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Seasonal Distribution of Copepods as Influenced by Physicochemical Parameters in Khanki Headworks, Pakistan

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ABSTRACT

Copepods are tiny aquatic animals. Their role in aquatic food chains is very crucial as they are involved in organic matter circulation and energy transfer between different trophic levels. They are important source of food for fish larvae and zooplanktivorous fish. **Objective:** To assess the influence of environmental variables on the abundance, diversity and seasonal variations of planktonic copepods in Khanki Headworks, Pakistan. **Methods:** For the analysis of environmental variables monthly water samples from four selected sites were collected for a period of one year (February 2021 to January 2022). Environmental variables were analyzed in laboratory by following standard procedures. Month wise samples of copepods were collected with planktonic net (mesh size: 37μm) from four locations (each with 3 sub-sites). **Results:** In total, 7 species of Copepods belonging to 4 genera were identified from February 2021 to January 2022. Mesocyclops was observed as the most diverse genus (4 species), while Mesocyclops edax was the most prevalent copepod species. Population density and biodiversity were highest in June and minimum in January. Shannon-Weaver diversity index described greater diversity among copepod species in June. Pearson correlation and canonical correspondence analysis (CCA) revealed that electrical conductivity (EC), temperature, turbidity, pH and dissolved oxygen (DO) were important environmental variables affecting the biodiversity and density of copepods. **Conclusions:** This investigation elucidated that physicochemical parameters generally regulate the population dynamics of Copepods.

INTRODUCTION

Freshwater zooplankton communities generally consist of Rotifera, Protozoa and Microcrustacea [1]. Aquatic ecosystems are colonized by different groups of planktonic microcrustaceans like copepods, ostracods and cladocerans [2]. Copepoda is considered as the most common zooplankton group in the aquatic ecosystems [3]. There are approximately 14,000 known copepod species, whereas, only 3000 species are present in freshwater [4, 5]. Copepods are an important source of fish food and their use as live fish feed is also common in aquaculture because they promote the development and growth of fish larvae [6]. Decline in abundance and diversity of copepods resulted in the downfall of aquaculture in many countries [7, 8]. Microcrustaceans perform a very significant role as an intermediate link in food webs and food chains by transferring primary production and energy from lower to upper trophic levels [9, 10]. Zooplankton body size plays an important role in improving the water quality of any aquatic

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reservoir because large zooplanktons control the algal blooms by grazing on them [11]. However, an increase in temperature and total phosphorus levels results in small mean size of zooplanktons [12, 13]. Environmental variables play a very crucial role in shaping the community structure of aquatic invertebrates [14]. Microcrustaceans (copepods) are very sensitive to environmental changes and show different preferences and specific responses to various environmental variables [15, 16]. River Chenab is very prominent freshwater ecosystem in Pakistan and it has very diverse aquatic flora and fauna [17, 18]. Khanki Headworks is an important site, providing habitat to prominent fish fauna, however, no previous study is conducted in this area regarding our present investigation. Present investigation is the first baseline study that was conducted for a period of one year (February 2021 to January 2022) at Khanki Headworks with following objectives (1) to study seasonal variations in community structure of planktonic copepods, (2) to investigate the role of environmental variables in shaping the population dynamics of microcrustaceans, (3) to emphasize the role of copepods in fresh water ecosystems as bioindicator of trophic status.

METHODS

The study area "Khanki Headworks" is one of the important headworks that are situated on Chenab River, Punjab, Pakistan. At first it was built in 1889 for flood control and irrigation of barren agricultural lands. Head Khanki irrigates vast area (three million acres) of agricultural lands by diverting water to the Lower Chenab Canal and 59 minor tributaries. It is also valuable site for fishing due to its notable fish fauna. For the analysis of environmental variables monthly water samples from four sites were collected for a period of one year (February 2021 to January 2022). For sampling pre-cleaned 1 liter plastic bottles were used. Water samples were usually taken in early hours (9.0 A.M to 12 P.M). Measurements of Temperature, total dissolved solids (TDS), pH, turbidity, electrical conductivity and dissolved oxygen (DO) were taken in the field immediately with their respective meters [19, 20]. Other environmental variables such as total hardness (TH) and total alkalinity (TA) were analyzed in laboratory by following standard procedures [21]. Month wise samples of copepods were collected with planktonic net (mesh size: 37μm) from four locations (each with 3 sub-site). Planktonic net was placed in water in such horizontal position that 60 liters of water could pass through it. Plankton sampling was carried out for one year from February 2021 to January 2022. Pre-cleaned plastic bottles (50 ml) were used for copepods sampling. After that formalin solution (5%) was added in bottles for fixation and further analysis [22, 23]. Copepods species were identified with standard keys and relevant literature [24-28]. Identification of copepods was made possible by observing their antenna shape, arrangement of ovarian bags, general body shape. Sedgewick-Rafter counting chamber was utilized for the enumeration of copepods [21]. Copepods were observed under an inverted microscope (LEICA HC 50/50) fitted with 5 mega pixel camera. Photographs of copepods were taken by using this inverted microscope. Living copepods stained with 1% neutral red were also observed under the microscope. Relative abundance and biodiversity of copepods were calculated with Shannon-Weaver (H) and Simpson (D) diversity indices [29, 30]. Species richness (SR) was estimated by Margalef [31], whereas species evenness was quantified by Pielou [32]. Relationships between copepods species and different environmental variables were evaluated by Pearson's correlation. Month wise copepods data were subjected to one way ANOVA to evaluate significant difference among population density of copepods in different months. ANOVA and Pearson's correlation were applied using R software. Relationships between Copepod species and various months were computed by Principal Component Analysis (PCA), whereas, correlations between microcrustacean fauna and environmental variables were determined by using CCA (Canonical Correspondence Analysis). PCA and CCA were performed using XL stat 2022.

RESULTS

We were able to identify 7 species of copepods. Copepod species were related to 4 genera and one family (Table 1).

Table 1: List of Copepod species identified from Khanki Headworks (February 2021 to January 2022)

No.	Family	Genus	Species
1	Cyclopoidae	Eucyclops	Eucyclops elegans
2		Mesocyclops	Mesocyclops aspericornis
3			Mesocyclops leuckarti
4			Mesocyclops edax
5			Mesocyclops inversus
6		Microcyclops	Microcyclops varicans
7		Ectocyclops	Ectocyclops phaleratus

Mesocyclops edax was the most abundant copepod species, whereas, Microcyclops varicans showed minimum abundance (Figure 1).

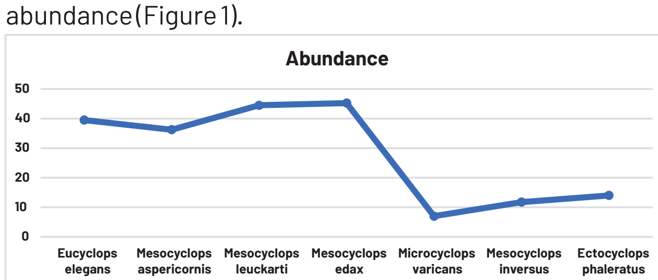

Figure 1: Relative abundance of copepod species isolated from

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Mesocyclops (4 species) was the most diverse genus, while Eucyclops was noted as the most abundant copepod genus (Figure 2).

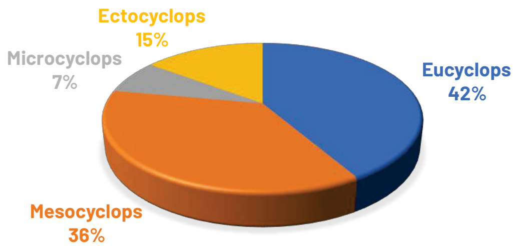

Figure 2: Percentage representation of copepod genera isolated from Khanki Headworks

Shannon-Weaver diversity index (H) exhibited highest values in June and lowest values in January. Similar trend was computed by Simpson diversity index and its high values for copepods were recorded in June and lowest values in January (Figure 3).

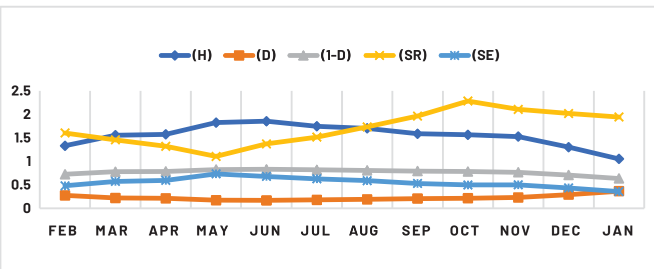

Figure 3: Variations of diversity indices of copepods isolated from Khanki Headworks

H (Shannon-weaver diversity index), D (Simpson index of dominance), 1-D (Simpson index of diversity), SR (Species richness), SE (Species evenness)

In the month of June, maximum diversity of copepods (7 species) was observed, whereas, in January, minimum diversity of copepods (3 species) was recorded. Statistically significant difference in copepods density from February 2021 to January 2022 was noted by ANOVA (Table 2).

Table 2: Analysis of variance of copepods ($p<0.05$) from Khanki Headworks (February 2021 to January 2022)

Source of Variation	SS	DF	MS	F	p-value	F crit
Between Groups	2785.338	1	2785.338	40.296	0.000	4.3009
Within Groups	1520.652	22	69.12056	-	-	-
Total	4305.99	23	-	-	-	-

SS= Sum of square, DF= Degree of freedom, MS= Mean of square, P= Probability, F= f-Distribution

Based on six principal components, 98.78% of total variance was represented by PCA. Axis F1 (45.95%) and axis F2 (33.77%) indicated total 79.82% variance in copepods community structure (Figure 4).

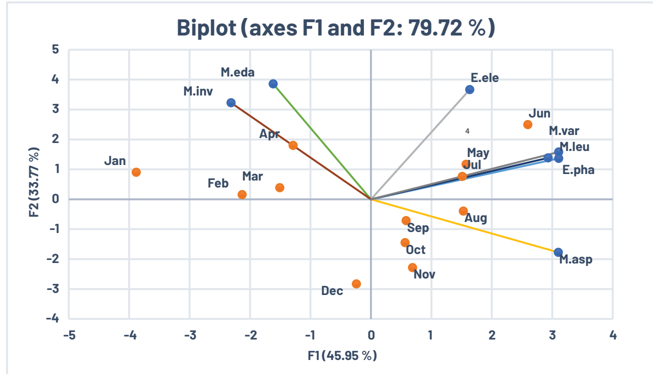

Figure 4: PCA biplot of 7 copepod species of Khanki Headworks from February 2021 to January 2022

CCA symmetric map reflected the effects of physicochemical

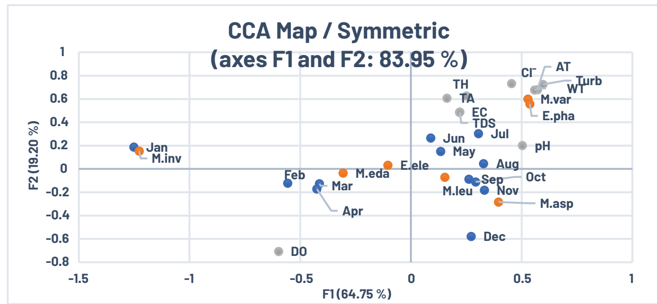

Figure 5: CCA ordination triplot depicting the copepod-environment relationships at Khanki Headworks

In present investigation temperature values varied between 39°C to 16°C, being maximum in June and minimum in January. Microcrustacean (copepods) fauna exhibited significant growth in summer months with maximum biodiversity (7 species) and abundance (27±5.92) recorded in June, whereas, minimum population abundance (10.5±1.75) and diversity (3 species) observed in January (Figure 6).

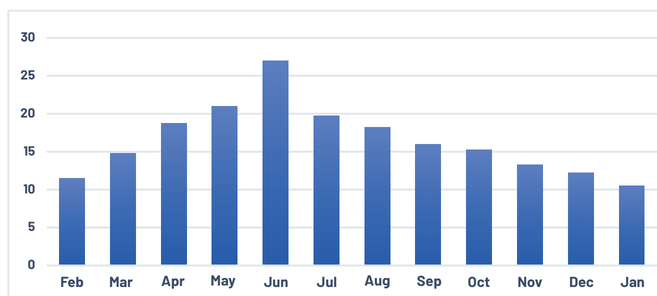

Figure 6: Population density of copepods on monthly basis from Khanki Headworks

Values of water pH were recorded moderately alkaline between 7-7.9, being highest (7.9) in July and lowest (7) in January. Seasonal fluctuations in DO values ranged from 5.34-6.97, being maximum (6.97) in January and minimum (5.34) in June. Electrical conductivity (EC) was recorded maximum (369 µs/cm) in June and minimum (270 µs/cm) in January. Seasonal variations in total hardness (TH) values ranged between 170 mg/l in June and 110 mg/l in January. Turbidity increased in summer months, whereas, it decreased in winter season. Maximum range (258 mg/l) of total dissolved solids (TDS) was noted in June while its minimum value

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(189 mg/l) was monitored in January. Water alkalinity was noticed maximum (180 mg/l) in June and lowest (100 mg/l) in January (Figure 7).

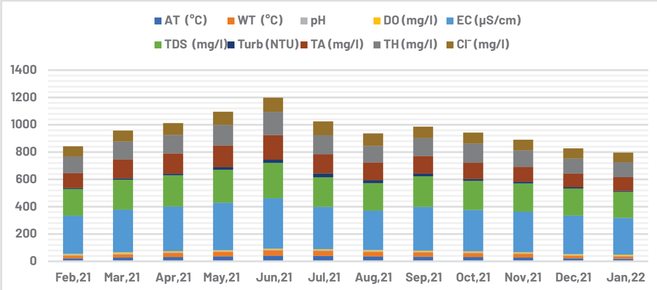

Figure 6: Figure 7: Variations of different physicochemical parameters, Temperature (°C), pH, Dissolved oxygen (mg/L), Electrical conductivity (µs/cm), Total hardness (mg/L), Turbidity (NTU), Total alkalinity (mg/L), Total dissolved solids (mg/L), Chlorides (mg/L) at Khanki Headworks

Temperature was observed to have positive influence on the population abundance of copepods. Water pH also exhibited positive relationship with microcrustacean community. According to Pearson's correlation dissolved oxygen (DO) had negative relationship with microcrustacean density and diversity. EC and TH were found to have positive correlation with microcrustacean fauna. Population abundance of copepods was found to be positively influenced by these above mentioned (Turbidity, TDS, Alkalinity) environmental variables (Table 3).

Table 3: Pearson correlations between copepods and physicochemical parameters

Vari-ables	Cope-pods	AT	WT	pH	DO	EC	TDS	Turb	TA	TH	Cl⁻
Cope-pods	1	-	-	-	-	-	-	-	-	-	-
AT	0.968	1	-	-	-	-	-	-	-	-	-
WT	0.961	0.994	1	-	-	-	-	-	-	-	-
pH	0.240	0.406	0.409	1	-	-	-	-	-	-	-
DO	-0.898	-0.956	-0.953	-0.486	1	-	-	-	-	-	-
EC	0.836	0.780	0.755	-0.136	-0.645	1	-	-	-	-	-
TDS	0.838	0.783	0.758	-0.133	-0.649	0.999	1	-	-	-	-
Turb	0.829	0.899	0.897	0.527	-0.939	0.537	0.539	1	-	-	-
TA	0.856	0.835	0.819	-0.022	-0.737	0.943	0.945	0.609	1	-	-
TH	0.837	0.821	0.789	0.021	-0.699	0.931	0.931	0.616	0.912	1	-
Cl⁻	0.939	0.957	0.950	0.298	-0.935	0.766	0.768	0.897	0.866	0.814	1

DISCUSSION

The motive behind this study was to explore the seasonal fluctuations in community structure of planktonic copepods. After qualitative analysis of samples, 7 Copepoda species were recorded. Mesocyclops edax was observed as most common and abundant species from available in all study months. Eucyclops (39.5%) was the most abundant genus of copepods. In summer months, microcrustacean copepods showed a significant growth in density and diversity, whereas, in winter season minimum abundance of copepods was recorded. High temperature was observed to be favorable for most species. Our findings were similar to previous studies [22, 33, 3]. Maximum diversity of copepods (7 species) was recorded in the month of June, whereas, minimum diversity was noticed in January (3 species). According to Maqbool et al., Photosynthetic activity also escalates as temperature rises which promotes the microcrustacean abundance [19]. During this study, pH values were moderately alkaline during sampling months, which were in accordance with previous reports [34, 23]. pH increased during summer months because of eutrophication and high levels of nitrates and carbonates in water, as reported by earlier researchers [22]. Population abundance and biodiversity of copepods increased at higher pH. Similar results were observed by previous researchers [35]. High DO levels were found during winter season, whereas, DO values decreased in summer season and lowest values were recorded in June (5.34 mg/l). During summer months BOD (biological oxygen demand) increases to decompose the organic matter that results in low level of DO in water. Rising temperature can reduce the water solubility of oxygen, as was reported in earlier investigations [20, 22, 36]. However, DO level was never recorded lower than 5 mg/l that is considered as threshold level for aquatic life [36, 37]. A negative correlation was established between microcrustacean abundance and DO level. This observation was similar to previous reports [38, 39]. In summer, high values of total dissolved solids (TDS) were noted because high temperature accelerates the decaying process of vegetation that results in more input of TDS in water. Previous studies also documented such results [40, 41]. Electrical conductivity (EC) values were higher in summer season and lower in colder months. These findings were consistent with previous studies [42, 43]. EC (electrical conductivity) ranged between 270 µS/cm to 369 µS/cm. High evaporation rate might be the reason of high EC values in summer months [19]. Both parameters (EC and TDS) positively affected the abundance of copepods. High turbidity was noticed especially in the month of July and August. During monsoon heavy rainfall unsettles the bottom sediments that causes an increase in water turbidity (43, 44]. According to Onyedineke et al., it favors the copepods abundance because of more food availability [45]. Total hardness (TH) and total alkalinity (TA) were recorded higher in summer months and lower in winter [8, 20]. For the measurement of hardness, generally magnesium (Mg^{++}) and calcium (Ca^{2+}) ions are considered, whereas, for the measurement of TA carbonates and bicarbonates ions are usually considered, as was mentioned [43]. Both variables (TH and TA) were found to have positive influence on population density of copepods [46]. According to Shannon-Weaver index of diversity (H),

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highest (1.85) biodiversity was calculated in June and lowest (1.05) in the month of January. Values of Simpson diversity index for copepods ranged from 0.63 to 0.83, being highest in June and lowest in January. Simpson index of dominance exhibited minimum value range for copepods (0.17) in June, whereas, maximum value range (0.36) in January. PCA findings revealed; 4 species in group 1(Upper right), 1 species in group 2 (bottom right), no species in group 3 (bottom left) and 2 species in group 4 (upper left). Maximum biodiversity was related to the months of May, June and July. CCA results depicted interactions between copepods and environmental variables. According to CCA symmetric map, different hydrological parameters (Temperature, EC, pH, turbidity and TH) had strong influence on the distribution and abundance of copepods. Dissolved oxygen (DO) manifested negative impact on the diversity of copepods.

CONCLUSIONS

Present study provided baseline knowledge and information about seasonal variations in distribution of copepods in relation to environmental variables in Khanki Headworks. Physicochemical variables (temperature, EC, DO, TH, TDS) shaped the community structure of copepods. High population abundance and diversity were observed in summer season as compared to winter. Our findings revealed that copepods are very sensitive to any physical and chemical change in surrounding environment. Thus, they can provide valuable information regarding the trophic status and water quality of freshwater ecosystems.

Authors Contribution

Conceptualization: NR
Methodology: MAR, MAI
Formal analysis: MAR, HA
Writing-review and editing: MAR
All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

The authors declare no conflict of interest.

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