 11) and is regarded as a criterion for the categorization of waters

based on using standard parameters (12). These Indices are mathematical tools to quantify the descriptive data on the quality level of waters (13, 14). The National Sanitation Foundation Water Quality Index (NSFWQI) and Iran Surface Water Quality Index (IRWQI_{SC}) Indices are two of the most complete and comprehensive water quality Indices (8, 9, 15, 16) used to study waters, monitor and detect pollutants in waters and qualitatively evaluate the zoning of surface waters by measuring physical and chemical features (17, 18). These two Indices are less problematic compared to other models and because of their simplicity and availability, they are frequently employed by researchers around the world (19–22). After coding and measuring the qualitative information, the sub-index amount of each parameter is measured based on the characteristic curve for quality score classification and weighting factor of each parameter listed in the tables (23–25). The above indicators present water quality in simple terms for experts and the public by presenting numbers in certain classifications. Nevertheless, in terms of ecology, one of the best and cheapest scientific methods, compared to chemical methods, is to determine the biological health of waters and determine the effects of human activities on water qualities by biological evaluations and monitoring (26–28). The biological index works based on the detection of arthropods in families (29, 30) and can pronounce logical judgments on ecosystems (31–33). Various national and international studies have been conducted on the investigation of water qualities in line with the two above-mentioned Indices (34, 35). For example, Shukrisarvey et al. studied the quality of the Tajan river in Mazandaran Province based on the biological Indices of the Hilsenhoff biological Index, Shannon-Wiener index, physicochemical parameters and environmental Indices (36). In another study, Heidari et al. evaluated the biological features of the Kashkan river in terms of the diversity and Macro Benthic population structure (37). Moreover, Ibiwumi et al. investigated the effects of physicochemical parameters on Nigeria Macro Benthics via various Indices (38).

The purpose of this study, which was conducted for the first time in northwest Iran, was to investigate the effects of the physicochemical parameters of surface waters receiving effluents from fish farms on aquatic insects, determine the relationship between physicochemical parameters and biological indices, and also measure the correlation coefficients between qualitative and biological indices and provide basic information for the study and management of water resources in Bolaghlar area of Nair city, Ardabil Province.

## Materials and Methods

This study is a cross-sectional descriptive study conducted to investigate the effects of physicochemical parameters in surface waters (Aglagan River) receiving the effluent of fish farms on biological Indices, determine the relationship between physicochemical parameters and aquatic insects and measure correlation coefficients between qualitative and biological Indices in Boulaghlar region, Nir City, over the period from March to August 2019. According to the topography of the region and the areas where the fish farms are located, as well as the status of other land uses in the region, nine stations were selected along the Aglagan river (Fig. 1). Six series sampling of aquatic insects were done from each station throughout the study period (54 samples collected in total), and they were detected down to family level by valid entomological keys (39, 40). To determine physicochemical parameters, three series of samples were collected from each station (27 samples in total). Based on the guidelines of the Standard Methods v. 20, the measured parameters in this study included fecal coliform (MPN/100 mL), biochemical oxygen (mg/mL), nitrate (mg/L), DO (mg/L), electrical conductivity, chemical oxygen demand (mg/L), ammonium (mg/L), phosphate (mg/L), turbidity (NTU), total hardness (mg/L.CaCO₃, total

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dissolved solids (mg/L), temperature (°C) and pH. In order to analyze the correlation and relationship between the parameters and the obtained indicators, the collected data were statistically analyzed by SPSS v.22 software. To assess relationships among physicochemical parameters, Pearson's correlation coefficient was used; to determine the effects of physicochemical parameters on qualitative Indices and the effects of qualitative Indices on Hilsenhoff biological Index, regression correlational analysis was employed. In this study, NSFWQI, IRWQISC and HBI Indices were used and calculated based on standard, relevant formulas (41, 42).

## Results

### Physicochemical parameters

The mean of measured physicochemical parameters in the samples used to measure NSFWQI and IRWQI$_{SC}$ Indices and also the amounts of NSFWQI, IRWQI$_{SC}$ and biological Indices are depicted in Table 1. As can be seen in Table 1, the minimum amount of electrical conductivity (0.38 µS/cm) was observed in the upper stations of fish farms. The maximum amount of pH was detected in Station 2 (7.59) and the minimum was detected in Stations 7 and 8 (7.27). The minimum amounts of turbidity (1.03 NTU), COD (4.33 mg/L), nitrate (3 mg/L), phosphate (0.09 mg/L) and fecal coliform (4 colonies/100 ml, 7.67 MPN/100ml) belonged to Station 1. The maximum amounts of electrical conductivity (0.47 µS/cm), hardness (189.67 mg/L CaCO$_3$), BOD (12.67 mg/L), ammonium (0.35 mg/L), nitrate (16.33 mg/L), phosphate (0.55 mg/L) and fecal coliform (27.33 colonies/100ml, 53.67 MPN/100ml) were detected in Station 3. The highest temperature parameter (17.53 °C) was reported in Station 5. The maximum amount of oxygen parameter (5.97 mg/L), and the minimum amounts of dissolved solids (284.33 mg/L), hardness parameter (94.27 mg/L CaCO3), BOD (1.33 mg/L), ammonium (0.09 mg/L) and temperature (16.51 °C) were detected in Sta-

tion 6. The maximum amounts of turbidity (12.78 NTU) and COD (17 mg/L) were observed in Station 7. The highest amount of dissolved solids parameter (342 mg/L) belonged to Station 8, and the lowest amount of DO (4.74 mg/L) belonged to Station 9. The NSFWQI index ranged from 54.45 to 76.21, which according to the results of this index, the studied stations can be put into 'good quality' and 'moderate quality' categories. The maximum (76.21) and minimum (54.45) amounts of this index belonged to Station 6 and based on the IRWQI$_{SC}$ index, the studied stations ranged from 41.32 to 77.40 which puts them in the three categories of 'good quality', 'moderate quality' and 'fairly bad quality'. The maximum amount of this index (77.40) was observed in Station 6, and the minimum amount (41.32) was observed in Station 3.

### Biological index

A total of 2,593 aquatic insects were collected from all the studied stations belonging to 9 families, 9 orders and 4 classes. The highest and lowest frequencies of the families of aquatic insects belonged to Gammarida (78.63%) and Enidae (0.11%), respectively. The Hilsenhoff biological Index ranged from 6.26 to 3.56; the highest index was observed in Station 9 with the biological quality of 'fairly poor', and the lowest index was observed in Station 1 whose biological quality was in accordance with the Hilsenhoff Index (Table 2).

### Relationships among physicochemical parameters, qualitative index and biological index

The results of Pearson's correlation analysis (Table 3) revealed that there was not a significant relationship between temperature and other parameters. Investigating relationship between turbidity, TDS, phosphate, fecal coliform, nitrate, BOD, COD, electrical conductivity, total hardness and ammonium parameters showed a negative correlation with pH, DO and its saturation percentage parameters;

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however, there were positive correlations between the above-mentioned parameters and the other parameters. The pH index had a significantly positive correlation with DO and its saturation ratio. The saturation percentage of DO had no significant relationships with fecal coliform. On the other hand, the investigation of the relationship between physicochemical parameters and the Hilsenhoff Index using linear regression showed that there were signifi

cant relationships between this index and pH, DO and TDS parameters. Nevertheless, this effect was positive on TDS (Sig=0.046, B 0.027) yet negative on DO (Sig=0.037, B=-1.588) and pH (Sig=0.029, B=-4.837). Finally, there were no significant relationships between the Hilsenhoff Index and NSFWQI (Sig= 0.151, B= 0.047) and IRQQI$_{SC}$ (Sig= 0.179, B= -0.027) Indices (Table 4).
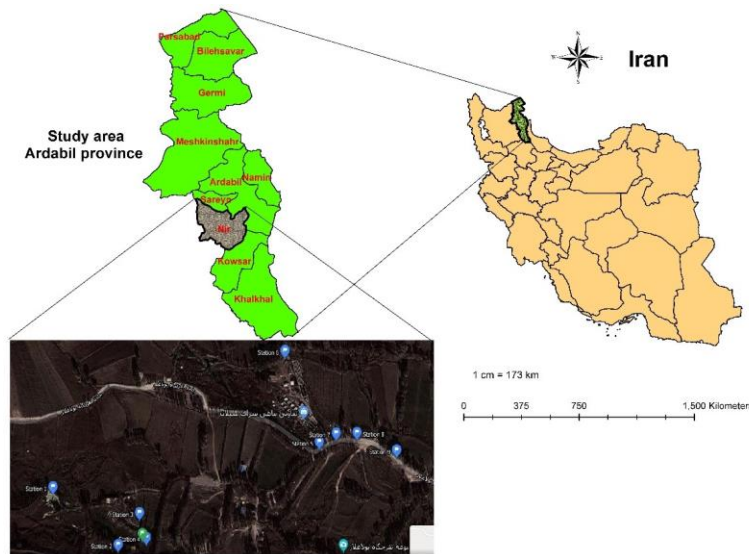


**Fig. 1.** Study area and location of samples sites in northwest of Iran, Ardabil Province, 2019
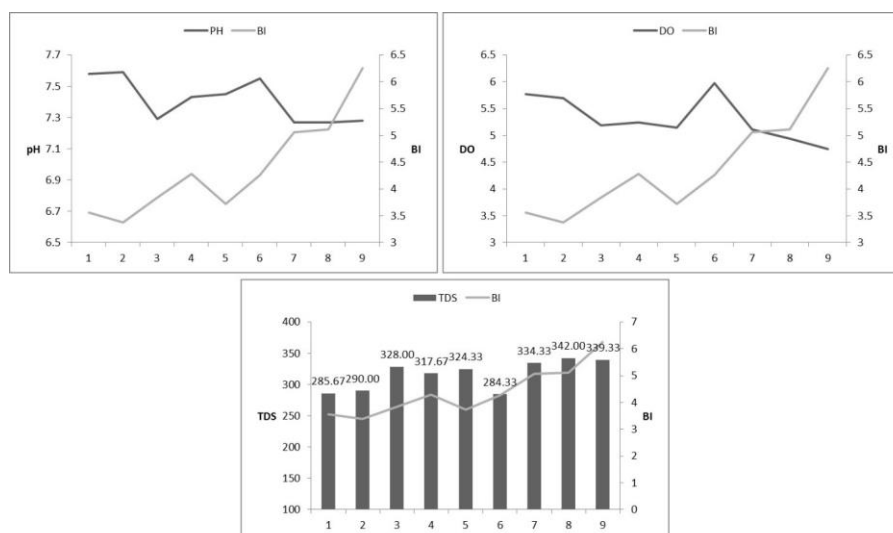


**Fig. 2.** Relationship between pH, Dissolved Oxygen (DO), Total Dissolved Solids (TDS) and Biological Index (BI) in studied stations (Northwest of Iran, Ardabil Province, 2019)

**Table 1.** Physicochemical parameters result in studies stations (Northwest of Iran, Ardabil Province, 2019)

| Parameters | Unite | Stations | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| Turbidity | NTU | 1.03 | 1.60 | 11.63 | 11.05 | 10.63 | 1.40 | 12.78 | 12.69 | 12.11 |
| pH | - | 7.58 | 7.59 | 7.29 | 7.43 | 7.45 | 7.55 | 7.27 | 7.27 | 7.28 |
| BOD | mg/L | 1.67 | 2.33 | 12.67 | 12 | 10 | 1.33 | 12 | 11.33 | 10.67 |
| Fecal Coliform | MPN/100 mL | 7.67 | 9.67 | 53.67 | 41.67 | 29.67 | 12.67 | 53.67 | 45.67 | 29.67 |
| Total Hardness | mg/L.CaCO$_3$ | 97.27 | 101.23 | 189.67 | 169.30 | 174.73 | 94.27 | 191.23 | 188.87 | 187 |
| DO | mg/L | 5.77 | 5.69 | 5.19 | 5.24 | 5.14 | 5.97 | 5.11 | 4.94 | 4.74 |
| Oxygen (Sat. Per.) | % | 72.83 | 71.43 | 63.52 | 59.99 | 59.26 | 73.22 | 65.12 | 61.76 | 57.56 |
| Temperature | °C | 17.07 | 17.06 | 17.19 | 17.20 | 17.53 | 16.51 | 16.61 | 16.62 | 16.76 |
| TDS | mg/L | 285.67 | 290 | 328 | 317.67 | 324.33 | 284.33 | 334.33 | 342 | 339.33 |
| Phosphate | mg/L | 0.09 | 0.1 | 0.55 | 0.51 | 0.46 | 0.13 | 0.46 | 0.45 | 0.43 |
| Nitrate | mg/L | 3 | 3.67 | 16.33 | 15 | 13 | 3.33 | 16 | 15 | 12.67 |
| Ammonium | mg/L | 0.16 | 0.17 | 0.35 | 0.31 | 0.27 | 0.09 | 0.31 | 0.30 | 0.27 |
| Colonies | Count | 4 | 5.33 | 27.33 | 24.67 | 21.67 | 5 | 26.67 | 26.33 | 24 |
| EC | µS/cm | 0.38 | 0.38 | 0.47 | 0.46 | 0.46 | 0.38 | 0.45 | 0.46 | 0.46 |
| COD | mg/L | 4.33 | 5 | 15.33 | 14.67 | 12.67 | 3.67 | 17 | 16 | 13.67 |
| NSFWQI | - | 72.99 | 74.58 | 55.86 | 54.45 | 56.16 | 76.21 | 55.83 | 55.25 | 55.13 |
| IRWQISC | - | 77.35 | 75.24 | 41.32 | 42.40 | 45.39 | 77.40 | 41.63 | 42.55 | 45.80 |
| BI | - | 3.56 | 3.38 | 3.84 | 4.28 | 3.72 | 4.26 | 5.06 | 5.11 | 6.26 |

**Table 2.** Aquatic insect specimens collected and the calculated bio-index values in river of Northwest of Iran, Ardabil Province, 2019

| Stations | Family | | | | | | | | | Total | BI | Water Quality |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Ephemeridae | Notonectidae | Hydropsychidae | Chironomidae | Culicidae | Asellidae | Gammaridae | Glossiphoniidae | Enidae | | | |
| 1 | 55 | 5 | 4 | 0 | 0 | 0 | 184 | 1 | 3 | 243 | 3.56 | Excellent |
| 2 | 38 | 1 | 2 | 0 | 0 | 0 | 85 | 2 | 0 | 128 | 3.47 | Excellent |
| 3 | 59 | 0 | 1 | 0 | 0 | 2 | 589 | 1 | 0 | 652 | 3.83 | Very good |
| 4 | 8 | 0 | 1 | 0 | 0 | 0 | 198 | 20 | 0 | 227 | 4.28 | Good |
| 5 | 67 | 2 | 0 | 0 | 0 | 0 | 404 | 1 | 0 | 474 | 3.76 | Excellent |
| 6 | 184 | 0 | 0 | 15 | 0 | 0 | 184 | 0 | 0 | 203 | 4.25 | Good |
| 7 | 0 | 0 | 4 | 55 | 0 | 0 | 152 | 1 | 0 | 212 | 5.05 | Fair |
| 8 | 2 | 0 | 4 | 69 | 0 | 0 | 172 | 1 | 0 | 248 | 5.11 | Fair |
| 9 | 9 | 0 | 5 | 72 | 4 | 37 | 71 | 8 | 0 | 206 | 6.26 | Fairly poor |
| Total | 422 | 8 | 21 | 211 | 4 | 39 | 2039 | 35 | 3 | 2593 | 3.97 | Very good |

**Table 3.** Pearson statistical test results and correlation between physicochemical parameters (Northwest of Iran, Ardabil Province, 2019)

| Parameter | Turbidity | pH | Temp. | DO | Oxygen (Sat. Per.) | TDS | Phosphate | Colonies | Nitrate | BOD | COD | EC | Total Hardness | Ammonium | Fecal Coliform |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Turbidity | 1 | -0.92 | 0.09 | -0.93 | -0.89 | 0.97 | 0.96 | 0.99 | 0.98 | 0.98 | 0.98 | 0.97 | 0.99 | 0.92 | 0.90 |
| Ph | | 1 | 0.25 | 0.88 | 0.72 | -0.95 | -0.84 | -0.91 | -0.88 | -0.88 | -0.91 | -0.86 | -0.93 | -0.83 | -0.88 |
| Temp. | | | 1 | -0.03 | -0.26 | -0.05 | 0.20 | 0.06 | 0.10 | 0.13 | 0.03 | 0.20 | 0.06 | 0.25 | -0.04 |
| DO | | | | 1 | 0.92 | -0.97 | -0.83 | -0.89 | -.85 | -.88 | -0.88 | -0.90 | -0.93 | -0.83 | -0.72 |
| Oxygen (Sat. Per.) | | | | | 1 | -0.87 | -0.88 | -0.87 | -0.84 | -0.87 | -0.83 | -0.92 | -0.88 | -0.80 | -0.65 |
| TDS | | | | | | 1 | 0.89 | 0.95 | 0.92 | 0.92 | 0.94 | 0.93 | 0.97 | 0.87 | 0.84 |
| Phosphate | | | | | | | 1 | 0.98 | 0.98 | 0.98 | 0.95 | 0.98 | 0.96 | 0.94 | 0.91 |
| Colonies | | | | | | | | 1 | 0.99 | 0.99 | 0.99 | 0.98 | 0.99 | 0.95 | 0.94 |
| Nitrate | | | | | | | | | 1 | 0.99 | 0.99 | 0.97 | 0.97 | 0.96 | 0.95 |
| BOD | | | | | | | | | | 1 | 0.98 | 0.98 | 0.98 | 0.96 | 0.93 |
| COD | | | | | | | | | | | 1 | 0.95 | 0.98 | 0.95 | 0.94 |
| EC | | | | | | | | | | | | 1 | 0.97 | 0.93 | 0.87 |
| Total Hardness | | | | | | | | | | | | | 1 | 0.93 | 0.90 |
| Ammonium | | | | | | | | | | | | | | 1 | 0.91 |
| Fecal Coliform | | | | | | | | | | | | | | | 1 |

Coefficients description: value of -1 meaning a total negative linear correlation,
0 being no correlation, and
+ 1 meaning a total positive correlation.

**Table 4.** Relationship between physicochemical parameters and biological index (Northwest of Iran, Ardabil Province, 2019)

| Model | Unstandardized Coefficients | | Standardized Coefficients | T | Sig.[*] |
|---|---|---|---|---|---|
| | B | Std. Error | Beta[**] | | |
| (Constant) | 40.235 | 13.127 | | 3.065 | .018 |
| pH | -4.837 | 1.771 | -.718 | -2.731 | .029 |
| (Constant) | 3.517 | .522 | | 6.739 | .000 |
| Turbidity | .104 | .054 | .591 | 1.939 | .094 |
| (Constant) | 12.816 | 3.283 | | 3.904 | .006 |
| DO | -1.588 | .617 | -.697 | -2.575 | .037 |
| (Constant) | 31.918 | 14.011 | | 2.278 | .057 |
| Temperature | -1.624 | .826 | -.596 | -1.965 | .090 |
| (Constant) | 9.871 | 3.151 | | 3.133 | .017 |
| %DO | -.084 | .048 | -.551 | -1.748 | .124 |
| (Constant) | -4.137 | 3.522 | | -1.175 | .278 |
| TDS | .027 | .011 | .676 | 2.426 | .046 |
| (Constant) | 3.652 | .671 | | 5.445 | .001 |
| Phosphate | 2.077 | 1.695 | .420 | 1.225 | .260 |
| (Constant) | 3.505 | .601 | | 5.832 | .001 |
| Colonies | .048 | .029 | .531 | 1.658 | .141 |
| (Constant) | 3.588 | .660 | | 5.440 | .001 |
| Nitrate | .073 | .054 | .455 | 1.353 | .218 |
| (Constant) | 3.634 | .594 | | 6.113 | .000 |
| BOD | .091 | .063 | .481 | 1.451 | .190 |
| (Constant) | 3.361 | .688 | | 4.882 | .002 |
| COD | .090 | .055 | .525 | 1.632 | .147 |
| (Constant) | -.426 | 3.336 | | -.128 | .902 |
| EC | 11.103 | 7.668 | .480 | 1.448 | .191 |
| (Constant) | 2.502 | 1.066 | | 2.348 | .051 |
| Total Hardness | .012 | .007 | .568 | 1.828 | .110 |
| (Constant) | 3.507 | .998 | | 3.515 | .010 |
| Ammonium | 3.547 | 3.822 | .331 | .928 | .384 |
| (Constant) | 3.755 | .635 | | 5.909 | .001 |
| Fecal Coliform | .020 | .018 | .393 | 1.132 | .295 |
| (Constant) | 5.867 | 1.032 | | 5.682 | .001 |
| IRWQI | -.027 | .018 | -.492 | -1.495 | .179 |
| (Constant) | 7.297 | 1.829 | | 3.989 | .005 |
| NSFWQI | -.047 | .029 | -.520 | -1.611 | .151 |

*p-value< 0.05: significant
**Slope of linear regression equation

## Discussion

Beneficial insects play an important role in human life. A group of these insects are considered as biological indicators of water quality, including Ephemeroptera, Plecoptera and Trichoptera. This group of insects indicates the appropriate quality of water for human use (43–46).

Based on the study findings in the previous section, the activity of fish farms can negatively affect the amount of DO by decreasing the amount of this parameter in fish farms' effluent stations as compared to upper stations. According to the findings of this study, the amount of this parameter gradually reduced from the upper sections to the lower sections of the river showing that the effluent contained a lot of

wastes of fishes, leading, in turn, to an increase in nitrate, phosphate and solid materials concentration levels, water turbidity, a higher chemical decomposition level of organic materials and a stronger need for oxygen (47). In an agreement with the findings of this study, previous research conducted in Kermanshah Province, Khuzestan Province, Yassoj, Iranian Kurdistan, the United States and Turkey have also reported that the amount of DO in the effluent of fish farms is significantly reduced (47–50).

This study revealed that the amount of pH in Stations 3 and 7 was influenced by the effluent of fish farms and was thus significantly reduced. In a study by Khoshakhlagh et al. (51), several reasons were discussed for such a decline in pH including the decomposition of wastes of fishes and food leftovers in the effluent and an increase in organic materials and pollution. The results of this study agree with the findings reported by Boventura et al. (52) and Zarzuela et al. (53) but disagree with the findings of studies by Selong and Helfrich and Dugel (49, 54).

The findings of this study showed a significant increase in the parameters of TDS, electrical conductivity, fecal coliform, nitrate, ammonium, phosphate, BOD and COD in the effluent of fish farms. Millard et al., Hynes et al., Homewood et al., Stephens et al., Pillary et al., Adem et al., Miller and Semens and Sobhani et al. have all reported in their studies that food leftovers and remaining, waste of fishes, metabolic fish products and cleansing pools can lead to such an increase in the above-mentioned parameters (9, 49, 55–60). The findings of this study agree with those previously reported from Khuzestan, Shahr-e Kurd, Yassoj, Mazandaran Province, Golestan Province, Kermanshah Province, Turkey and Minnesota in the United States (45, 47, 49, 50, 61–63). A rise in dissolved solids can also increase the amount of turbidity as also reported by Hosseini et al. (45). Because the physicochemical parameters including turbidity, TDS, phosphate, faecal coliform, nitrate, BOD, COD, electrical conductivity, total hard-

ness, and ammonium are all somehow related to oxygen consumption, so they can lead to a decrease in DO and, as a result, oxygen saturation percentage. Also, some of these compounds, such as nitrate and ammonium, cause changes in the pH of water due to some chemical interactions, especially with the production of acidic compounds, so the reason for the negative correlation between these parameters can be explained.

The findings of this research revealed an increase in the temperature of the effluent of fish farms throughout the year. Khoshakhlagh et al. named several possible reasons for such a boost in the temperature: the repelled heat of fish metabolism, water touching the bed and walls of the pools, absorbing more sunlight because of suspended food leftovers, extra energy produced by pollutant influents and decomposition of organic compounds by microorganisms in the water (51). A study conducted by Axeler et al. also corroborates the findings of this study (64) which revealed an increase in the total amount of hardness in the effluent of fish farms. These results are in agreement with previous research findings conducted in Minnesota in the United States, Kermanshah Province and Chaharmahal and Bakhtiari Province (48, 52, 64). Boyd has stated that using calcic materials produced by limestone in fish farms can lead to an increase in the overall hardness of water (65).

Stations 1, 2 and 6 located in the upper section of fish farms, and hence safeguarded against effluents, can fit into the 'good quality' category based on NSFWQI and IRWQI$_{SC}$ Indices. The changes resulted from fish farms activities significantly reduced the amount of NSFWQI and IRWQI$_{SC}$ Indices in fish farms' effluent stations; thus, they can be categorized into the 'moderate quality' and 'fairly bad quality' categories, respectively. Based on the NSFWQI index, all the stations located in the lower section of the river belong to the 'moderate quality' category whereas based on IR-WQI$_{SC}$ index, stations 4 and 8 fall into the

'fairly bad' category and stations 5 and 9 fit into the 'moderate quality' category. Such findings are in line with the results observed in Golestan Province based on the NSFWQI index (63). All the studies conducted on rivers receiving the effluent of fish farms in East Azerbaijan Province, Golestan Province and Gorgan University based on the NSFWQI index have proved the effects of fish farms' activities on the quality of rivers by various degrees (23, 66–69).

According to the results, stations 1 and 2, which were located in the upper section of the first fish farm and were guarded against the pollutants of this farm, hosted a higher number of Amphipoda and Ephemeroptera species, which mostly live in freshwaters; their biological index was also excellent in quality. The reason for the higher numbers of Amphipoda and Ephemeroptera in the effluent station of the first fish farm (Fig. 2) is probably because the water was not polluted enough to negatively affect such arthropods. The relative effect of effluent pollutants from the first fish farm on station 4 was significant in a way that the number of the above-mentioned species drastically reduced in this station; Leach species was also detected and collected in this station. An increase in the number of such species in Station 5 shows that the environment for their thriving is quite felicitous. Located in the upper section of the second fish farm, Stations 6 hosted Amphipoda and Chironomidae species (two species which are less sensitive to pollution). Thus, Stations 3 and 6 fit into the 'very good quality' category regarding biological index. The number of Amphipoda species decreased in Station 7 and other later stations, but the number of other less-sensitive species, such as Chironomidae, increased; some species which are frequently observed in polluted waters, such as Diptera and Isopoda, were also observed in Station 9. Therefore, Stations 7 and 8 fall into the 'good quality' category and Station 9 into the 'moderate quality' category. In general, it seems that because of the short-

covering distance of the study, the negative effects of pollutants resulting from the effluent of fish farms were higher in lower stations (70).

The existence of a positive correlation between fecal coliform and dissolved solids is acceptable because an increase in the amount of fecal coliform could increase the load of organic compounds in water, leading in turn to a rise in the electrical conductivity index. The negative correlation between fecal coliform and DO is also acceptable because when the coliform-containing effluent enters the river, the oxygen level drastically decreases. The positive correlations among dissolved solids, turbidity and electrical conductivity result from their mutual effects on each other because when the number of dissolved solids increases, the amounts of turbidity and electrical conductivity of water also increase. The positive correlations among nitrate, electrical conductivity, phosphate, BOD and COD parameters are a result of deriving from the same source in a way that an increase in the amount of nitrate can raise the amounts of other mentioned parameters (71).

The investigation of the effects of physiochemical parameters and Hilsenhoff biological Index on other parameters of the study by the linear regression analysis revealed that there were significant relationships between these two Indices and pH, DO and dissolved solids parameters. These results agree with the findings of the studies conducted in Nigeria in terms of pH parameter, in Mazandaran Province and Lorestan Province in terms of dissolved solids parameter and finally in Nigeria, Mazandaran Province and Lorestan Province in terms of DO (36–38). This effect was positive regarding dissolved solids parameter (Sig= 0.046, B= 0.027) but negative regarding DO parameter (Sig= 0.037, B= -1.588) and pH parameter (Sig= 0.029, B= -4.837). The quantity and quality of the influent organic materials pouring into water resources (72) are influenced by fish farm activities and the effluent discharge of such farms (52, 71). This can pol-

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lute waters and negatively affect the quality and quantity of water resources (66, 72, 73), and effective parameters can bring about drastic changes in the ecosystem, such as losing diversity, losing the whole population of one species, increasing the number of species feeding on organic materials, losing sensitive species and replacing resistant species (72). Finally, such changes can result in an increase or decrease in the biological index as observed in the stations in this study. The relationship between effective parameters and the Hilsenhoff biological Index is represented in Fig. 2. As observed, there were no significant relationships between the Hilsenhoff biological Index and the two qualitative Indices of NSFWQI and IRWQI showing that the measured parameters are not similar in the above-mentioned Indices.

To conduct this study, there were some limitations that faced challenges, including economic limitations and problems related to sampling. These problems were solved by conducting composite sampling and managing points and the number of samples so that the resulting data have the maximum compatibility with the real conditions of the studied area. Also, there were problems in the analysis of some parameters, which were solved by using portable devices.

## Conclusion

The stations located in the upper section of fish farms can be categorized into the 'good quality' category based on the NSFWQI and IRWQI$_{SC}$ Indices. The activities of fish farms can negatively impact relevant parameters that affect the NSFWQI index and its lower values in the effluent receiving stations and even in lower-section stations, hence putting these stations in the 'moderate quality' category. However, in terms of the IRWQI$_{SC}$ index, the effluent stations and the lower ones all belong to 'fairly bad quality' and 'moderate quality'

categories. The main reason for the changes in the above indicators along the river and especially in the downstream stations of the farms is related to receiving effluents from the farms and the presence of relatively high amounts of various pollutants based on the results of the tests. The two water quality indicators generally had almost the same trends in all stations and minor changes are caused by the parameters including BOD, nitrate, phosphate and etc. in their calculation. Therefore, it is necessary to consider the necessary arrangements for different uses of water in downstream of the river and act with caution for drinking and household uses. The linear regression analysis revealed a significant relationship between the Hilsenhoff biological Index and the physiochemical parameters of pH, DO and total dissolved solids. Given that the activity of fish farms and their effluent discharge can negatively change the physical, chemical and biological parameters of receiving waters, it is essential to consider necessary treatments for such effluents so that health and ecological risks are efficiently reduced. For purposes such as drinking, it is recommended to use different water treatment methods, including observing the physical distance of water harvesting for the possibility of self-purification, physicochemical treatment and disinfection, as much as possible depending on the local conditions, so that the quality of water could be acceptable in terms of national standards for various uses as well as surface water. Moreover, it is of high importance to conducting more studies in this field, continuous monitoring and measure Indices relevant to the type of utilization and check them against with the standards.

## Acknowledgements

and also to the experts working at Water and Wastewater Laboratory of School of Health who cooperated in conducting relevant experiments.

## Ethical considerations

## Conflict of interest statement

The authors declare there is no conflict of interest.

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