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Effects of cage culture of rainbow trout, *Oncorhynchus mykiss* on phytoplankton and zooplankton communities (Case study: Golestan Reservoir 1, Gorgan, Iran)

Javid Imanpour Namin[*1], Kam Safarbibi[1], Hamid Allaf Noveirian[1], Korosh Amini[2]

1. Fisheries Department, Faculty of Natural Resources, University of Guilan, Sowmeh-Sara, 1144, Guilan, Iran
2. Inland Water Aquatics Stocks Research Center, Gorgan, Iran

** Corresponding author's E-mail: imanpour@guilan.ac.ir, javidiman@gmail.com*

ABSTRACT

The environmental effects of fish cage culture have poorly been studied in reservoirs. Since this activity is increasingly practiced, investigations on the impacts of cage culture on physicochemical parameters and biodiversity of the reservoir is imperative. The aim of this study was to study the effects of rainbow trout cage culture on water quality, as well as plankton population structure and communities in a reservoir (Golestan Reservoir 1, Gorgan, Iran). Four sampling stations were selected in the following manner: sampling station immediately below the cages, stations some distances including 50 m, 150 m and 1000 m (control station) away from the cages. Phytoplankton, zooplankton and water samples were obtained on a monthly basis for 4 months (December, January, February and March 2016). The depths of the stations were the same. Water temperature, pH, dissolved oxygen, EC, transparency, TDS and TSS did not exhibit statistically significant differences among the stations. Significant increases were detected in the ammonium, nitrate, phosphate and chlorophyll *α* concentrations at the cage station ($P<0.05$). Five phytoplankton phyla including Bacillariophyta, Chlorophyta, Cyanophyta, Euglenophyta and Dinophyta and also four zooplankton phyla including Protozoa, Rotatoria, Cladocera and Copepoda were identified in the reservoir. The obtained results revealed that cage culture exerts significant effect on the population and communities of planktonic organisms. Increase

Keywords: Golestan, Reservoir, Net cages, *Oncorhynchus mykiss*, Phytoplankton, Zooplankton.
Article type: Research Article.

INTRODUCTION

Cage culture is the practice of farming of aquatic organisms in cages and nets, commonly practiced worldwide in both freshwater and marine environments, including open ocean, estuaries, lakes, reservoirs, ponds and rivers (Beveridge 1987; Huang *et al*. 2012). Cage aquaculture is an old practice. It dates back to early 10th century when Chinese fishermen used to grow fish fry in cages made of bamboo sticks (Beveridge 1996; Degefu *et al.* 2011). Cage aquaculture has certain advantages over other aquaculture systems that are potentially important in terms of uptake by rural poor and landless people; use of existing ponds, lakes and reservoirs that are currently not utilized; ease of feeding; ease of stocking and harvesting; less expense associated with treating or preventing disease; easier stock management and monitoring compared with pond culture (Mondal *et al*. 2010; Onyema 2011: Oniye *et al*. 2014). Nonetheless, numerous concerns have been raised about the environmental impact of cage culture, mainly on the water quality and biotic composition of small water bodies such as fish ponds and reservoirs (Degefu, Mengistu & Schager 2011). According to Cowx, Grady, Welcomme & Bartley (1998) and Borges *et al.* (2010), fish production in artificial reservoirs has the potential to significantly contribute to the global fish supply, especially in Asia (De Silva 2002) and South America (Petrere 1996). Since the exchange time of freshwater

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systems is shorter than that in marine environments, the environmental effects of wastes produced by freshwater cage fish culture are much stronger than those of marine cage farming (Alpaslan & Pulatsü 2008). Cage cultured fish are entirely dependent on formulated diets. A relatively small portion of the organic matter and inorganic nutrients in feed applied to cages is transformed to fish biomass. It is estimated that for every ton of fish production in cage culture, 132.5 kg of nitrogen and 25.0 kg of phosphorus are released into the environment (Islam 2005; Anusuya *et al*. 2015). Nutrients such as phosphorus, and nitrogen, as well as other chemical residues are released into the water column from the breakdown of excess feed, as well as through fish excretion and fecal waste often leading to nutrient loading, and eutrophication. Environmental impacts are not restricted to areas within cages, as wind, waves and bottom currents can also allow nearby areas to be affected by farming activities. Phytoplankton and zooplankton are good indicators for changes in nutrient pollution over time because they respond quickly to changes in nutrient. The aim of this study was to evaluate the effects of a fish (*Onchorhynchus mykiss*) cage farm on water quality and phytoplankton and zooplankton structure in the Golestan Reservoir 1. Since this is the first research on the impacts of fish cage culture in a reservoir in Iran, it is an important contribution to the future development of similar farms.

MATERIALS AND METHODS

Study site

The Golestan Reservoir 1 (Fig. 1) is located 13 km from Gonbad Kavous at 37˚ 19′ longitude and 55° 17′ latitude. Its basin covers an area of 5,000 square kilometers and is located between 57° 36′ North longitude to 46° 37′ and 13° 55′ and 28° 55′ East latitude. Gorganrood River, the main river in the region, supplies the reservoir. The volume of the reservoir is 86 million cubic meters with surface area of 1500 ha. The reservoir is a storage type dam with maximum depth of 25 m. The reservoir provides services to agriculture, industry, aquaculture sectors and acts as flood control infrastructure.

Sampling

The study was carried out in a rainbow trout cage culture farm located at the Golestan Reservoir 1 (Fig. 1) as a part of Gorganrood River basin during 2016. The studied fish farm was composed of 40 cages (150 m^3 volume each) spread over an area of approximately 2 km^2 with a total production capacity of 32 tons of rainbow trout, *Oncorhynchus mykiss*, per year. The cages were made of a steel frame and polyethylene net with mesh size of 16 mm.

Rainbow trout were stocked in cages in December 2016. Feeding was performed with extruded commercial food. The fish with a mean size of 400 ± 5 g were introduced into the cages and harvested at 900 g ± 10 g by the end of four months. Four stations were selected in the reservoir as follows: immediately below the cages (St_1), and also some distances including 50 m (St_2), 150 m (St_3) and 1000 m (St_4 = control station) far from the cages. The depth of the stations was 7 m. The first sampling was carried out one week before the cage installation. Dissolved oxygen (DO), pH, specific conductivity, TDS and water temperature were measured using a multi-parameter probe (Model HQ40d, HACH Instruments) in-situ during sampling events. Water transparency (vertical visibility) was estimated using a standard Secchi disc of 20 cm diameter and was measured in-situ during sampling as well. The replicated water samples were collected using Ruttner sampler on a monthly interval at the depth of 1 m. Water samples (2 L) were filtered through Whatman GF/C filter papers. Chlorophyll-α concentration was analyzed spectrophotometrically after extraction with 90.0% acetone (APHA 1998). Nitrate, phosphate and ammonium were measured using a photometer (Palintest 8000, Gateshead, UK) following standard method described for the examination of water and wastewater (APHA 1998). Plankton samples were taken monthly by Ruttner sampler and were filtered by a 20 µ plankton net for qualitative and quantitative analyses. Samplings were carried out at subsurface during the morning. The water samples (5 mL) were sedimented after preservation with 4% formalin solution in counting chambers. Phytoplankton counting followed the standard inverted microscope method as described by Lund *et al.* (1958). Colonies and filamentous algae were counted as an organism (APHA 1998). In the case of zooplankton counting, the water samples were preserved in a 4% formaldehyde solution and allowed to be settled down in a chamber (Wetzel & Likens 1991), then counted and identified (Edmondson 1959; Harding & Smith 1974).

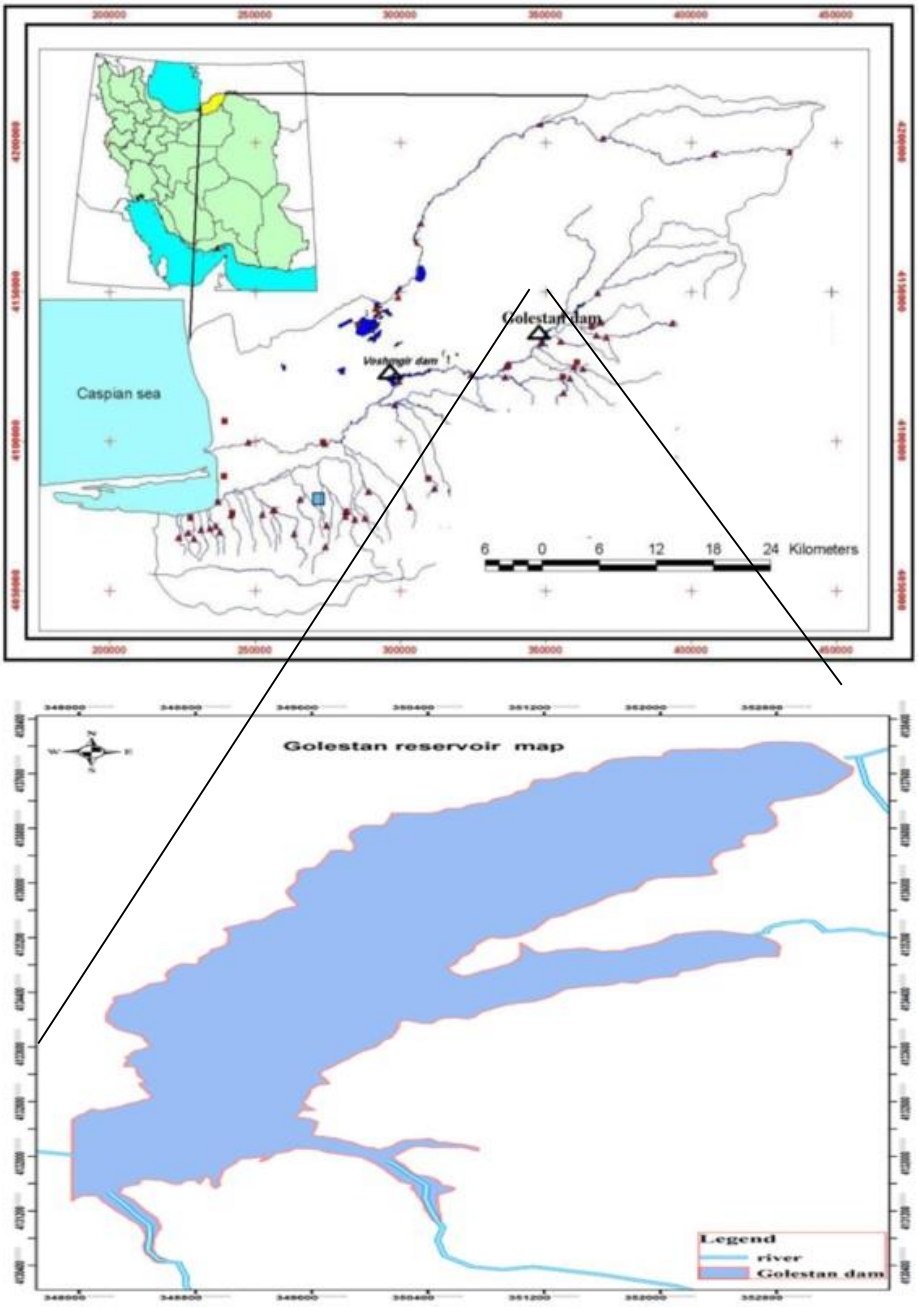

Fig.1. Location of the Golestan Reservoir 1

Plankton density was calculated in cubic meters (APHA 1998). To obtain the weight of the living organisms, their lengths were measured and calculated using their geometric shapes (Lawrence *et al.* 1987). The total biomass (mg m^{-3}) was determined for each taxon using length–weight regressions from Amerasinghe *et al*. (2008), Dumont *et al.* (1975), Claps *et al*. (2004) and also Maia-Barbosa & Pinto-Coelho (2013). The Shannon index $H = -\sum[(p_i) \times \log(pi)]$ was used to calculate the species diversity (Shannon & Weaver 1963). The index uses the number of species and distribution of species and their relationship, where H is the index of species diversity and P_i is the ratio of the number of species to the total number of the species. The Shannon index value varies between 0-6. Greater Shannon index shows great diversity of species in the ecosystem.

Data Analysis

Statistical analyses were performed using Excel 2016 and statistical software SPSS version 16. Two-way variance analysis (ANOVA) and Tukey post hoc test were used to evaluate differences in water quality, phytoplankton and zooplankton between different sampling stations and months. The Shannon diversity was calculated using Ecological Methodology software. Principal Components Analysis (PCA) was used to reduce the data

dimensionality and identify the main variables influencing the structure of the plankton community. The PCA was performed using Canoco 5.0.

RESULTS

The parameters corresponding to the physical and chemical variables (mean ± SE) are presented in Table 1. The mean water temperature at the sampling sites varied between 12.0 and 18.16 ºC. Transparency varied between 5.04 m (December) and 5.54 m (January), the highest values were in station 4 while the lowest at St_1.

The lowest dissolved oxygen was 8.03 ± 0.18 mg L^{-1} at St_1 in January, while the highest value, 8.46 ± 0.06 mg L^{-1} in February at St_3. The pH was lower in January than in the other months. Electrical conductivity (EC) of the water varied between 558- 678 µm cm^{-1}. Total dissolved solid (TDS) slightly varied in the range of 144 -152 mg L^{-1} and total suspended solid (TSS) values fluctuated between 3-4.66 mg L^{-1}. There were no statistically significant differences in these parameters among sampling stations ($p > 0.05$).

The lowest concentration of Chlorophyll α was observed at St_4 (March) while the highest at station 1 (February). The chlorophyll α concentration showed statistically significant differences in all sampling months ($p < 0.05$) except for November. The values of nitrate, phosphate and ammonium were also analyzed and compared between stations. These values were higher at St_1 in comparison with the others stations and significant differences were evident ($p < 0.05$) for nitrate, phosphate and ammonium except for November. In this study five phytoplankton phyla including Bacillariophyta (7 species), Chlorophyta (6 species) Cyanophyta (5 species), Euglenophyta (2 species) and Dinophyta (1 species) were identified (Table 2). Holoplanktic zooplankton including Protozoa (4 species), Rotatoria (8 species), Cladocera (5 species) and Copepoda (4 species) were also identified (Table 3).

The highest densities of phytoplankton were recorded in February at St_1 with a mean number of 9.8×10^7 ± 1509900 cells per cubic meters at the actual location of the cages. The highest concentration of phytoplankton in the rest of sampling months at stations 2, 3 and 4, were 8.52×10^7 ± 1587400, 6.9×10^7 ± 1216500 and 4.8×10^7 ± 69280 cells m^{-3} respectively, recorded in February. The highest phytoplankton concentration was observed at St_1 during the whole sampling period followed by stations 2, 3 and 4 (Fig. 2). Tukey post hoc test showed statistically significant differences ($p < 0.05$) in phytoplankton density between the station immediately below the cage and control stations except in November.

In November, December, January, February and March, Bacillariophyceae was dominant at all stations (Fig. 3). The percentages of diatoms were 38.05%, 38.43%, 36.6%, 33.32% and 44.56% of the total phytoplankton density in November, December, January, February and March respectively.

The analysis of ANOVA revealed significant ++differences ($P < 0.05$) in density and biomass of phytoplankton at different sampling stations and months. Shannon index for phytoplankton groups varied between 3.77- 4.03 (Fig. 4). There were no significant differences in terms of biodiversity at different stations ($P > 0.05$).

The results depicted in Fig. 5 exhibited that the density and biomass of zooplankton in November, December, January, February and March at St_1 was higher than those at stations 2, 3 and 4, reflecting the effects of cage fish farming. The highest density of zooplankton during the study was recorded at St_1 with an average of 5.2×10^4 ± 11269 cells m^{-3} in December.

This sampling station showed the highest density in all other monthly samplings (Fig. 5). In November, December, January, February and March, Rotatoria was the dominant taxa in all stations (Fig. 6). The percentages of Rotatoria were 36.83%, 42.17%, 56.48%, 41.49% and 48.57% of the total zooplankton composition in November, December, January, February and March respectively.

There was a significant difference between the density and biomass of zooplankton in different months ($p < 0.05$). Shannon index for the groups of zooplankton varied from 3.14-3.93 (Fig. 7). No significant difference was observed in terms of biodiversity at different stations ($p > 0.05$).

Table 1. Results of physical and chemical analyses at the studied sampling stations, in Golestan Reservoir One from November to March (2016-2017) *

Physical and Chemical Parameters/month/station		Water temperature (°C)	DO (mg L^{-1})	pH	Transparency (m)	Conductivity (µs cm^{-1})	TDS (mg L^{-1})	TSS (mg L^{-1})	Chl. α (µg L^{-1})	PO_4^- (mg L^{-1})	NO_3^- (mg L^{-1})	NH_4^+ (mg L^{-1})
November	1	18.16 ± 0.6	8.26 ± 0.17	7.6 ± 0.05	5.4 ± 0.03	596.33 ± 3.3	152 ± 5	3 ± 1.00	2.45 ± 0.05	0.03 ± 0.002	0.23 ± 0.01	0.021 ± 0.001
	2	17.73 ± 0.37	8.3 ± 0.2	7.54 ± 0.04	5.43 ± 0.01	593 ± 1.52	148 ± 5.4	3.33 ± 1.5	2.4 ± 0.045	0.026 ± 0.001	0.23 ± 0.01	0.22 ± 0.004
	3	17.96 ± 0.37	8.1 ± 0.2	7.49 ± 0.02	5.44 ± 0.06	590 ± 2.3	1.49 ± 5.5	3 ± 1.00	2.43 ± 0.02	0.028 ± 0.001	0.23 ± 0.01	0.021 ± 0.003
	4	17.9 ± 0.49	8.16 ± 0.13	7.53 ± 0.02	5.41 ± 0.04	593.67 ± 2.72	147 ± 4.04	3 ± 1.00	2.36 ± 0.06	0.028 ± 0.006	0.23 ± 0.017	0.021 ± 0.003
December	1	16.33 ± 0.33	8.16 ± 0. 8	7.6 ± 0.01	5.04 ± 0.01	627.67 ± 17.3	147 ± 0.88	4.33 ± 0.66	2.83 ± 0.08^{a}	0.055 ± 0.002^{a}	0.32 ± 0.01^{a}	0.025 ± 0.004
	2	16.3 ± 0.26	8.2 ± 0.11	7.5 ± 0.03	25.11 ± 0.02	649.67 ± 1.45	144 ± 1.45	4 ± 0.00	2.65 ± 0.12^{ab}	0.052 ± 0.002^{a}	0.32 ± 0.00^{a}	0.024 ± 0.003
	3	16.7 ± 0.17	8.16 ± 0.08	7.5 ± 0.03	95.12 ± 0.03	608.33 ± 30.6	143 ± 2.33	3.66 ± 0.33	2.52 ± 0.09^{ab}	0.05 ± 0.001^{a}	0.31 ± 0.00^{a}	0.023 ± 0.002
	4	16.16 ± 0.16	8.26 ± 0.26	7.5 ± 0.05	5.12 ± 0.03	643.67 ± 3.17	144 ± 6.92	3.33 ± 0.33	2.38 ± 0.055^{b}	0.03 ± 0.003^{b}	0.22 ± 0.08^{b}	0.02 ± 0.001
January	1	14.66 ± 0.33	8.03 ± 0.18	7.48 ± 0.33	5.32 ± 0.15	647.67 ± 27.3	146 ± 1.33	4.66 ± 0.33	2.98 ± 0.069^{a}	0.059 ± 0.002^{a}	0.38 ± 0.00^{a}	0.029 ± 0.001^{a}
	2	14.96 ± 0.3	8.43 ± 0.12	7.34 ± 0.14	5.44 ± 0.14	672.33 ± 3.38	145 ± 2.51	4.33 ± 0.33	2.81 ± 0.032^{a}	0.052 ± 0.002^{b}	0.35 ± 0.017^{a}	0.025 ± 0.002^{ab}
	3	14.83 ± 0.16	8.43 ± 0.12	7.34 ± 0.19	5.47 ± 0.077	680 ± 1.52	145 ± 1.73	4 ± 0.57	2.51 ± 0.062^{b}	0.049 ± 0.002^{b}	0.32 ± 0.02^{a}	0.026 ± 0.003^{ab}
	4	14.83 ± 0.44	8.3 ± 0.06	7.27 ± 0.03	5.5 ± 0.04	678.33 ± 2.40	144 ± 2.18	3.33 ± 0.33	2.31 ± 0.03^{b}	0.031 ± 0.002^{c}	0.22 ± 0.01^{b}	0.021 ± 0.001^{b}
February	1	13.16 ± 0.16	8.43 ± 0.05	7.5 ± 0.14	5.23 ± 0.088	558 ± 2.00	147 ± 0.57	4.66 ± 1.2	2.92 ± 0.04^{a}	0.057 ± 0.003^{a}	0.43 ± 0.00^{a}	0.03 ± 0.004^{a}
	2	13.23 ± 0.12	8.4 ± 0.14	7.36 ± 0.23	5.3 ± 0.11	565.33 ± 2.9	145 ± 1.2	4 ± 0.57	2.85 ± 0.05^{ab}	0.058 ± 0.001^{b}	0.39 ± 0.00^{ab}	0.028 ± 0.002^{ab}
	3	13 ± 0.01	8.46 ± 0.06	7.39 ± 0.06	5.43 ± 0.02	564 ± 4.1	144 ± 1.73	3.66 ± 0.33	2.61 ± 0.09 ^{ab}	0.046 ± 0.001^{b}	0.34 ± 0.02^{b}	0.026 ± 0.001^{ab}
	4	13.23 ± 0.23	8.4 ± 0.06	7.25 ± 0.07	5.43 ± 0.02	559.67 ± 5.54	144 ± 1.76	3 ± 0.00	2.34 ± 0.05^{b}	0.031 ± 0.002^{c}	0.23 ± 0.00^{c}	0.021 ± 0.001^{b}
March	1	12.16 ± 0.16	8.23 ± 0.23	7.7 ± 0.11	5.27 ± 0.06	564 ± 3.00	147 ± 0.66	4.33 ± 0.66	3.14 ± 0.06^{a}	0.058 ± 0.001^{a}	0.46 ± 0.01^{a}	0.03 ± 0.001^{a}
	2	12 ± 0.01	8.33 ± 0.06	7.4 ± 0.15	5.34 ± 0.02	562.67 ± 2.1	145 ± 2.02	3.66 ± 0.33	2.71 ± 0.07 ^{b}	0.046 ± 0.002^{b}	0.4 ± 0.01^{a}	0.029 ± 0.001^{a}
	3	12.2 ± 0.01	8.4 ± 0.23	7.3 ± 0.08	5.37 ± 0.03	560.33 ± 2.02	145 ± 1.2	3.66 ± 0.33	2.31 ± 0.07^{c}	0.044 ± 0.004^{b}	0.31 ± 0.01^{b}	0.027 ± 0.002^{a}
	4	12 ± 0.01	8.4 ± 0.4	7.3 ± 0.17	5.37 ± 0.02	561.33 ± 1.7	144 ± 2.18	3.66 ± 0.66	2.17 ± 0.02^{c}	0.031 ± 0.002^{c}	0.23 ± 0.01^{c}	0.021 ± 0.001^{b}

*. Values are presented as mean (± SE). Differences between means with the same superscripts in a row are not significant ($p>0.05$).

Table 2. Presence (+) or absence (-) of the phytoplankton in Golestan Reservoir 1 in the different stations and months.

Phytoplankton Taxa	Abbreviation	November				December				January				February				March			
		1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
Bacillariophyceae																					
Asterionella sp.	Asterio	-	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+
Coscinodiscus sp.	Coscino	-	-	-	-	-	-	-	-		+	-	-	+	+	+	+	+	+	+	+
Cyclotella sp.	Cyclote	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Cymbella sp.	Cymbe	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Fragilaria sp.	Frag	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Navicula sp.	Navi	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Nitzschia sp.	Nitzs	+	+	+	+	+	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-
Chlorophycaea																					
Ankistrodesmus sp.	Ankis	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Chlorella sp.	Chlore	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	
Closteriopsis sp.	Closterio	-	-	-	-	-	-	-	-	+	+	-	+	+	+	+	+	+	+	+	+
Closterium sp.	Closteri	+	+	+	+	+	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-
Cosmarium sp.	Cosmar	-	-	-	-	-	-	-	-	+	-	+	+	+	+	+	+	+	+	+	+
Eudorina sp.	Eudor	-	-	-	-	-	-	-	-	+	+	-	+	+	+	+	+	+	+	+	+
Cyanophyceae																					
Anabaena sp.	Anaba	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Anabaenopsis sp.	Anabaeno	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Oscillatoria sp.	Oscilla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Spirulina sp.	Spirul	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Gomphosphaeria sp.	Gompho	+	+	+	+	+	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
Euglenophyceae																					
Euglena sp.	Eugl	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Trachelomonas sp.	Trach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Dinophyceae																					
Gymnodinium sp.	Gymno																				

Table 3. Presence (+) or Absence (-) of the zooplankton in Golestan Reservoir 1 in the different stations and months.

Zooplankton Taxa	Abbreviation	November				December				January				February				March			
		1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
Protozoa																					
Difflugia sp.	Difflug	+	-	+	+	+	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-
Paramecium sp.	Parame	+	-	+	+	+	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-
Raphidophrys sp.	Raphido	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+
Rotatoria																					
Adineta sp.	Adinet	+	-	+	+	+	-	+	+	+	+	+	+	+	+	-	+	+	+	+	-
Asplanchna sp.	Asplanch	+	-	+	+	+	+	+	+	+	+	-	+	+	+	+	+	-	+	+	+
Brachionus sp.	Brachio	+	+	-	+	+	-	+	+	+	+	+	+	+	+	+	+	+	-	-	+
Keratella sp.	Kerate	+	+	+	-	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+
Philodina sp.	Philod	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pleurotrocha sp.	Pleuro	-	-	-	-	-	-	-	-	+	+	+	+	+	-	+	+	+	+	+	+
Rotaria sp.	Rotaria sp.	+	+	+	-	+	+	+	+	+	+	+	+	+	+	-	+	+	-	+	+
Trichotria sp.	Trichot	-	+	+	+	+	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-
Cladocera																					
Daphnia sp.	Daphni	+	+	+	-	+	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-
Nauplius Cladocera	Naup Clado	+	+	-	+	+	+	-	+	+	-	+	+	+	-	+	+	+	+	+	+
Diaphanosoma sp.	Diaphano	+	+	-	+	+	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-
Leptodora sp.	Leptodo	-	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+
Copepoda																					
Eudiaptomus sp.	Eudiap	+	+	+	+	+	-	+	+	+	-	+	+	+	+	+	+	+	+	+	+
Cyclops sp.	Cyclop	-	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	-	+	+
Eucyclops sp.	Eucyclo	+	-	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+
Nauplius copepoda	Nau cope	-	+	+	+	+	+	-	+	+	-	-	-	-	-	-	-	-	-	-	-
Thermocyclops sp.	Thermoc	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+

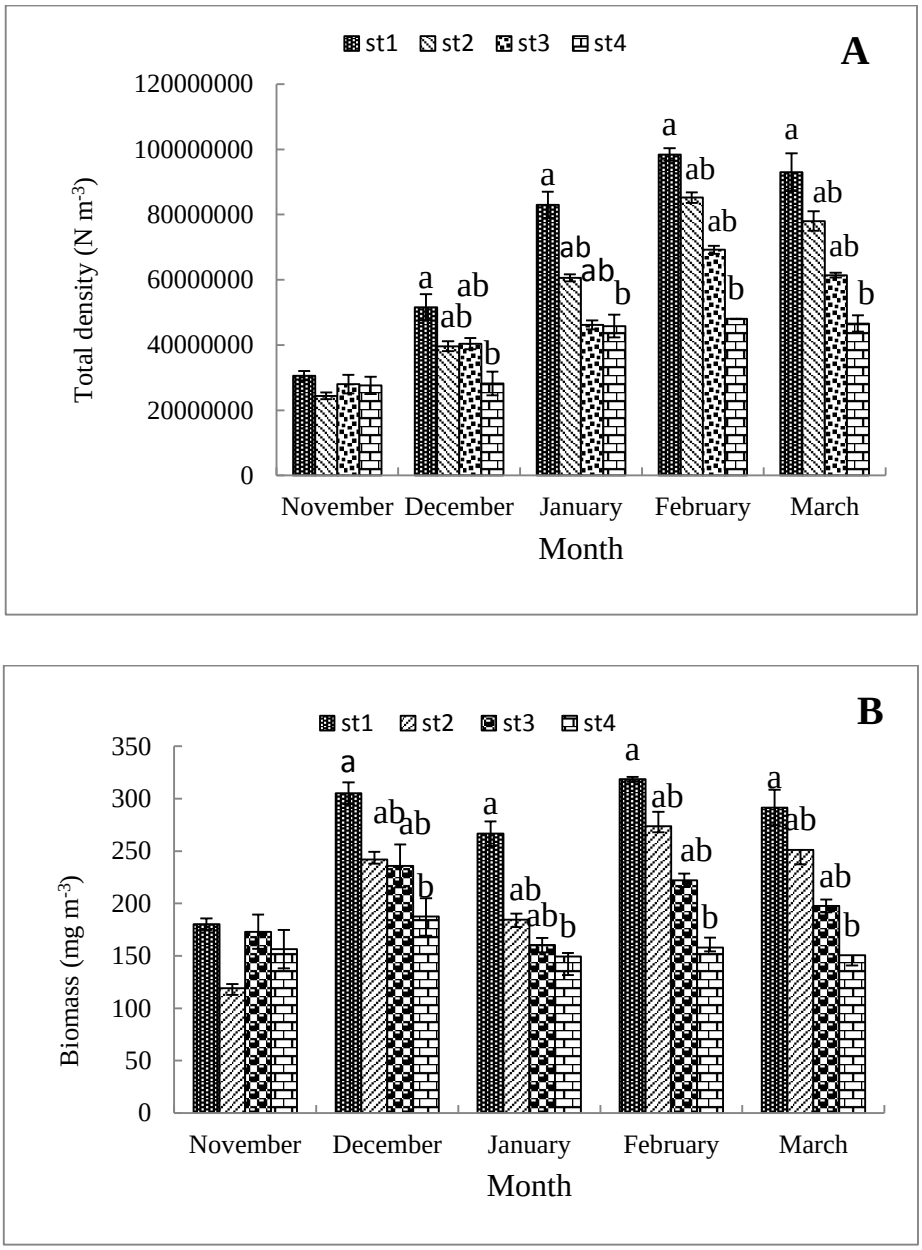

Fig. 2. Mean (± SE) density (A) and biomass (B) of phytoplankton in different months and stations of Golestan Reservoir 1 in 2016-2017. Different letter subscripts represent significant different among the stations at the level of $p < 0.05$.

PCA showed highly significant scores (P = 0.001) for axis 1 and all canonical axes, thus the ordination results are authentic for phytoplankton and zooplankton. These 10 environmental variables in the PCA explained 81.4% of the total variation in the phytoplankton communities. The eigenvalues of axis 1 and 2 were 0.815 and 0.086, respectively, revealing 81.1% and 89.74% of the total variance, respectively. The species-environment correlations were 0.988 for axis 1 and 0.754 for axis 2, indicating a significant relationship between the environmental variables and dominant species. For zooplankton community, these 10 environmental variables in the PCA explained 78.7% of the total variations. The eigenvalues of axis 1 and 2 were 0.639 and 0.118, respectively, exhibiting 63.92% and 75.73% of the total variance, respectively. The species-environment correlations were 0.986 for axis 1 and 0.862 for axis 2, displaying a significant relationship between the environmental variables and dominant species.

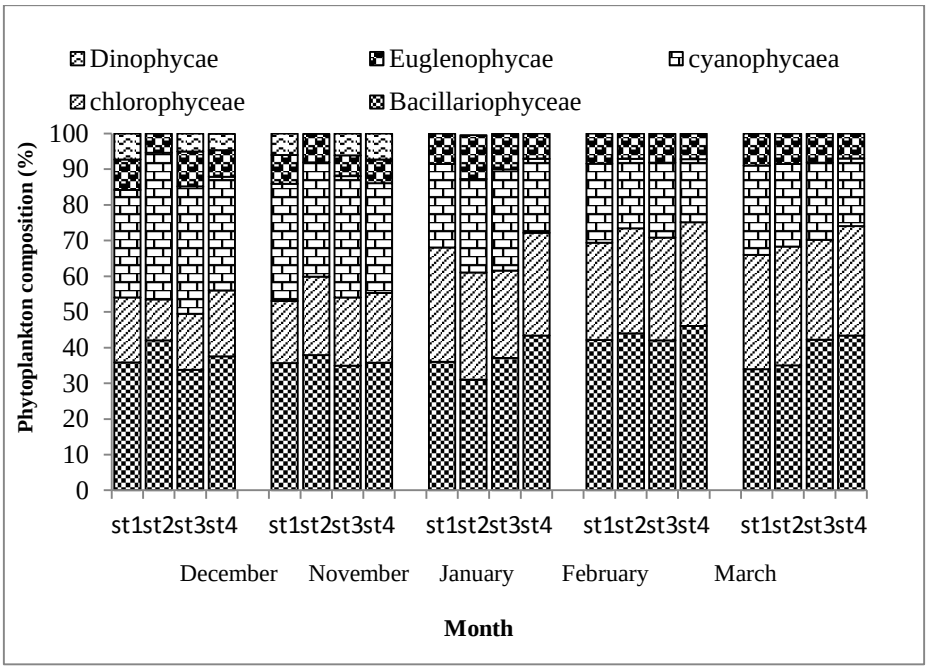

Fig. 3. Phytoplankton composition in Golestan Reservoir 1 in different months and stations during 2016-2017.

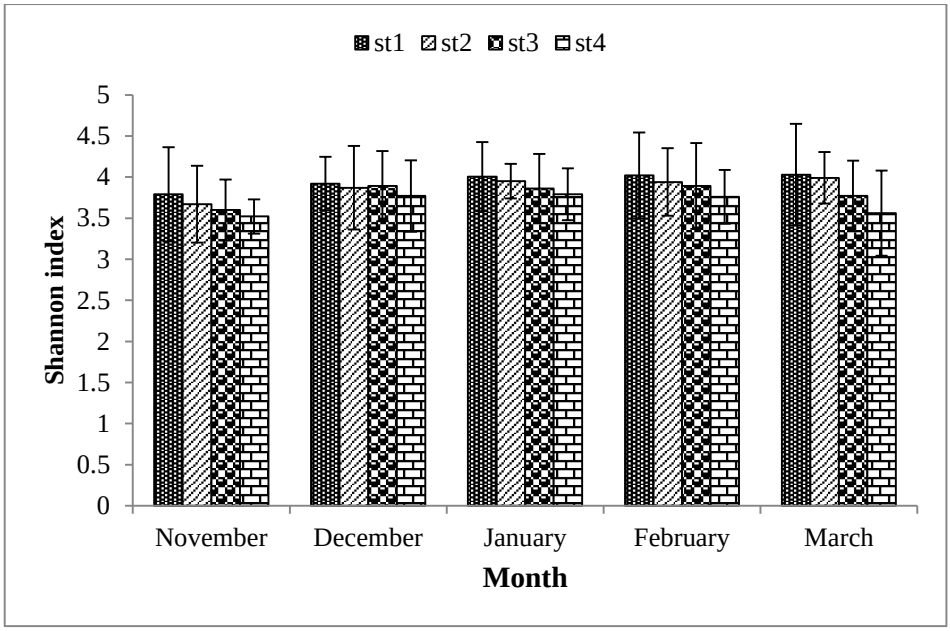

Fig. 4. Mean (±SE) Shannon diversity index values of phytoplankton in Golestan Reservoir 1 in different months and stations during 2016-2017.

Some parameters like temperature, EC and pH showed significant positive association with phytoplankton species of *Gymnodinium* sp., *Gomphosphaeria* sp., *Closterium* sp. and *Nitzchia* sp. while a negative relationship with transparency and DO. A significant positive relationship was recorded between *Eudorina* sp., *Asterionella* sp., *Coscinodiscus* sp. and *Cosmarium* sp. with dissolved oxygen (DO) and transparency, while a negative relationship with water temperature, EC and pH. Some species like *Ankistrodesmus* sp., *Navicula* sp., *Euglena* sp., *Cyclotella* sp., *Trachelomonas* sp., *Spirulina* sp., *Cymbella* sp., *Oscillatoria* sp., *Anabaena* sp., *Anabenopsis* sp., *Closterioplsis* sp., *Chlorella* sp. and *Fragillaria* sp. displayed a significant positive relationship with water TDS, TSS, nitrate, phosphate and ammonium (Fig. 8).

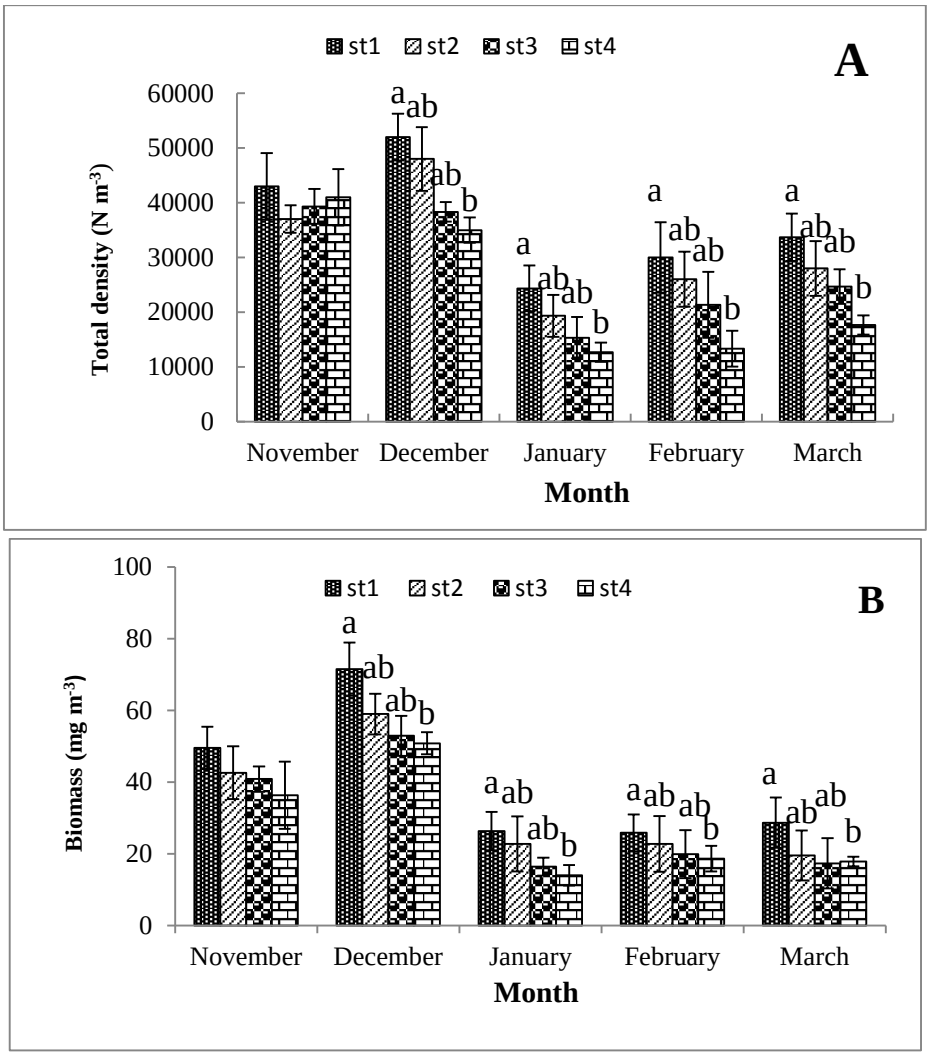

Fig. 5. Mean (±SE) density (A) and biomass (B) of zooplankton in different months and stations of Golestan Reservoir 1 in 2016-2017**.** Different letters represent significant difference among the stations at the level of $p < 0.05$.

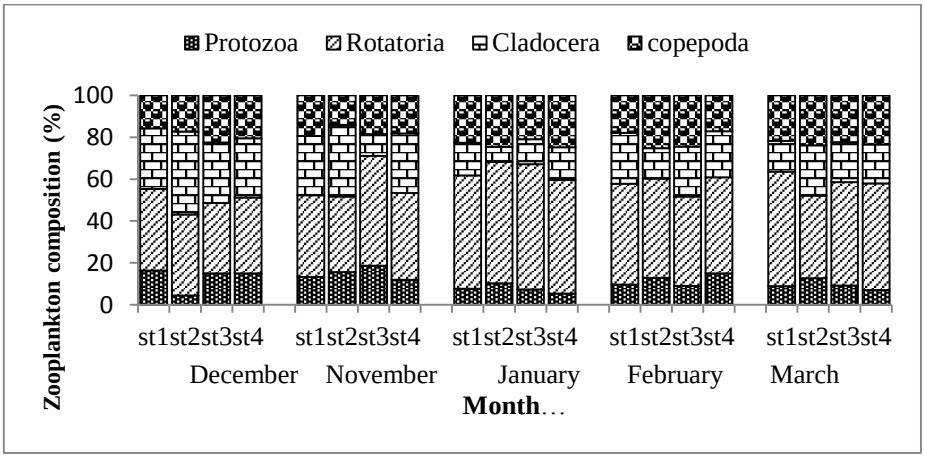

Fig. 6. Zooplankton composition in different months and stations of Golestan Reservoir 1 during 2016-2017.

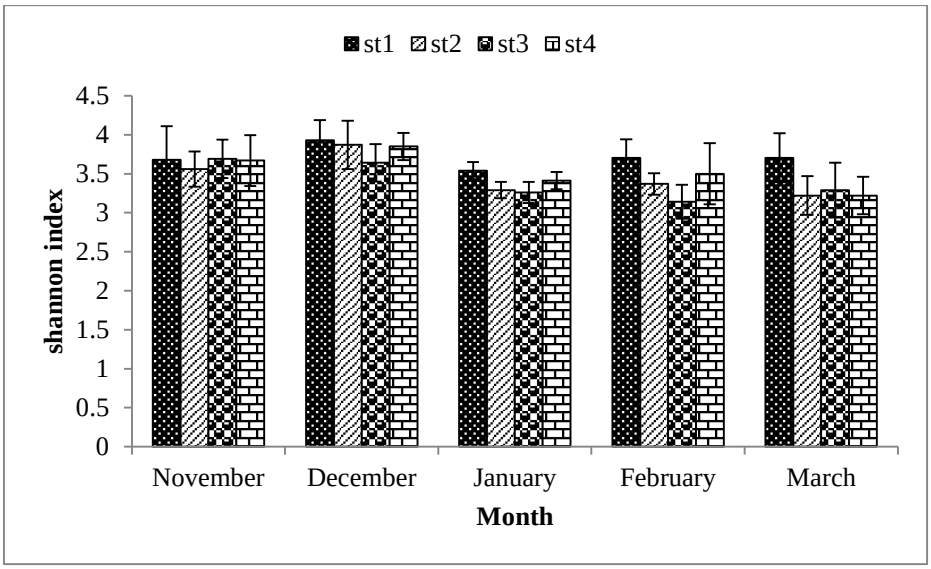

Fig. 7. Mean (± SE) Shannon diversity index values of zooplankton in different months and stations of Golestan Reservoir 1 during 2016-2017.

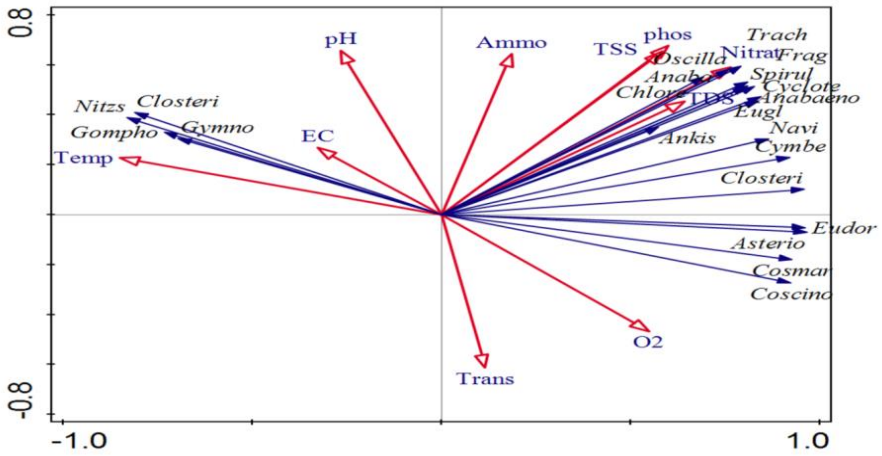

Fig. 8. PCA bi-plot showing the relationships between different physicochemical parameters and phytoplankton species associated with the trout cage culture in Golestan Reservoir 1.

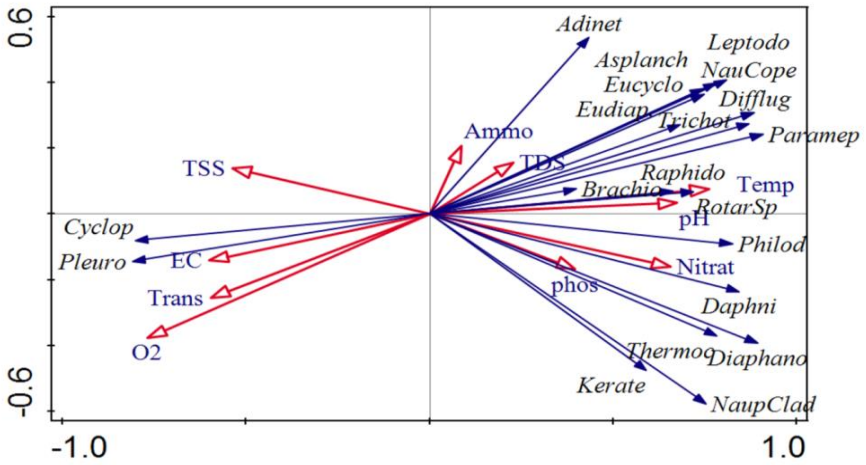

Fig. 9. PCA bi-plot showing the relationships between different physicochemical parameters and zooplankton species concerning to the cage culture in Golestan Reservoir 1.

Transparency, EC and DO showed significant correlation with the zooplankton species including *Cyclops* sp., *Pleurotrocha* sp. while a negative relationship with TDS, pH, temperature and ammonium. *Keratella* sp., *Thermocyclops* sp., *Daphnia* sp., *Philodina* sp., *Diaphanosoma* sp. and Nauplii of Cladocera demonstrated significant positive relationship with nitrate and phosphate, while a negative relationship with TSS. Some species including Rotifera sp., *Brachionus* sp., *Paramecium* sp., *Trichotria* sp., *Difflugia* sp., *Asplanchna* sp., *Leptodora* sp., *Eucyclops* sp., *Adineta* sp., *Paramecium* sp., nauplii of copepoda, *Raphidophrys* sp. showed significant positive relationship with TDS, Temperature, pH and ammonium, while a negative relationship with EC, DO and Transparency. Figs. 8 and 9 illustrate that the temperature and DO were the main variables associated with the distribution of plankton communities.

DISCUSSION AND CONCLUSION

The present study showed that the rainbow trout cage culture farm (with 30-ton capacity) in the Golestan Reservoir 1 has significantly affected water quality, as well as phytoplankton and zooplankton distributions in several ways. Water temperature variation between stations was not statistically significant and apparently cage culture did not have any measurable effect on water temperature which should normally be below 20 °C for rainbow trout culture (Atay 2000; Alpbaz 2005). The lowest values of dissolved oxygen were measured at station 1, although there was no substantial oxygen depletion in the immediate vicinity of the cages. However, the recorded oxygen reduction in water mass surrounding the cages may be due to the respiration by fish (Cornel & Whoriskey 1993). Its level should be higher than 6.00 mg L^{-1} for rainbow trout culture (Atay 2000, Özdemir *et al.* 2014).

There were no statistically significant differences in pH between stations ($p > 0.05$). Cornel & Whoriskey (1993) reported that pH values were similar in cage station and other stations, indicating that the cage culture did not have any measurable impact on pH values. Other authors have reported that the deposition of waste material in cage culture set up may drop (Beveridge 1984; Pitta *et al*. 1999). The results of the present study indicated that the pH fluctuation in the Golestan Reservoir 1 is within the range suitable for rainbow trout culture. All the nutrients were high in cage station as compared to control station except in November and differences in nutrient levels were statistically significant between stations ($p< 0.05$). Several authors have reported that nutrient levels might be increased by fish cage culture depending on the site and size of farms, water exchange rates and other characteristics of the water body (Phillips *et al*. 1985; Stirling & Dey 1990; Pitta *et al*. 1999).

In this study, the effect of nutrient discharge on dissolved oxygen (DO) was not noticeable. So that, a relatively good dissolved oxygen level and Secchi disk depth of transparency were observed throughout the study period at all four sampling stations. These findings are in line with those of Neofitou & Klaoudatos (2008) who reported no significant impact on DO levels by fish cage culture. The mean Secchi disk transparency and chlorophyll α values (5.33 m and 2.59 µg L^{-1}, respectively) indicated the oligotrophic conditions in the reservoir (OECD 1982). The highest concentration of chlorophyll α was recorded at station 1. There were statistically significant differences in the chlorophyll α level at station 1 with all other stations ($p < 0.05$). The highest density and biomass of phytoplankton and zooplankton were recorded in station 1. Demir *et al*. 2001 reported similar trend in phytoplankton density at the cage station in comparison with open water stations in their study in Bodrum, Turkey. Nutrient load enhancement is one of the most severe negative consequences of fish farming in lakes and reservoirs. Low feeding efficiency, high fish density and feed quantity due to intensive cage farming usually leads to mass loss of nutrients. Massik & Costello (1994) reported that 82% of P (phosphorus) in effluents of salmonid farms was directly bioavailable to phytoplankton species. Thus, salmonid aquaculture represents an important point source of nutrient loads in the lake and reservoir systems, which are in a highly biologically available form. Given this fact, the main concern is that in a long run with ever increased production pressure and capacity, there are tangible potentials for eutrophication in such water bodies including Golestan Reservoir 1. The composition of organisms did not vary significantly in different stations. Generally, organic enrichment leads to an elevation in the abundance of species without alteration in the composition of the species at the first stage. Habitats with advanced degree of eutrophication enter the second stage where alterations in the dominance of species occur with ultimate evolve into changes in the composition of the species. The locale of the net cages is still in the first stage of the alterations predicted to occur as a consequence of eutrophication, and actions are already required to avoid further development in the process of eutrophication and changes in biodiversity. Phytoplankton and zooplankton assemblages at both reference and cage farm stations on the Golestan Reservoir 1 were often dominated by the Bacillariophyceae (diatom) and Rotifera, respectively, in terms of the number of genera and their densities

compared to other taxonomic groups. This is in agreement with Demir *et al*. (2001), who reported a higher abundance of diatoms and Rotifera in an Anatolian dam lake compared to other phytoplankton and zooplankton groups. Rotifer dominance in a reservoir could be related to their opportunistic characteristics (r selected strategy, rapid population growth during short favorable seasons), which allow them to flourish in unstable and dynamic environments. Perhaps these characteristics, combined with low predation pressure due to their small size, grant them a competitive advantage over the other groups (Dumont 1977).

The zooplankton density peak was associated with upraised nutrient concentration in the reservoir, which probably stimulated algal growth. Arcifa (1984) presented evidences exhibiting that predation by invertebrates is also an important factor influencing the structure of the whole zooplankton community. Shannon diversity index based on phytoplankton and zooplankton groups in different months was in its highest in December at station 1 where fish cage is located. This can be due to the elevated loads of nutrients at the site of the cage and provision of conditions for the presence of different species (Penczak *et al*.1982). Sidik *et al.* (2008) and Skejic *et al.* (2011) observed a close diversity of phytoplankton among the fish cage culture and those in the control places. The results of the present study highlighted the effects of trout cage culture on population structures of phytoplankton and zooplankton. Generally, water renewal time is shorter in reservoirs than in natural lakes (but not always the case), and changes in ecosystems such as eutrophication resulting from fish cage culture may be less harmful. Therefore, reservoirs are somehow appropriate water bodies for cage culture.

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