

# **Influence of Physico-Chemical Factors, Zooplankton Species Biodiversity and Seasonal** Abundance in Anzali International Wetland, Iran

#### Delaram Golmarvi<sup>1</sup>, Maryam Fallahi Kapourchali<sup>2\*</sup>, Ali Mashinchian Moradi<sup>1</sup>, Mohammadreza Fatemi<sup>1</sup>, Rezvan Mousavi Nadoshan<sup>1</sup>

<sup>1</sup>Department of Marine Biology, Science and Research Branch, Islamic Azad University, Tehran, Iran <sup>2</sup>Inland water Aquaculture Center, Iranian Fisheries Science Research Institute (IFSRI), Agricultural Research Education and Extension Organization (AREEO), Bandar-e Anzali, Iran

Email: \*m\_fallahi2011@yahoo.com

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Abstract

Anzali International wetland is located in the south coast of the Caspian Sea. Physicochemical analysis and zooplankton survey of the wetland were carried out on monthly basis for the period of one year from January 2012 to December 2013 at 9 different stations of Anzali wetland and its related channel to the Caspian Sea. Water temperature of the wetland followed more or less similar trend as that of air temperature. pH determined alkaline nature of the wetland ranging between 7.05 to 9.47, dissolved oxygen was recorded in the range of 3.36 mg/l to 10.51 mg/l while other parameters recorded were water temperature (10 to 23 C), Nitrate (0.48 to 4.36 mg/l), Total Phosphates (0.15 to 0.67 mg/l), Salinity (220 to 692 mg/l), Electrical Conductivity (235 to 1369 µs/cm), TDS (246 to 1971 mg/l), BOD (2 to 36 mg/l) and COD (4 to 74 mg/l). During study period, total 60 species of zooplankton were identified in four main groups such as Protozoa (22 sp.), Rotifera (29 sp.), Copepoda (5 sp.) and Cladocera (4 sp.). The highest numbers of zooplankton were recorded in summer months and lowest in winter which is the second rainy season of this area. The water body is receiving domestic discharge, agricultural run-off and industrial wastes leading to large amount of nutrient inputs to the ecosystem which indicates the eutrophic statue of the wetland. The aim of present study was to investigate the interrelationship between physicochemical factors and zooplankton population in context of their seasonal abundance.

#### **Keywords**

Physicochemical Factors, Zooplankton Population, Seasonal Abundance, Anzali Wetland

### **1. Introduction**

Zooplankton is a very tiny organism found in every aquatic ecosystem. As the second

producer, they are the main energy transferor between primary producers and other levels of food chain including fish. They are also the most significant biotic organism influencing all the aspects of every aquatic ecosystem, such as food webs, energy and material cycle meanwhile they have an undeniable role in natural and artificial fish nutrition. Mutually there are different environmental factors that effect on zooplankton abundance and distribution such as water temperature, presence of nutrients and physico-chemical factors [1].

As Zooplanktons respond immediately to environmental changes, their species composition enables to show any signs of pollution or any decline in environmental qualification of ecosystems. Anzali international wetland provides various ecological values that are frequently overlooked. Physicochemical factors such as temperature, pH, Do and electrical conductivity form part of abiotic elements of anaquatic ecosystem [2]. As an example, intolerable levels of water temperature would limit the abundance of zooplanktons well as high pH levels may lead to the death of zooplankton, moreover sensitivity to low amount of dissolved oxygen would influence on zooplankton various life stages and different biological functions including feeding, growth, and reproduction.

Experimental results from the Anzali wetland recommend that there are currently a lot of resources of industrial and agricultural run-offs and urban wastes which are discharging to the wetland and affecting the aquatic species abundance and diversity specially by unlimited volume of nutrients, which has led to undesired algal blooms, fish mortality, and loss of zooplankton population and diversity and consequently would enter to the Caspian sea.

This study pointed at the influence of physicochemical factors on zooplankton seasonal species abundance in Anzali International wetland. The results obtained from this research hopefully will assist a clear understanding of zooplankton role in survival and environmental qualification of Anzali wetland and the related aquatic communities.

#### 2. Materials and Methods

Anzali wetland (49°55'E and 37°20'N) is the largest coastal wetland in the south of the Caspian Sea in Guilan province, Iran, covering an area of 500 ha. It has an average depth of 1.2 m and gets deeper toward the west. The annual rainfall in the catchment area varies from 2000 to 2200 mm. The water is mainly used for irrigation, fisheries and boating.

Anzali wetland can be divided into 4 sections. In the present study, 9 stations have been selected to represent its four sections and the interaction with the Caspian Sea. These stations designated as A, B, C, D, E, F, G, H and I. Station A is located on the furthest part of the west section of wetland. Station B is located in the middle of the wetland which is also wildlife refuge and manages by Department of Environment (DoE). Station C is located in the south part of the wetland and also manages as conserved area by DoE. Station D is the old wave breaker and previous entrance of the wetland to the open sea. Station E is in the east section of the wetland and receives effluents from agricultural and urban estate. Station F is selected in the entrance of open sea and near the new wave breaker that is under construction. Station G is located in the central section of the wetland and manages as wildlife refuge by DoE. Station H is in the main output river of the wetland to the Caspian Sea and Station I is situated in the entrance of the west section of the wetland where a new path after mechanical drainage process has been opened.

For zooplankton sampling Water samples (1000 ml) were collected for qualitative analysis from approximately 20 cm below the water surface at each station and were filtered through 30  $\mu$ m mesh size plankton net and were poured in new, clean 300 CC polyethene bottles and permanently labeled, fixed in 4% formalin [3]. For zooplankton identification and quantitative analysis the sample in each bottle was collected with Stempe 1ml pipette. The pipette content was transferred unto Bogorov counting chamber for species enumeration (each sample having three replicates of the concentrate and totally there were 27 samples per month and 324 samples for the whole period of sampling) which were examined under the Canon binocular invert microscope using the scanning, low-power and high-power objectives at 100 - 200× magnifications [4]. The different organisms identifies were classified into different families and species and their frequencies were noted. Finally the statistical analysis of species and determination of standard deviation have been done by the usage of SPSS version16.

#### 3. Determination of Physiochemical Parameters

**Water Temperature:** It was determined using mercury-in glass thermometer by dipping it into the water and allowed to stabilize for 5 seconds, removing and reading immediately recorded according to APHA, 2005 method.

**pH, Total Dissolved Solids, Electrical Conductivity and Salinity:** These were measured using pH/EC/TDS/Salinity meter (HANNA 3100 Model) by dipping the probes into the water until the screen showed a fixed reading as described by the manufacturers.

**Dissolved Oxygen (DO):** It was determined using DO meter (HANNA model HI 9146) in which the probe was inserted into the water until DO reading in mg/l was recorded as described by the manufacturers.

5-Day Biochemical Oxygen Demand (BOD5) and Chemical Oxygen Demand (COD): It was measured after collecting the samples into a labeled 250 ml brown bottle, kept in an incubator at research laboratory at 21°C for 5-days, then DO was measured again.  $BOD_5$  was obtained by subtracting the 5-day DO reading from the 0-day DO reading APHA, 2005. For determination of Chemical Oxygen Demand (COD) the Open reflex has been used.

Nitrate (NO<sub>3</sub>): A simple and rapid method has been proposed for the determination of nitrite using methylanthranilate as a coupling agent. Sulfanilic acid was diazotized in acidic medium and coupled with methylanthranilate to give a colored dye having absorption maximum at 493 nm. Determination of nitrate is based on the reduction of nitrate to nitrite in the presence of Zn/NaCl. The produced nitrite issubsequently diazotized with sulfanilic acid and then coupled with methylanthranilate to form an azo dye and was measured at 493 nm.

**Total Phosphate (T- PO\_4^{3-}):** A sensitive spectrophotometric manual determination method of phosphorus for the T-  $PO_4^{3-}$  has been used based on formation of the blue

bismuth-phosphomolybdate complex (BiPMo), was developed. Ascorbic acid is used for the reduction of bismuth-phosphomolybdic acid to bismuth-phosphomolybdate complex. Beer's law is obeyed for the concentration range to 0.6 mg· $\ell^{-1}$  (aqueous solution) and to 1.2 mg· $\ell^{-1}$  P (IBMK).

#### 4. Results

Physicochemical characterization of Anzali wetland was summarized in **Tables 1-4**. The amount of pH was nearly similar during the year but tends to be more alkaline. The parameters like Nitrate ( $NO_3$ ), Phosphate (T-PO<sub>4</sub>), Salinity and TDS where recorded maximum in summer which shows their relativity with water temperature, while by lack of enough dissolved oxygen (DO) in summer and spring, the amount of Biological Oxygen Demand (BOD<sub>5</sub>) and Chemical Oxygen Demand (COD) showed a sharp

Table 1. Physicochemical parameters of 9 stations in Anzali wetland during spring.

Spring	Station								
	1	2	3	4	5	0	/	0	9
DO	7.81	6.51	6.57	8.97	7.19	9.52	8.22	7.05	7.84
Water Temp (°C)	16.33	18.33	18	16.33	18	16.33	18.33	17.33	16.67
pH	7.94	8.42	7.94	8.18	7.39	8.39	8.14	7.68	7.58
NO <sub>3</sub> (mg/l)	1.28	1.46	1.99	2.38	1.64	2.26	2.43	2.35	2.94
T-PO <sub>4</sub> (mg/l)	0.25	0.29	0.25	0.25	0.25	0.25	0.23	0.28	0.28
Salinity (mg/l)	240.33	296.67	279.33	463.67	293	492.33	246.67	354.33	240.33
Conductivity (µS/cm)	246.67	603	570.67	1283	623.33	1347.33	519.33	817	478.33
TDS (mg/l)	276.33	304.33	460.67	1564.33	518.67	1947	261.67	442.67	230.67
BOD (mg/l)	12.33	14.33	10.83	12.33	13	13.33	15.33	13.33	10.83
COD (mg/l)	25.67	29.67	22.33	24.67	26.67	27.67	31.33	27.33	32

\*Characteristics were shown as average value for all stations.

Table 2. Physicochemical parameters of 9 stations in Anzali wetland during summer.

Summer	Station 1	Station 2	Station 3	Station 4	Station 5	Station 6	Station 7	Station 8	Station 9
DO	5.76	5.55	4.91	8.39	5.04	8.81	7.56	6.28	6.55
Water Temp (°C)	19.33	20.07	20.6	19.47	20.6	19.07	20.27	19.67	18.53
pН	8.11	8.34	7.96	8.15	7.15	80.25	8.11	7.64	7.2
NO <sub>3</sub> (mg/l)	3.86	3.44	4.13	4.03	3.8	3.69	3.67	4.05	3.87
T-PO <sub>4</sub> (mg/l)	0.35	0.24	0.55	0.27	0.46	0.37	0.3	0.3	0.32
Salinity (mg/l)	491.67	430.53	146.07	89.53	468.6	612.67	406.13	504.27	383.47
Conductivity (µS/cm)	265.67	617.6	582.73	293.4	638.47	1353.47	546.07	870	492.07
TDS (mg/l)	291.62	316.19	489.24	583.33	527.95	1957.14	273.24	471.1	242.24
BOD (mg/l)	7.17	5.67	4.57	9	5.2	9	12.47	9.07	8.77
COD (mg/l)	15.67	12.33	10.07	19.67	19.67	19.65	25.67	19.27	18.2

A t	Station								
Autumn	1	2	3	4	5	6	7	8	9
DO	8.18	6.9	7.13	9.19	6.46	8.94	8.01	6.89	7.32
Water Temp (°C)	16.33	16.67	17	16.33	17	16.33	16.67	16.67	16.67
рН	8.43	7.8	7.82	7.96	7.52	8.1	7.84	7.7	8.06
NO <sub>3</sub> (mg/l)	2.65	2.57	0.76	2.27	2.03	1.19	0.99	0.97	2.57
T-PO <sub>4</sub> (mg/l)	0.28	0.26	0.27	0.21	0.28	0.24	0.24	0.24	0.27
Salinity (mg/l)	383.33	396	399	534	421	548.33	388.67	500	385.33
Conductivity (µS/cm)	294	581.33	534	292.67	702	1352.67	443.33	767.67	403.33
TDS (mg/l)	247.33	318	318	85.67	507	1739.67	240.33	363	234.33
BOD (mg/l)	6	9	13.33	11.7	15	15	7	15.67	5.33
COD (mg/l)	13.67	20	28.67	24.81	31.33	31	15.67	34	11.67

Table 3. Physicochemical parameters of 9 stations in Anzali wetland during Autumn.

Table 4. Physicochemical parameters of 9 stations in Anzali wetland during winter.

Winter	Station 1	Station 2	Station 3	Station 4	Station 5	Station 6	Station 7	Station 8	Station 9
DO	8.62	7.46	7.92	10	7.63	10.34	8.86	8.37	8.34
Water Temp (°C)	10.67	11.67	11.33	10.67	11.33	10.67	11	11.3	10.67
pH	8.96	8.33	8.03	8	8.05	8.23	8.2	7.78	8.5
NO <sub>3</sub> (mg/l)	1.55	1.69	1.59	1.26	0.77	1.23	3.67	2.17	1.25
T-PO <sub>4</sub> (mg/l)	0.24	0.22	0.21	0.21	0.23	0.24	0.19	0.27	0.26
Salinity (mg/l)	257.33	264.33	269.67	440.33	279.67	470.33	257.33	336.67	252
Conductivity (µS/cm)	293	655.67	538.33	1260	831.33	1328	388	621	442.33
TDS (mg/l)	200.67	373.67	330	1298.33	513.33	1755.33	238.67	317.67	236
BOD (mg/l)	9.33	16	21.33	16.67	24	19.33	10.3	22.67	8.67
COD (mg/l)	19.67	33.3	43.69	34.67	49.33	40	21.67	47.33	18.33

decrease. Electrical Conductivity (EC) was approximately equal in all seasons and did not show a special seasonal change. The annual investigation represented that the highest annual amount of DO was 10 mg/l in station 4 in winter and the lowest one happened in station 5 in summer 5.04 mg/l. Nutrients like nitrate and phosphate were ranged between 0.77 - 4.13 mg/l and 0.21 - 0.46 mg/l respectively from winter to summer with a clear sharp rise in compare with other parameters. During the study period, zooplankton biodiversity of Anzali wetland was represented by 61 species consisting of 30 species (49.23%) of Rotifera, 22 species (28.54%) of Protozoa, 5 species (11.67%) of Copepoda, 4 species (10.56%) of Cladocera (**Table 5**).

In the present investigation, Rotifera was found as the most dominating group in the wetland and represented by species like *Brachionus calyciflorus, Filinia longiseta, Keratella quadrata, Lecane ludwigii, Lepadella patella, Monostyla cornuta* and *Philodina erythrophthalma* (Table 6). Rotifers are chiefly freshwater forms and presence of these

Sp. phylum	Spring (Ind/l)	Summer (Ind/l)	Autumn (Ind/l)	Winter (Ind/l)	Percentage (%)
Protozoa	$36.4\pm0.23$	$115.7\pm0.36$	$58.2\pm0.32$	$47.3\pm0.26$	49.23
Rotifera	$187.5\pm0.5$	$376.4\pm0.13$	$131.7\pm0.32$	$114.5\pm0.35$	28.54
Copepoda	$27.3\pm0.14$	$76.8\pm0.41$	$38.5\pm0.47$	$31.4 \pm .18$	11.67
Cladocera	$23.5\pm0.09$	$65.4\pm0.5$	33.6 ± 0.29	$25.7\pm0.11$	9.56
Rotifera Copepoda Cladocera	$187.5 \pm 0.5$ $27.3 \pm 0.14$ $23.5 \pm 0.09$	$376.4 \pm 0.13$ $76.8 \pm 0.41$ $65.4 \pm 0.5$	$131.7 \pm 0.32$ $38.5 \pm 0.47$ $33.6 \pm 0.29$	$114.5 \pm 0.35$ $31.4 \pm .18$ $25.7 \pm 0.11$	28.54 11.67 9.56

Table 5. Seasonal variation in densities of different groups of zooplankton in Anzali wetland.

<b>Table 6.</b> List of zooplankton species recorded in Anzali wetland d	luring 2012-2013.
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Protozoa	Rotifera
Arcella arenaria	Polyarthra dolichoptera
Arcella costata	Polyarthra vulgaris
Tintinopsis sinensis	Brachionus calyciflorus
Tintinopsis conicus	Brachionus angularis
Tintinopsis cratera	Brachionus variabilis
Tintinopsis lacustris	Brachionus rubens
Tintinnidiumentzii	Keratella quadrata
Tintinnidiumwangi	Cephalodella gibba
Difflugia acuminate	Philodina erythrophthalma
Difflugia oblonga	Filinia longiseta
Difflugia oviformis	Monostyla cornuta
Difflugia lebes	Monostyla hamata
Euglypha alveolata	Lepadella patella
Euglypha tuberculata	Lepadella ovalis
Actinosphaerium eichhornii	Lecane curvicornis
Centropyxis aculeata	Lecane lodwigii
Cyphoderia alveolata	Synchaeta pectinata
Paramecium caudatum	Synchaeta vorax
Amoeba polypodia	Synchaeta stylata
Marituja pelagica	Synchaeta oblonga
Trinema lineare	Trichocera pussilla
Vorticella campanula	Trichocera porcellus
	Colurella adriatica
	Euchalanis diltata
	Dissotrocha aculeata
	Trichotria tetractis
	Trichotria pocillum
	Testudinella patina
	Epiphanes brachionus
	Lophocharis oxysternon
	Copepoda
	Naplious copepoda
	Cyclops vicinus
	Cyclops scutifer
	Cladocera
	Thermocyclops dybowskii
	Alona rectangular
	Alona costata
	Bosmina coregoni

organisms in abundance is related to suitable conditions for their survival. In Anzali wetland *Brachionus* sp. and *Polyarthra* occurred as most abundant species. In other

fresh water ecosystems in the north part of Iran has been registered as dominated plankton community in this area Polyarthra dolichoptera was also abundantly observed in Anzali wetland that the ccurrence of this genus along with *Brachionus* sp. indicate nutrient rich status of the waterbody. Rotifer richness and its biodiversity were found to be at the peak in summer in Anzali wetland indicating the influence of temperature which was supported by direct relation between summer temperature and Rotifer population. In Anzali wetland, biodiversity of Protozoans was represented by species such as Difflugia acuminate, Arcella arenaria, Centropyxis aculeate, Tintinopsis sinensis, Amoeba polypodia, Paramecium caudatum, Euglypha alveolata (Table 6). Protozoan population was recorded at peak in summer months and their count remains low during cold rainy seasons. Dilution of water caused by heavy rain and snow in winter can partly explain low protozoan count observed during winter while maximum population during spring and summer months indicated a positive relationship with temperature, nutrients and high population of phytoplankton (Boyden, 2007)<sup>9</sup>. Common Copepoda species in Anzali wetland were Cyclops vicinus, Cyclops scutifer, Thermocyclops dybowskii and the most observed Cladoceran species were Alona rectangular, Alonacostata, Bosmina coregoni and Bosmina longirostris (Table 5). During the present investigation, Cladocera were found to be high in spring followed by summer and minimum in winter months. The maximum population of Cladocera in spring and summer may be attributed to favorable temperature and availability of food and form of bacteria, nonplankton and suspended detritus in these seasons and physicochemical parameters like water temperature, dissolved oxygen and nutrients play an important role in controlling the diversity and density of Cladocera. In the present investigation Cyclops and Nauplii were recorded as copepods. Anzali wetland richness in organic matter support higher number of Cyclopoids, thus suggesting their preponderance in higher saprobe state of water [5]. Maximum number Copepods population was recorded in early summer months while minimum in winter which showed the close relationship between copepod population and higher temperature.

#### 5. Discussion

According to similar studies it has been found out that among all physic-chemical factors the 9 selected factors are the most effective and important ones as they have direct influence on zooplankton population and abundance moreover these factors are the most sensitive and variable factors of all in recent decades. This research showed that the significant source of anthropogenic perturbation in wetland is nutrients (input) enrichment so the increased amount of BOD, COD, TDS, Nitrate and Phosphate in stations B, E and G suggests the impacts by human interferences more than other stations whilst such increase in station H, indicates the huge volume of output of the wetland discharges to the entrance of the sea where in rainy seasons an extra amount of chemical and organic nutrients discharges in this station. It also has been found out that total nitrate concentration significantly contributed to zooplankton variability in addition to water temperature hence it has been deduced the increase of nitrate concentration in Anzali wetland could raise the zooplankton abundance and diversity where of the primary productivity of Anzali wetland increased which led to the increase of zooplankton abundance and species biodiversity. Similar result has also been observed by other researcher where they reported that zooplankton are favored in nutrient rich environments particularly wetlands. Furthermore, zooplankton is more dependent on dissolved oxygen during summer and late spring, therefore an increase in dissolved oxygen during these seasons would increase their abundance [6].

Also worthy of note is the positive correlation between total dissolved solids and zooplankton diversity and abundance in spring and summer [7]. TDS is basically used to define the total ions in solution in the water. It could therefore be implied from the positive correlation that an increase in the total ions in solution increases the density and abundance of zooplankton. High concentrations of nutrients such as phosphate and nitrate usually give rise to the high abundance of some zooplankton species in aquatic environment. As a large wetland in this province, Anzali wetland has rich diversity of rotifers such as Dissotrocha aculeate, Lecane ludwigii, Monostyla hamata due to the abundant of vegetation overgrowth in Anzali wetland. meanwhile observation of species such as Brachionus angularis, Brachionus calciflorus, Brachionus vulgaris, Euchlanis dilatata, Filinia longiseta, Lecane curvicornis, Testudinella patina indicate that a lot of zooplankton taxa in Anzali wetland have evaluation toleration to the salinization. This observation was also reported by [8] who noted that salts like magnesium and calcium promotes zooplankton growth. Water level rise in the rainy seasons also decreased zooplankton density as observed during autumn and winter. This is due to pressure impacted on the cell by the water level. There is a strong relation between water level, biochemical oxygen demand (BOD), chemical oxygen demand (COD), TDS, alkalinity and rainfall [9]. This indicates the major source of organic pollution is through surface run-off and showed that the source of contamination is washed in with the rain, therefore organic pollution increases with the rain.

#### 6. Conclusion

During the research period, some drainage operations had been done in various parts of the wetland, so some stations were difficult to access, also vegetation overgrowth, algal bloom and sensible decrease in water level led to determination of stations in suitable stations with related records of previous studies, but because of lack of enough regular studies on Anzali wetland in recent years specially before the drainage operations, it was arduous to compare the results with the related background. Biological and seasonal differences influence distribution and abundance of zooplankton communities. According to their exobiological characteristics, zooplankton fauna of a wetland may vary considerably at spatial scale, temporal scale and time scales. Rotifers appeared to be the most dominating community throughout the study period. Overall zooplankton population fauna of the wetland was much more diversified indicating the Anzali wetland as nutrient rich water body which undergoes the state of eutrophication in the recent decade, if not managed properly. Pollution-tolerant species density in most polluted areas such as station B, E, G and H had nutrients, TDS and BOD as specific physic-chemical factors, while all species had strong correlation with DO and water temperature. In this study, most of the zooplankton species were freshwater species despite many of them have been sampled in saline stations such as stations D and F. The achievements of this research could be contributed to understating of anthropogenic influences on ecosystem stability and their specific effects on species abundance and could be utilized in integrated monitoring projects for reducing opposing effects reduction and pollution control.

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