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**Biodiversity of Rotifera and Cladocera in Al-Hilla  
River, Babylon Province, Iraq**

*A Thesis*

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of Doctorate Of Philosophy In Biology

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*Dedication*

*To*

*My Family*

*Wameedh 2023*

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## Summary

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Five stations along Al-Hilla River have been utilized to study the morphological and genetic diversity of zooplankton (Rotifera and Cladocera). The first station (st1) represent Al-Hilla River as it enters the province of Babylon at the village of Anana, the second station (st2) as it approaches the city center (near the Betta Bridge), the third station (st3) as it leaves the city center in the Al-Farsi area, The fourth station (st4) is a branch that arises from Al-Hilla River and flows through the village of Al-Ghalis and the fifth station (st5) is a branch that diverges from Al-Hilla River at Nazim Dora and travels through the region between Al-Dolab and Al-Dabla. Moreover, zooplankton samples (Rotifera and Cladocera) are obtained seasonally using a zooplankton net with 30 cm diameter and 50  $\mu\text{m}$  mesh size. All samples are counted and photographed before being categorized using taxonomic keys.

Some physical and chemical characteristics of water are investigated seasonally between spring 2021 and winter 2022. These characteristics included the air temperature, which ranged between (11.73 - 46.01)  $^{\circ}\text{C}$ , water temperature (10.51- 35.21)  $^{\circ}\text{C}$ , pH (7.4-8.38), electrical conductivity (520.66 - 1375.6)  $\mu\text{s}/\text{cm}$ , total dissolved solids (362.66 - 964)  $\text{mg}/\text{l}$ , salinity (347.5 - 691)  $\text{mg}/\text{l}$ , and dissolved oxygen (4.94 - 10.25)  $\text{mg}/\text{l}$ .

Zooplankton (Rotifera and Cladocera) densities ranged from the lowest value (0.32)  $\text{Ind}/\text{L}$  in the fifth station during the autumn season to the maximum value (1.386)  $\text{Ind}/\text{L}$  in the fourth station during the summer season. For the majority of the study stations, Cladocera and Rotifera density increased in the spring and the summer, respectively. The lowest

mean number of taxa (5.33) is recorded in the fifth station during the fall season, while the maximum mean number of taxa (14) is recorded in the summer season in the first, second, and fifth stations. The current study also revealed variation in species densities, with the dominance of each of the following species representing the dominance of Rotifera: *Bdelloidea*, *Trichocerca rattus*, *Euchlanis dilatata*, *Trichotria tetractis*, *Keratella cochlearis*, and *Testudinella patina*, while the dominance of each of the following species representing the dominance of Cladocera: *Eubosmina longispina*, *Acroperus angustatus* and *Daphnia magn.*

To compare the diversity and abundance of species in the stations during the study period, some biodiversity indices are also investigated, including Shannon-Wiener index, where values ranged between (1.721-2.83), Margalef index (1.278-3.351), The index of dominance (0.071-0.250), Equitability (Evenness) Index (0.717-0.913), Menhinick index (0.343-0.776), as well as the Berger-Parker index (0.13-0.4). The findings showed that the majority of the stations had an average level of biodiversity, which is impacted by changes in the physical and chemical properties of water. According to Jaccard's similarity index, the second and third stations shared the most similarities, while the first and fifth stations shared the least similarities.

According to phenotypic identification, this study included the recording of (69) taxa of Rotifera, which included (54 species and 12 genera) that belonged to 17 families, as well as the recording of three species (*Ascomorpha ovalis*, *Horaella brehmi*, and *Lecane arcula*) for the first time in Iraq and four species (*Lecane quadridentata*, *Lecane tenuiseta*, *Lecane unguulate*, and *Scaridium longicaudum*) for the first time in the Hilla river. While (14) taxa of Cladocera are recorded and included

(8 species and 2 genera) belonging to 4 families, in addition to recording five species (*Acantholeberis curvirostris*, *Acroperus angustatus*, *Anthalona verrucosa*, *Alonella exigua* and *Eubosmina longispina*) for the first time in Iraq. Regarding genetic identification, it identified 83 species of Rotifera based on the analysis of the gene sequence and comparison of the results with the National Center for Biotechnology Information (NCBI), including two species (*Bryceella stylata* and *Ascomorpha ovalis*) that are recorded for the first time in Iraq and three species (*Lecane tenuista*, *Lecane unguolata*, and *Scaridium longicaudum*) that are recorded for the first time in the Hilla river, and (40) Cladocera species are identified, including seven species (*Daphnia dubia*, *Daphnia laevis*, *Daphnia ambigua*, *Daphnia pulicaria*, *Daphnia longicephala*, *Polyphemus pediculus* and *Acantholeberis curvirostris*) that are recorded for the first time in Iraq. Also, the current study is the first to evaluate the Hilla River in terms of the environment and diagnostic of Cladocera, as well as the genetic diversity of zooplankton.

The current study also focused on defining the phylogenetic tree of the investigated species and stations. The findings showed a significant genetic convergence (82%) between the three species of Rotifera (*Trichocerca tenuior*, *Testudinella reflexa*, and *Polyarthra dolichoptera*), as well as a significant genetic convergence (92%) between Cladocera species (*Scapholeberis mucronata*, *Leptodora kindtii*, *Cercopagis pengoi*, *Evadne nordmanni*, *Podon leuckartii*, *Bythotrephes longimanus*, *Daphnia longicephala*, *Daphniopsis truncata*, *Daphnia ambigua* and *Daphnia occidentalis*). Also, the results showed that there is convergence between the first and fifth stations ( 85% ) for the Rotifera and (75%) for the Cladocera.

Some of the analyzed environmental parameters and zooplankton species have positive and negative correlations, according to SPSS statistical analysis and CCA analysis. While there is an inverse correlation between temperature and the amount of dissolved oxygen in the water, the air and water temperatures are both favorably significant. The findings also showed a positive significant correlation between salinity, total dissolved solids and electrical conductivity, while a negative significant correlation is found with pH. When it comes to the various Rotifers and Cladocera species, the majority of them showed a significant positive correlation with pH and dissolved oxygen, whereas they showed a significant negative correlation with electrical conductivity, total dissolved solids and salinity. The correlations between the species also differed.

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# *Chapter One*

## *Introduction and Literature Review*

## **1 : Introduction and Literature Review**

### **1 – 1 : Introduction**

The term "biodiversity" is perceived differently, depending upon the sociological group involved. Taxonomists, economists, agronomists and sociologists each have their own partial view of the concept. Biologists tend to define biodiversity as the diversity of all living beings. Farmers are interested in exploiting the manifold potential deriving from variations over soils and regions. Industry sees a reservoir of genes useful in biotechnology or a set of exploitable biological resources (timber, fish, etc.). As for the general public: its main concern is with landscapes and charismatic species threatened by extinction (Leveque and Mounolou, 2003).

Biodiversity emerged as an environmental issue in the early 1980s, culminating in the Conference on Sustainable Development held in Rio de Janeiro 1992, defined three levels of biodiversity: Intraspecific genetic diversity, species diversity and diversity of ecosystems. The impact of the Intraspecific genetic diversity on the ecological processes in ecosystems may be comparable with that of species diversity (Hughes *et al.*, 2008).

Freshwater habitats provide humans with arguably the single most important natural resource. Furthermore, although freshwater only covers 0.8% of the Earth's surface, this area supports nearly 6% of all known diversity at least 100,000 species (Dudgeon *et al.*, 2006). Despite the importance of freshwater resources to society and the natural world, these fragile ecosystems and their associated species are being severely degraded by human activities (Moyle *et al.*, 2011). Human demands on freshwater are so great that declines in biodiversity are much worse in

freshwater ecosystems than in terrestrial ecosystems (Dudgeon *et al.*, 2006).

Morphological identification of freshwater invertebrates used in many bioassessment and biomonitoring programs is exceptionally difficult because many are small and/or have numerous instars, dynamic life histories and systematic uncertainties (McCafferty *et al.*, 2008). Furthermore, species-level identifications generally require adult specimens, not the aquatic larval forms (Baird and Sweeney, 2011; Webb *et al.*, 2012 and Stein *et al.*, 2014). Genetic sequencing techniques have been found to be effective at identifying a wide variety of organisms, including macroinvertebrates (Hebert *et al.*, 2003; Hebert and Gregory, 2005; Sweeney *et al.*, 2011 ; Webb *et al.*, 2012).

Global biodiversity is currently threatened by a variety of stressors, including climate change (Pecl *et al.*, 2017). As environmental conditions continue to change, species are faced with limited response options: relocate, adapt to the new conditions, or go locally extinct (Bellard *et al.*, 2012; Singer *et al.*, 2013 ; Kadowaki *et al.*, 2016). For example, understanding the relative importance of abiotic conditions such as temperature, pH, salinity and dissolved oxygen for aquatic biodiversity is urgent because human activities alter the conditions of aquatic systems worldwide. Many rivers are still threatened by organic pollution in the form of run-off fertilizers and pesticides from intensive agriculture (Valle *et al.*, 2015), paper pulp factories (Karrasch *et al.*, 2006), potato or cassava starch companies (Arimoro *et al.*, 2008) and urban sewage (Couceiro *et al.*, 2007 ; Wen *et al.*, 2017).

In each work concerning the study of the diversity of communities, the use of diversity indices is a necessary tool to calculate and quantify the diversity status (Van Strien *et al.*, 2012). In addition, these indices estimate biological and ecological quality of an ecosystem through the structure of the community (Danilov and Ekelund, 1999), they are also possible indicators to monitor the level of pollution in environment (Washington, 1984). In so far as the use of a single numerical index to describe the structure of a community and the ecological quality of an ecosystem oversimplifies its real biodiversity, the literature proposes different indices to study the diversity of communities in aquatic ecosystems (De Pauw *et al.*, 1992 ; Beaugrand *et al.*, 2002).

A zooplankton community is a dynamic system in which the species composition may change significantly during the season. Temporal changes of the zooplankton species found in are influenced by many factors including temperature, food, competition and predation (Cimdin, 2001; Wetzel, 2001; Chang and Hanazato, 2004 ; Harris *et al.*, 2012). It is well-known fact that zooplankton are very sensitive to the biotic and abiotic influences which lead to structural and functional changes on a polymorphism level (De Meester *et al.*,1995). This makes them good bioindicator for the evaluation of water quality and toxicity as well as for experimental purposes in ecology and genetics (Sloka,1998; Dussart and Defaye, 2001). Rotifera are a group of microscopic animals (< 2 mm) that encompasses approximately 2000 species (Segers, 2007). They are globally distributed in freshwater and brackish habitats, along with a small number of marine habitats (Wallace *et al.*, 2006). Rotifera play a pivotal role in aquatic food webs by transferring energy to higher trophic levels (Xiang *et al.*, 2017). The subclass Monogononta which includes most species in the phylum comprises planktonic, littoral and benthic

organisms (Segers, 2008). Cladocera (water fleas) are primarily freshwater small-sized (0.2– 6 mm) inhabiting pelagic and littoral zones. Four Cladocera orders are recognized (Fryer, 1987) and about 620 species are currently known. They inhabit most types of continental fresh and saline water habitats, occurring more abundantly in both temporary and permanent stagnant waters (Forro *et al.*, 2008). The Cladocera have an important and active role in the ecosystem because of its location in the food chain between phytoplankton and fish, predatory invertebrates, and considered an important food for fish especially small fish, as well as an indicator to the environmental condition because of its sensitivity to any changes happening in the environment (Monaghan and Milner, 2010 ; Lotter *et al.*, 2012).

Molecular ecology concerns the application of molecular techniques to questions in ecology, evolution, behavior, diversity and conservation biology (Carvalho, 1998). Several technical and methodological developments in molecular biology in the 1980s facilitated such application among these, refinement in DNA extraction protocols, polymerase chain reaction (PCR), design of conserved primers which allowed amplification and sequencing of genes of virtually any organism. The genetic variation between and within species not studied at molecular level in Iraq and especially in the Hilla river before and these are the aims of the current study.

**1 – 2 : The Aim of Study**

The aim of this study is to investigate the seasonal diversity of zooplankton (Rotifera and Cladocera) in Al-Hilla River and some streams branched from it. This aim accomplished through following objectives :

- 1 – Assess some physical and chemical characters of water.
- 2 - Identify and classify the zooplankton (Rotifera and Cladocera).
- 3 - Utilizing genetic diversity for Rotifera and Cladocera.
- 4 - Calculate the Biodiversity Indices depending on morphology and genetic methods.

## **1 – 3 : Literature Review :**

### **1-3-1 :Biodiversity**

To enumerate the true diversity on the planet, we must first define biodiversity and understand how we might assess it. Biodiversity can be discussed and studied in different ways, including species diversity, genetic diversity, and ecosystem diversity (Spangenberg, 2007). Mostly, biodiversity is assessed using species as the units of measure (De Queiroz, 2007). Biological diversity or biodiversity are expressions that refer to the variety of life on the planet or to the property of living systems to be distinct. It includes plants, animals, microorganisms, ecosystems and ecological processes in a functional unit (Dias and Coradin,1999). The diversity is regarded as an indication of the ecosystem well-being. It indicates directly the protection of certain place, the higher the value, the larger the biodiversity of the ecosystem in question (Ricklefs, 1990).

The Prior term of Biodiversity is most commonly used to replace the more clearly defined and long established terms, species diversity and species richness (Walker, 1992). There are alternate term for biodiversity which define as the totality of genes, species and ecosystems of a region (Tor-Bjorn, 2001 ; Davis, 2011). In 1992, United Nations Earth Summit defined biological diversity as the variability among living organisms from all sources including inter alia, terrestrial, marine and other aquatic ecosystems and the ecological complexes of which they are part, this includes diversity within species, between species and of ecosystems (Hawksworth, 1996). As well as, The Food and Agriculture Organization of the United Nations (FAO) defines biodiversity as the variability that exists among living organisms (both

within and between species) and the ecosystems of which they are part (Bélanger and Pilling, 2019).

Biodiversity generally tends to cluster in hotspots (Myers *et al.*, 2000) which is defined as a region with a high level of endemic species that have experienced great habitat loss. The term hotspot is introduced in 1988 by Norman Myers, while hotspots are spread all over the world, the majority are forest areas and most are located in the tropics (Myers, 1988).

Biodiversity is an important aspect of the natural world and a cornerstone of resilience in ecosystems (Folke *et al.*, 2004). Despite its importance, some investigators examining the diversity of organisms have found that species concepts based on morphology have underestimated the actual biodiversity of systems (Kieneke *et al.*, 2012 ; Gebiola *et al.*, 2012). Biodiversity is usually explored at three levels , genetic diversity, species diversity and ecosystem diversity. These levels work together to create the complexity of life on Earth (Sahney *et al.*, 2010 ; Campbell, 2003). Other investigators examining the genetic diversity of freshwater organisms have contributed to a variety of scientific discussions including those on species concepts and species delimitation (Pons *et al.*, 2006; De Queiroz, 2007 ; White *et al.*, 2014), concerns about the accurate estimation of biodiversity (Isaac *et al.*, 2004 ; Zachos *et al.*, 2013) and the relationships between diversity and geography (Szpiech *et al.*, 2008 ; Spitzer, 2014). Nevertheless, much remains to be learned about the patterns and dynamics of genetic diversity. Other investigators have found evidence for cryptic species, which are morphologically similar but genetically appear to be separately evolving lineages when described by methods such as an arbitrary threshold of genetic divergence (De Queiroz, 2007).

Genetic methods are particularly useful for examining biodiversity of freshwater invertebrates for many reasons (Mynott *et al.*, 2011; Webb *et al.*, 2012 ; Stein *et al.*, 2014). For example, Carter and Resh, (2013) showed that the species-level identifications generally require adult specimens of invertebrate rather than the aquatic larval forms that are the life stage collected in many bioassessment and biomonitoring programs to assess the quality of freshwater habitats. As well as, McCafferty *et al.*, (2008) explained that morphological identification of freshwater invertebrates, particularly the immature stages is difficult because many are small or have numerous instars, dynamic life histories and systematic uncertainties .

Many investigators such as (Parmesan, 2006 ; Schlacher *et al.*, 2008) showed that shifts in biodiversity may indicate climatic or anthropogenic environmental change or impacts on a variety of temporal and spatial scales. Changes in biodiversity can affect ecosystem function (Hooper *et al.*, 2005 ; Isbell *et al.*, 2011) and food web dynamics (Cardinale *et al.*, 2002). At the same time, human impacts can increase diversity at multiple scales through mechanisms including changing disturbance regimes (Devictor and Robert, 2009) and addition of exotic species (Bruno *et al.*, 2004). Despite a growing number of local scale long-term biodiversity studies like (Vellend *et al.*, 2013 ; Dornelas *et al.*, 2014), major gaps in the understanding of biodiversity change outside of developed nations and for underrepresented biomes seriously impede our ability to accurately quantify biodiversity change across the planet (Gonzalez *et al.*, 2016).

Finally, Biodiversity can be expressed in different levels: diversity of genes, species and ecosystems. It can be appreciated by the number

(wealth) of different biological categories, relative abundance (evenness) of these categories and also for variability in local level (alpha diversity), the complementarity between biological habitats (beta diversity) and variability between landscapes (gamma diversity) (Whittaker, 1972).

### **1-3-2 :Factors affecting Biodiversity**

Defining the factors that affect biological diversity is a major goal of ecologists principally in situations of environmental disturbance caused by human activities. This knowledge supports conservation actions and proposals for environmental monitoring and management to mitigate biodiversity loss (Butchart *et al.*, 2010) and to maximize the use of environmental services through the maintenance of ecosystem processes (Lindenmayer and Likens, 2010 ; Simoes *et al.*, 2013).

Aquatic environments, mainly rivers are among the most threatened natural ecosystems worldwide due to the damming of their courses including the construction of reservoirs (Nilsson *et al.*, 2005 ; Dudgeon *et al.*, 2006). Fresh waters face an unprecedented array of environmental threats, many of which are anthropogenic, as we enter the first decades of the 21<sup>st</sup> century (Covich *et al.*, 2004). Many studies such as (Covich *et al.*, 2004; Woodward *et al.*, 2005; Balvanera *et al.*, 2006; Brooks *et al.*, 2006 ; Schindler, 2007) have proven that the Eutrophication, climate change, toxic pollution, acidification, exotic species, overharvesting and habitat destruction can all have catastrophic impacts on biodiversity and ecosystem functioning and ultimately on the supply of 'goods and services' of value to humans. However, it is still far from clear how stressor impacts are manifested across different levels of biological organization (e.g. populations, communities, food webs and ecosystems) .

Zooplankton are susceptible to variations in a wide number of environmental factors including biotic factors such as food availability (algae, bacteria), predation by fish and invertebrates (Patalas and Salki, 1993) and abiotic factors such as temperature, pH, DO and electrical conductivity (Verma and Agarwal, 2007). 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 (Delaram *et al.*, 2017). Many studies showed that the Zooplankton communities often respond quickly to environmental change because most species have short generation times (usually days to weeks in length). For examples, Ivanova and Kazantseva, (2006) studied the combined influence of lake surface area, pH and total dissolved solids on the zooplankton communities in lakes and showed that extremely high or extremely low values of pH and TDS inhibit lake zooplankton; Okogwu and Nwani, (2009) studied physicochemical variables and zooplankton of Ologe Lagoon and showed there is increase in zooplankton abundance and diversity with increase in rainfall; Jonathan *et al.*, (2010) examined the association between zooplankton species richness in lakes and environmental variability on inter annual, seasonal and shorter time scales, as well as long-term average conditions and suggest that temporal fluctuations in the chemical variables such as pH and DO tend to exclude zooplankton species while temperature variability promotes greater richness; Julies and Kaholongu, (2013) studied the relationship between dissolved oxygen and distribution of zooplankton of the Namibian coast and showed that the dissolved oxygen does not have a direct effect on the diversity of zooplankton but an indirect effect and it is the interplay and

interaction between several biological and environmental factors that affects zooplankton assemblage composition; Malik and Shikha, (2016) examined the diversity and distribution pattern of zooplanktons in Bhimtal lake along with evaluation of the relationships among different zooplankton groups and abiotic factors and found positive correlation between zooplankton growth, water temperature and pH while the growth is adversely affected with increasing alkalinity and dissolved oxygen; Claire Ann Malaras *et al.*, (2019) analyze the physico-chemical characteristics of Bitanag creek and to assess zooplankton diversity and showed significant difference on the abundance and diversity of zooplankton between sites. Thus, the distribution and abundance of zooplankton were greatly influenced by the condition of its physico-chemical such as conductivity, water temperature, TDS, turbidity, salinity, water current, COD, DO, pH and nutrient chemistry; Abdullah *et al.*, (2020) studied the temporal distribution of zooplankton communities in coastal waters of the northern bay of Bengal, Bangladesh and showed that species evenness and diversity significantly correlated with salinity, transparency and TDS; Vilas-Boas *et al.*, (2021) assessed the single and combined effects of these stressors on zooplankton communities collected from a Mediterranean coastal lagoon and showed that temperature can influence the direct and indirect effects of salinity on zooplankton communities and diversity; Neelgund and Kadadevaru, (2021) study the seasonal variation in zooplankton abundance at Kadasgatti minor irrigation tank (MIT) located in India and observed the highest zooplankton abundance in summer while minimum in monsoon season.

Multiple studies have found that biodiversity declines have increased in the last century due to human impacts (Barnosky *et al.*, 2011 ; Ceballos *et al.*, 2015) and these declines in biodiversity are likely to alter the

functioning of ecosystems (Naeem *et al.*, 2012). It is therefore essential to understand how global change is likely to impact biodiversity so that researchers and policy makers can make better decisions regarding conservation and restoration of ecosystems, habitats and species (Bellard *et al.*, 2012). Not all species are likely to respond to global changes in the same way. For example, cosmopolitan species with broad biogeographic distributions and tolerances to a wide variety of environmental conditions might be less susceptible to the effects of climate change. However, endemic species those with narrower biogeographic distributions tend to have limited geographic ranges because of their restricted physiological tolerances to abiotic conditions (Malcolm *et al.*, 2006). As temperatures continue to rise, Malcolm *et al.*, (2006) indicated to that endemic species have been declining in abundance, causing changes in community assembly and structure.

Climate change has proven to affect biodiversity and evidence supporting the altering effects is widespread such as increasing atmospheric carbon dioxide (Ainsworth and Long, 2004), acidifying (Doney *et al.*, 2009) and temperature affects species ranges (Lovejoy and Hannah, 2005). In 2004, an international collaborative study on four continents estimated that 10 % of species would become extinct by 2050 because of global warming (Brown, 2004).

In general, The threats to global freshwater biodiversity can be grouped under five interacting categories: overexploitation, water pollution, flow modification, destruction or degradation of habitat and invasion by exotic species (Rahel, 2002; Postel and Richter, 2003; Revenga *et al.*, 2005). Overexploitation primarily affects vertebrates, whereas the other four threat categories have consequences for all

freshwater biodiversity from microbes to megafauna. Habitat degradation is brought about by an array of interacting factors. It may involve direct effects on the aquatic environment (such as excavation of river sand) or indirect impacts that result from changes within the drainage basin. Flow modifications are ubiquitous in running waters (Nilsson *et al.*, 2005). Widespread invasion and deliberate introduction of exotic species adds to the physical and chemical impacts of humans on fresh waters, in part because exotics are most likely to successfully invade fresh waters already modified or degraded by humans (Bunn and Arthington, 2002 ; Koehn, 2004).

### **1-3-3 : Biodiversity Indices**

Various diversity indices are used to determine the distribution of invertebrates related to habitat quality. Diversity index is a statistical method which is planned to evaluate the variety of a data group consisting of different types of components. Features of a population such as number of existing species (Richness), distribution of individuals equally (Evenness) and total number of existing individuals underlie the basis of diversity indices (Wilhm and Dorris, 1968 ; Allan, 1975). Thus, any changes in any of these three features will affect the whole population, so that the diversity indices depending upon these features are used effectively to determine the changes in a population (Mandaville, 2002 ; Dügel, 1995).

Undisturbed habitats are characterized by high diversity and number of different species. Rich nutrient sources occur with the contributing the organic contaminants to a stream. While the pollution is increasing, the number of tolerant species increases and sensitive species begin to disappear. Both diversity and abundance in a population are decreasing

(Hellawell, 1986). Important findings about the structure of a stream are gained by using the diversity indices. Some diversity indices such as Shannon Diversity Index, Simpson Index, Margalef Index and evenness indices such as Evenness Index are used in this study. In most of the published studies, species variation as a biological diversity (DeLong, 1996 ; Williams, 2004) consists of two components: species richness and evenness (Omernik, 2003 ; Liu *et al.*, 2008) which can be regarded as sustainability and healthiness factors in the rangeland ecosystems (Williams, 2004 ; Jouri, 2010). Species richness is the total number of taxa collected, whereas evenness depends on the relative abundance of individuals representing each taxa (Begon *et al.*, 1990). Different factors have been already affected by the community's diversity as follows: sampling size and rate, destruction and human activity, farming, topographical factors, succession, underground waters, rivers and livestock grazing (Afshani *et al.*, 2009).

Biodiversity represents the variety and heterogeneity of organisms or traits at all levels of the hierarchy of life from molecules to ecosystems. Typically, the focus is on species diversity but other forms of diversity such as genetic diversity are also important and informative. Even after deciding which form of diversity to measure, quantifying biodiversity remains problematic because there is no single index that adequately summarizes the concept (Purvis and Hector, 2000). Richness or the number of species or attributes present, is the simplest metric used to represent diversity (Whittaker, 1972) and it remains the most commonly applied (Magurran, 2004). Intuitively, species or trait abundance is also important for diversity and the proportional abundance of species can be incorporated into indices representing diversity. The simplest of these indices are proposed by Berger and Parker, has an analytical relationship

with the geometric series of the species abundance model (Caruso *et al.*, 2007) and reports the proportional abundance of only the most abundant species in the population (Berger and Parker, 1970). Foremost among these are the Shannon's diversity ( $H'$ ) and Simpson's diversity ( $D1$ ) indices which differ in their theoretical foundation and interpretation (Magurran, 2004). Shannon's diversity ( $H'$ ) has its foundations in information theory and represents the uncertainty about the identity of an unknown individual. In a highly diverse (and evenly distributed) system, an unknown individual could belong to any species leading to a high uncertainty in predictions of its identity. In a less diverse system dominated by one or a few species, it is easier to predict the identity of unknown individuals and there is less uncertainty in the system (Shannon, 1948). This metric is common in the ecological literature despite its abstract conceptualization (Magurran, 2004).  $D1$  is the complement of Simpson's original index and represents the probability that two randomly chosen individuals belong to different species (McCune and Grace, 2002).  $D2$  is closely related to  $D1$ , being the inverse of Simpson's original index (Simpson, 1949). Both of these transformations serve to make the index increase as diversity intuitively increases and although both are used,  $D2$  is more common (Magurran, 2004).

One of the main problems with diversity indices are that the components are interrelated and often compete with each other. The indices of diversity thus try to put in adequacy the data on abundance with the number of species of a community in only one number, from which the structure of the community can theoretically be apprehended. The diversity of the species should be a function of the number of species and the distribution of the abundance of these species. It is possible for diversity to grow while the number of species decreases if the distribution

increases. Diversity under this definition becomes an aspect of the structure of the community in which the rare species are structurally of no importance. Diversity indices can be called "ecological indices" or "species diversity" (for a meaningful difference between diversity and biodiversity (Marquès, 2001).

### **1-3-4 : Biodiversity of Zooplankton**

Zooplanktons are free swimming, microscopic consumers of aquatic ecosystem. They play important role in aquatic ecosystem and food chain. Zooplanktons are valuable food sources for fishes and other aquatic animals. They are responsible for eating millions of little algae that may otherwise grow to an out of control state (Wilkinson, 2011). Zooplankton community is composed of both primary consumers and secondary consumers. They provide direct link between primary producers and higher trophic levels such as fishes (Ali, 2010). Because of their central position in aquatic food webs, zooplankton communities are reliable indicators of aquatic ecosystem. Some of the notable workers who have investigated in detail various aspects of zooplanktons were Kim *et al.*, (2000) who demonstrate that zooplankton community especially rotifers played an important role in transferring both bacterial and phytoplankton carbon to higher trophic levels; Tan *et al.*, (2004) who describe the composition, abundance, distribution and grazing impact of dominant components of the zooplankton community in the Pearl River Estuary (PRE) and showed that the grazing impact of zooplankton changed seasonally and spatially. Ugale *et al.*, (2010) stated that zooplanktons are at the top of the aquatic food chain and that zooplankton diversity is the most crucial biological criterion for determining water quality. As well as, Howick and Wilhm, (1984) showed that presence of zooplankton in

water body helps to increase economically important fish populations and they play a major role in energy transfer between phytoplankton and fish.

Zooplankton diversity is also influenced by the presence of phytoplankton for outcome of nutrients for better survival in aquatic life (Ali *et al.*, 1985). Fluctuations in zooplankton community composition and species diversity can indicate their distribution in any environment (Caroni and Irvine, 2010 ; Kehayias *et al.*, 2014). In addition, the distribution and diversity of zooplankton in aquatic ecosystem depend on the standing limnological properties of water (Kumar *et al.*, 2010). It is difficult to understand the biological phenomenon fully because the chemistry of water reveals much about the metabolism of the ecosystem and explain the general hydrobiological relationship (Basavaraja *et al.*, 2011). For these reasons, zooplankton communities are commonly used for experiments to test many ideas in community ecology including the relationships between diversity and stability or ecosystem function (Downing and Leibold, 2002), meta-community theory (Forbes and Chase, 2002; Howeth and Leibold, 2008), as well as alternative states theory of the ecosystem (Chase, 2007).

In zooplankton, Diel migration can vary temporally and spatially among developmental stages of species, and between co-existing species (Bohrer, 1980 ; George, 1983). There are three types of diel migrations that commonly occur within species of zooplankton: horizontal, vertical and reversed vertical migration (Iglesias *et al.*, 2007 ; Meerhoff *et al.*, 2007).

Zooplankton and other freshwater invertebrates have several life history characteristics that could promote rapid dispersal. First, a wide variety of invertebrates employ some mode of asexual reproduction (Bell,

1982) which allows clonal propagation and also avoids the problem of mate limitation. Because most Rotifera and Cladocera reproduce parthenogenetically (formation of eggs not requiring fertilization), a single individual can found a local population. Second, most zooplankton employ diapause to persist in unstable environments. Bdelloid rotifera undergo cryptobiosis and monogononts form diapausing eggs (Wallace and Snell, 2001). Diapausing eggs are also widespread in Cladocera, other branchiopods and freshwater species of calanoid copepods and ostracods (Ca´ceres, 1996). Diapausing eggs are known to resist freezing, drying and digestion by predators (Dodson and Frey, 2001). The vectors for transporting zooplankton to colonize new habitats including wind (Cohen and Shurin, 2003), flowing water (Magnuson *et al.*, 1998), by animals (Bilton *et al.*, 2001) and Human effects (Stelzleni-Schwent, 2000).

According to Wallace and Snell, (1991), freshwater zooplanktons is divided into three major groups (Rotifera, Cladocera and Copepods). Whereas Johnson and Allen, (2005) divided the freshwater zooplanktons on their size into Macrozooplankton (Cladocera and Copepods) and Microzooplankton (Rotifera and nauplii of Copepods). In present study, we have focused on the Rotifers and Cladocerans.

### **1-3-4 -1 : Rotifera:**

The rotifera known as “wheel animalcules” are a phylum of small (50 – 2000 µm), primarily freshwater zooplankton. There are approximately 2200 different rotifera species identified. Currently, their taxonomy is undergoing change. Seisonidea, Bdelloidea, and Monogononta are the three classes that make up the phylum Rotifera (Barnes *et al.*, 2001). With over 1500 species, the Monogononta is the biggest group, followed

by the Bdelloidea with roughly 350 species. Seisonidea are only known in two genera and three species (Aisha *et al.*, 2000).

Kingdom: Animalia  
--Phylum : Rotifera  
----Super class: Eurotatoria  
-----Class Bdelloidea  
-----Class: Monogononta  
----Super class: Pararotatoria  
-----Class: Seisonidea

Collectively, these groups include an estimated more than 1900 species capable of inhabiting fresh water, though this may be an underestimate, given incomplete taxonomic knowledge and the likely presence of cryptic species (Thackeray, 2021). Rotifera are widely recognized as being important components of freshwater ecosystems and whether this assessment is based on numbers or biomass, their contribution to trophic dynamics in these waters is striking (Wallace *et al.*, 2006) . Rotifera can be found across a wide variety of freshwater habitats from large lakes and reservoirs to small ponds, temporary puddles, birdbaths and even the interstitial waters within sediment layers and films of water on mosses and liverworts (Thackeray, 2021).

Rotifera is a group of primary freshwater invertebrates. They commonly occur in densities up to 1000 individuals per liter and are important filter-feeders on algae and bacteria (Wallace *et al.*, 2006). They are minute metazoans characterized by the presence of an anterior ciliated corona, a stiff body wall named lorica bearing variable appendages and a specialized pharyngeal organ, the mastax, containing hard elements termed trophi (Seger, 2008) which composed of five main pieces fulcrum, rami, incus, unci and manubria, it is great taxonomic significance for the distinction of families, genus and species (Abd Al-Rezzaq,2014). In some forms, classification to species may be ascertained

based on trophi alone (Wallace, 2002). In many species, the corona is developed as two concentric rings of cilia (trochus and cingulum) that beat in a metachronous pattern. This action resulting in an illusion of a rotating wheel, informs the allusion used to conceive the phylum's etymon (L., rota + ferre, wheel-bearers) (Wallace, 2002). Especially, the rotifer's small size capability of phenotypic plasticity and highly adaptable masticatory apparatus are important elements explaining the success of the group (Segers, 2008). Their propagules consist of single, hard-shelled, and durable encapsulated cysts (Monogononts) or anhydrobiotic individuals (Bdelloids). These propagules being small and drought-resistant, makes rotifers perfectly adapted to passive, aerial or phoretic dispersal. Monogononts and Bdelloids reproduce parthenogenetically. In monogononts, periods of parthenogenetic reproduction are interspersed with sexual phases (heterogony) but Bdelloids are unique in being the most diverse group of metazoans in which reproduction is by diploid, mitotic parthenogenesis only. The combination of their high dispersal capacity and their parthenogenetic reproduction enabling them to establish or renew a population starting off from a single resting stage, and to reach high effective population sizes relatively quickly makes them theoretically superbly apt (re)colonizers. The ability of many Bdelloids to shift from active to anhydrobiotic stage enables them to live in particularly ephemeral even predominantly dry conditions such as lichens or mosses. Bdelloids, however, can only be identified while alive and need to be examined during feeding and creeping (Segers, 2008).

The majority of the species are monogononts, while Bdelloids were represented only by *Rotaria* sp. The largest fraction (76%) belonged to the families: Lecanidae > Collurelidae > Brachionidae > Trichocercidae > Notommatidae > Flosculariidae. Bonecker *et al*, (2005) examined the

diversity and abundance of the rotifera in different environments of the upper Parana river floodplain in Brazil and showed that the highest species richness is in rivers and open floodplain lakes, the highest abundances in the isolated floodplain lakes and the highest values of species diversity in the channels especially during the rainy period,  $\beta$ 2-diversity values are higher in the channels especially during the dry period; Tijare and Thosar, (2008) studied the rotifera diversity in three lakes of Gadchiroli in the North-Eastern part of Maharashtra state of India and indicated that the population of rotifers is maximum in winter months while minimum in the summer and it is observed that Brachionodae is the dominant family of rotifera. In all 11 genera and 25 species are recorded from the three lakes as well as the maximum diversity of rotifera is observed in Gadchiroli lake due to its eutrophic condition. Sulehria *et al.*, (2009a) studied the rotifera species richness, diversity and evenness in a fish pond and observed the significant differences in the physico-chemical characteristics of water such as temperature, pH, DO and conductivity as well as rotifera diversity in different seasons. Contreras *et al.*, (2009) studied seasonal changes in the rotifera diversity from a tropical high altitude reservoir in Mexico and found *Keratella*, *Polyarthra* and *Trichocerca* were the dominant genera in addition to the highest diversity value is observed during January, while the lowest during March, as well as, they showed that temperature, dissolved oxygen, water column depth and chlorophyll *a* have combined effects on the abundance of dominant rotifer species. The highest rotifer density was observed in April and lowest is recorded in January; Sulehria and Malik, (2012) determined the population dynamics of rotifera in Balloki Head Works (Pakistan) and records new twenty four species. Three families dominated (Brachionidae, Lecanidae and Trichocercidae) with *Brachionus*, *Lecane* and *Trichocerca* respectively being the most

important genera. Rotifera density is positively correlated with water temperature being the highest in summer and the lowest in autumn/winter. Conductivity also had positive correlation with rotifera density. Negative correlation was however observed with pH, dissolved oxygen and total hardness. Finally, Dastgeer *et al.*, (2020) explored seasonal distribution and diversity of rotifera at Marala Headwork lake and selected fish ponds of district Sialkot, Punjab, Pakistan. *B. angularis*, *B. calyciflorus*, *F. minuta*, *K. cochlaeris*, *K. valga*, *L. lunaris* and *P. roseola* are common species recorded at both habitat types. Brachionidae is the most prevalent family throughout the year at both habitat types and recorded significantly different diversity in fish ponds and Marala lake during different seasons, as well as diversity indices showed higher population diversity of rotifera in Marala Lake than fish ponds.

### 1-3-4 -2 : Cladocera :

Cladocera "water fleas" are primarily freshwater small-sized (0.2– 6 mm and up to 18 mm in single case of *Leptodora kindtii*) branchiopod crustaceans, inhabiting pelagic, littoral, and benthic zones. Four cladocera orders are recognized : Anomopoda, Ctenopoda, Onychopoda, and the monotypic Haplopoda (Fryer, 1987).

Kingdom: Animalia  
 --Phylum : Arthropoda  
 ---- Subphylum : Crustacea  
 ----- Class : Branchiopoda  
 -----Order : Anomopoda  
 ----- Suborder: Cladocera

Most species occur in continental fresh or saline waters, although two ctenopods and several onychopods from the family Podonidae are truly marine and a few more ctenopod, anomopod and onychopod species occur in brackish waters. The trunk and appendages of most cladocera

(Anomopoda and Ctenopoda) are enclosed in a bivalved carapace. Tagmosis of the body is obscure (except in *Leptodora kindtii*, the single representative of Haplopoda), a single eye and ocellus are usually present. Antennules are uniramous, while antennae are biramous (except in females of *Holopedium*), natatory with 2–4 segments per branch. Four to six pairs of trunk limbs are either mostly similar in shape (Ctenopoda, Onychopoda, Haplopoda) or modified individually for various functions (Anomopoda) (Forro *et al.*, 2008). Anomopoda have five pairs of thoracic limbs, though occasionally having six pairs (Khanna, 2004). While the abdomen region gradually combines with the anterior of the trunk, the head of the anomopoda lacks a distinct separation from the trunk and the posterior (Dumont and Negrea, 2002).

Cladocera represent one of the major taxonomic groups colonizing freshwaters, both in terms of their species diversity and secondary production (Walseng *et al.*, 2006 ; Lemke and Benke, 2009). Most species are filter-feeders but onychopods and haplopods are predatory. They usually reproduce by cyclical parthenogenesis (but asexual lineages are known as well) and populations are mostly dominated by females. Sexual dimorphism is normally rather distinct. Sexually produced diapausing eggs are resistant to desiccation and other unfavorable conditions and may even survive passage through the digestive track of birds (Figuerola and Green, 2002); thus, they are important propagules for passive dispersal. Cladocera undergo parthenogenesis in suitable environments and form large populations. However, sexual reproduction of cladocera occurs under bad conditions and fertilized eggs are produced (Jiang and Du, 1979 ; Benzie, 2005).

Cladocera are important components of the food chain in aquatic ecosystems (Forro *et al.*, 2008) which forms the second trophic level of the food chain in the aquatic environments, they are play an important component of the faunal biodiversity of the freshwater ecosystems. They feed on phytoplankton, bacteria and detritus and thus transporting energy from plant materials into animal tissue and in turn constitute the basic food for higher animals including fishes particularly their larvae (Goswami, 2004). Some species of this group have a wide variation in size structure due to genetic factors (Black,1980) and environmental factors as temperature (Kappes and Sinsch, 2002a), food availability (Kappes and Sinsch, 2002b), pH (Korosi *et al.*, 2008) and acidification (Labaj *et al.*, 2016). *Bosmina* genera is a group of small cladocera filter feeders with variations in body size, shape, length of antennules and mucron (Labaj *et al.*, 2016 ; Hang and Hanazato, 2003). Cladocera are found in lakes, ponds, slow-moving streams and rivers. They are found from the Arctic to the Southern Oceans. Some cladocera are benthic, other species live on sediments or on vegetation, virtually anywhere water is present including swamps, puddles, ditches, and ground water (Oltra and Todolr, 1997; Dodson and Frey, 2001).

Cladocera is an Arthropoda, Crustacea, Class Branchiopoda which is divided into 4 suborders, 11 families, 80 genera and 400 species (Dumont and Negerea, 2002). Chydorids are a distinct group of organisms within the family Chydoridae. Many taxa within the family Chydoridae are important indicators for temperature, pH and nutrient dynamics (Whiteside and Swindoll, 1988). The taxa differ greatly with seasons and habitat types (Whiteside, 1974 ; Whiteside *et al.*, 1978).

In Europe, relatively warm climate-preferring taxa belonging to small *Alona* groups such as *Alona guttata* were predominantly recorded from the low-altitude temperate regions (Korhola, 1999), while the cold climate-preferring taxa such as *Acroperus harpae* and *Alona affinis* predominantly occurred in sub-arctic and high-altitude sites (Hofmann, 2000). Temperature also plays a profound role in the life history of these cladocerans (Nevalainen and Luoto, 2010). Adult females produce a large number of parthenogenetic females during spring (Frey, 1982). However, under adverse climatic conditions (e.g. dry period) gamogenesis occurs when females produce haploid reproductive or resting eggs (Frey, 1982).

Species of the genus *Bosmina* Baird, 1845 (Anomopoda: Bosminidae) is small cladocera (approximation 0.254 ~ 0.319 mm) that exist in lakes and reservoirs in every biogeographical region (Goulden and Frey, 1963; Havens and Decosta, 1985). The head of *Bosmina* is evenly rounded anteriorly, ventrally it protrudes into a fairly long ventrally directed rostrum looking like it is a part of the body (Havens and Decosta, 1985). The genus not only purifies the water by the highly efficient filtration of bacteria, organic detritus and phytoplankton (top-down control) in the water bodies but also is rich in protein and fat which can be used as bait for fish (bottom-up control). Therefore, *Bosmina* plays an important role in energy flow, the carbon cycle and the nutrient salt mineralization of waters (Huibin *et al.*, 2015). Moreover, *Bosmina* may achieve high biomass under a variety of conditions that are unfavorable for other cladocera. And its dominant position has become increasingly more apparent in recent decades (Yun *et al.*, 2016).

*Daphnia* spp., commonly called water fleas, are small planktonic crustaceans (1- 5 mm long), suborder of Cladocera that are ubiquitous in

freshwater aquatic environments (Tatarazako and Oda, 2007). *Daphnia* body is enclosed by shell known as the carapace. It has a double wall, between which hemolymph flows and which is part of the body cavity. *Daphnia* have up to 10 pairs of appendages which are antennules, antennae, maxillae and mandibles followed by 5 or 6 limbs on the trunk. The limbs form an apparatus for feeding and respiration. At the end of the abdomen is a pair of claws (Freyer, 1991; Lampert and Sommer, 1999). They occupy a key position in the aquatic food chain as the intermediate link between primary and secondary productivity (Miner *et al.*, 2012) and they also serve as model species in environmental toxicology because of their high sensitivity to water quality (Dodson and Hanazato, 1995). Under favorable conditions, *Daphnia* reproduces by parthenogenesis in which clonally offspring are produced by females asexually (Hebert and Ward, 1972). Induced by environmental changes, some females can produce haploid eggs that need fertilization by males. The fertilized eggs remain in a dormant state being enclosed by several protective membranes, the ephippium and can survive strict conditions for many decades before hatching (Caceres and Tessier, 2004; Mark and Solbe, 1998).

Many investigators have been interested in studying of Cladocera, as they represent one of the main taxonomic groups that colonizing the freshwater both in terms of their species diversity and secondary production. Among these studies, Borges and Pedrozo, (2009) investigated the richness, diversity and abundance variation of the zooplankton community of Jacuí River Delta as well as the abiotic variables analyzed were: fluviometric level, water temperature, Secchi transparency and chlorophyll-*a*. and showed that richness and Shannon diversity index are higher at the maximum fluviometric level influenced

by washout of species associated to the environment and by probable competition reduction; Diniz *et al.*, (2013) described the composition and structure of cladocera of littoral areas with and without macrophytes from a temporary shallow pond in the Caatinga of Pernambuco state (Brazil). Twenty-two species are recorded with two new occurrences: *Chydorus* cf. *brevilabris* and *Macrothrix superaculeata*. The species richness of non-planktonic cladocerans was higher than that of planktonic ones. Warmer months had higher densities and richness of cladocera and the months of greatest rainfall had lower richness especially for the Chydoridae family; Sinev and Yusoff, (2015) evaluated the fauna of cladocera of Sabah state of Malaysia, Borneo Island for the first time and revealed 31 species of cladocera including three species of Sididae, one species of Daphnidae, one species of Moinidae, four species of Macrothricidae, two species of Ilyocryptidae, and 20 species of Chydoridae; Neretina *et al.*, (2017) studied the Cladocera of Ethiopia and found 63 species of Cladocera in water bodies: 26 species of Chydoridae, 18 of Daphniidae, 6 of Moinidae, 5 of Macrothricidae, 5 of Sididae, 2 of Ilyocryptidae and 1 of Bosminidae. The highest cladocera diversity (39 taxa) is in the mountainous lake Tana. Diverse lakes, rivers and temporary pools have lower species diversity (24 taxa in total). Water bodies of Bale Mountains are characterized by significantly low cladocera diversity and high proportion of endemic taxa; Francisco and Elmoor-Loureiro, (2018) investigated the cladocera from the Upper Xingu River Basin, Central Brazil and found eighteen species belonging to three families. Also describe a new and highly specialized genus of Chydoridae: *Kisakiellus aweti* *gen. nov.*, *sp. nov.* which shares some morphological traits with members of Chydorinae; Tiang-nga *et al.*, (2020) studied the cladocera diversity in lake Kud-Thing, a Ramsar site of Bueng Kan Province, Thailand and recorded 58 species belonging to 38 genera. The most

species rich family is Chydoridae (32 species, 55%) followed by Sididae (8 species, 14%) and Macrothricidae (8 species, 14%) while only a single species of Ilyocryptidae is observed; Klemetsen *et al.*, (2020) examined the diversity, abundance and life histories of littoral chydorids in subarctic European lake and showed that three out of eight recorded species of Chydorids are common with abundance minima in late summer and maxima in autumn. *Eurycercus lamellatus* and *Acroperus harpae* are monocyclic whereas *Chydorus sphaericus* is seemingly acyclic.

### **1-3-5 : Genetic Diversity and Phylogeny**

Genetic diversity of organisms have contributed to a variety of scientific discussions including those on species concepts and species delimitation (DeSalle *et al.*, 2005; Pons *et al.*, 2006; De Queiroz, 2007 ; White *et al.*, 2014), concerns about the accurate estimation of biodiversity (Isaac *et al.*, 2004 ; Zachos *et al.*, 2013) and the relationships between diversity and geography (Hughes *et al.*, 2003; Szpiech *et al.*, 2008 ; Spitzer, 2014). Nevertheless, much remains to be learned about the patterns and dynamics of genetic diversity.

An understanding of the dynamics of genetic diversity in freshwater ecosystems is especially important because of recent biodiversity losses in these environments (Jenkins, 2003; Dudgeon *et al.*, 2006 ; Moyle *et al.*, 2011). Increased genetic structure which is one aspect of population diversity can contribute to a stabilizing portfolio effect. A portfolio effect describes the increased production and resiliency of a population that comprises spatially or temporally segregated subpopulations with diverse adaptations (Carlson and Satterthwaite, 2011). The portfolio effect can be an important component in the stability and survival of freshwater species (Schindler *et al.*, 2010).

Phylogeny refers to the evolutionary history of the development of a species or of a taxonomic group of organisms. The phylogenetic relationships are depicted in the form of a phylogenetic tree, i.e. a tree diagram depicting how one taxon is closely or distantly related to another taxon. The tree diagram demonstrating phylogenetic relationships is based on the molecular sequencing data analyses as well as on morphological data matrices (Kutschera, 2016). The term "Phylogeny" is derived from a combination of Greek words Phylon stand for "tribe" or "clan" or "race" and genesis means "origin" or "source". The term can also be applied to the genealogy of genes derived from a common ancestral gene (Patwardhan *et al.*, 2014).

While phylogeny is concerned with the evolutionary relatedness and history of organisms, it is not concerned with the identification of these organisms. Rather, it is the main concern of taxonomy. To be more precise, taxonomy is the branch of science that is concerned chiefly with identifying, naming and classifying organisms. It puts organisms into taxonomic ranks, e.g. domain, kingdom, phylum or division, class, genus and species. Nevertheless, the classification is based on morphology and phylogeny of organisms and phylogenetics provides information for use during the identification and classification of organisms. So while taxonomy is concerned chiefly with the identification and classification of organisms, phylogeny provides data for such purpose and a phylogenetic classification would be one that is largely based on molecular phylogeny data (Doolittle, 1999 ; Panchen, 1992).

Molecular phylogeny, a branch of phylogeny makes use of molecular sequencing to study evolutionary relationships and histories. Molecular

sequencing, in this regard is a useful tool to understand the phylogenies of different taxa. Basing the relatedness on morphology, anatomy, physiology and life cycles can be confounding. There are instances wherein some traits are easily identified as either similar or disparate. However, there are also instances when their similarities and disparities are vague. Furthermore, there are also situations when two species seem to belong to a common taxon but after analyzing their genomes they turned out to be evolutionary distant (Yokoyama, 1994 ; Gray *et al.*, 1999).

Fortunately, more advanced tools for study and research have become available and they have provided scientists a more reliable basis for determining and analyzing phylogeny. Nucleic acids such as DNA and RNA, store and retain certain genetic information that scientists use as hints of plausible evolutionary origins and history. It is because these biomolecules are heritable (Young, 2016).

With significant challenges to traditional morphological methods, the use of molecular techniques must be further investigated including DNA barcoding and metabarcoding approaches (Bucklin, 2010 ; Cristescu, 2014). DNA barcoding is a standardized approach using molecular data from one or a few specific gene regions, to identify a specimen as belonging to a particular species (Hebert *et al.*, 2003a,b). Metabarcoding utilizes the DNA barcoding approach but instead of applying the method to a single specimen at a time, it uses DNA barcoding across large collections of specimens at the same time (Cristescu, 2014). A metabarcoding approach uses the high-throughput capabilities of next generation sequencing platforms in an attempt to assess all diversity for a large taxonomic group within a mixed-species sample. The performance

of DNA barcoding and metabarcoding among widely distributed geographically and taxonomically diverse zooplankton such as crustaceans has been largely untested (Young, 2016).

Biodiversity is currently under threat and our perception of species loss is highly dependent on accurate estimates of species richness. However, estimates of species richness are often impaired by the occurrence of cryptic species (i.e. species that are impossible or difficult to distinguish based on their morphology) in diverse groups such as rotifers (Go´mez and Snell, 1996). Evidence on cryptic species diversity in rotifera, subclass Monogononta is growing and challenges our understanding of rotifer biodiversity. In Monogononta, cryptic species complexes have been described for species such as *Brachionus plicatilis* (Go´mez and Carvalho, 2000; Go´mez *et al.*, 2002 ; Papakostas *et al.*, 2005), *B. calyciflorus* (Schro¨der and Walsh, 2007; Xiang *et al.*, 2011 ; Papakostas *et al.*, 2016), *B. quadridentatus* (Garcı´a -Morales and Domı´nguez-Domı´nguez, 2019 ), *Lecane* spp.(Garcı´a-Morales and Eli´as-Gutie´rrez, 2013), *Lecane bulla* (Garcı´a -Morales and Domı´nguez-Domı´nguez, 2020), *Polyarthra dolichoptera*, *Synchaeta* spp. (Obertegger *et al.*, 2012), *Testudinella clypeata* (Leasi *et al.*, 2013), *Keratella cochlearis* (Segers and De Smet,2008), *Euchlanis dilatata* (Kordbacheh *et al.*, 2017) and bdelloid rotifers (Fontaneto *et al.*, 2009). All these studies provide evidence of how molecular approaches are important tools for understanding the nature of genetic differentiation and are also relevant to rotifera taxonomy. In Cladocera, Brakovska *et al.*, (2013) studied the occurrence of cladocera and the genetic diversity of *Daphnia cucullata* in Latvia and showed the dominant species of cladocera are *Diaphanosoma brachyurum*, *Daphnia cucullata*, *Bosmina crassicornis*, *Bosmina longispina* and *Bosmina longirostris*. *Daphnia*

*cucullata* used as a good model for ecological genetics research; Sharma, (2014) examined the Australian *Ceriodaphnia* (Cladocera: Daphniidae) using morphological attributes and two mitochondrial DNA (COI and 16s rRNA) and one nuclear DNA (28s rRNA) gene fragments to differentiate the species. The sequence data supports the existence of three species and indicated that mitochondrial markers, COI and 16s rRNA are appropriate molecular markers for species discrimination and identification of *Ceriodaphnia*; Wang *et al.*, (2016) examined the genetic diversity of *Daphnia pulex* of the Yangtze River using 16s rRNA, cytochrome c oxidase subunit I (COI), and 18s rRNA genes as molecular marks and indicating that *D. pulex* may have evolved into different subspecies.

### **1-3-6 : Local studies**

#### **1-3-6-1 : Zooplankton:**

In Iraq, there are some studies concerned with the identification and biodiversity of zooplankton such as Rotifera and Cladocera. As for the rotifers, Ghazi and Ahmed, (2008) studied the abundance and diversity of Rotifera in ponds in Garmat Ali region and recorded 26 species and determined the diversity of rotifera during the water salinity changes in selected stations at Shatt Al-Arab River and Shatt Al-Basrah canal; Salman *et al.*, (2014) listed 105 species of rotifera from the Southern Iraqi marshes; Al –Kalidy, (2014) studied the diversity of zooplankton in Al-Shamyia river and identified 79 species of rotifera; Al–Karawi, (2014) studied the diversity of zooplankton in Al-Kufa river and identified 81 species of rotifera; Abdulwahab and Rabee, (2015) investigated the biodiversity of zooplankton in the Tigris River in Baghdad City, central of Iraq, as well as analyzed some physical and chemical parameters such as water and air temperature, pH, EC, TDS, DO, etc and identified 106

taxonomy units of zooplankton including 65 taxa belonging to rotifera, 25 taxa to copepod and 16 taxa to cladocera. Also, measured the diversity indices and the values of species richness index of rotifera varied from 1.051 to 12.98, for Cladocera from 1.285 to 3.41 and for copepod from 1.5 to 7.2. The Shannon–Weiner index of rotifera varied from 0.67-3, 0.50–1.72 for cladocera and from 0.91-2.51 for copepoda. The uniformity index of zooplankton varied from 0.41-0.93 for rotifera, 0.33–1 for cladocera and 0.36–1 for copepoda; Hammadi *et al.*, (2016) studied the diversity of rotifera in Shatt Al-Arab Region, South of Iraq and recorded 99 species of rotifera belonging to 23 genera and 15 families, *Synchaeta lakowitziana* and Bdelloidea are the most dominant groups, in addition to measured the ecological indices: richness (1.32-8.43), Shannon-Weaver diversity index (0.99-2.36), evenness (0.59-0.91) and similarity index and Majeed *et al.*, (2022) evaluated the effects of Tharthar Arm on the composition and diversity of Rotifera in Tigris River and showed that low density of Rotifera in Tharthr Arm decreased the density in Tigris as well as declined the mean values of richness, evenness and Shannon diversity indices.

In the Hilla river, studies concerned with the identification and biodiversity of rotifera are limited, of which, Akbar, (1994) studied the effect of some environmental factors on the diversity of zooplankton in Hilla river and recorded seven species of cladocera, three species of copepoda, two species of ostracoda and rotifer without addressing the density or species; Abd Al-Rezzaq *et al.*, (2014) studied the rotifera in Hilla river which is passing through the city of Babylon – Iraq and was the first study in this river and identified 128 rotifera taxa (108 species +19 genera + one class), 40 taxa are recorded for first time in Iraq (39

species + one genus). *K. cochlearis* showed the highest abundant (39%) of the rotifera, *Lecane* showed the highest species number (14 species).

As for the Cladocera, the first research is published by Gurney, (1921) in which about 40 species of aquatic invertebrates are collected by Dr. Bu Buxton and reported during 1917 and 1918 in Amara Province and its environs. They recorded 18 species of cladocera. Then Mohammad, (1965) study in the center and south of Iraq, recorded 23 species of the cladocera including 15 species as a first record in Iraq; Khalaf and Smirnov, (1976) recorded some crustaceans including cladocera from the littoral zone of the central and southern Iraq, they reported 23 species of cladocera; Salman *et al.*, (1986) studied the seasonal variations of zooplankton at Shatt Al-Arab, they found that cladocera is dominating the zooplankton followed by copepoda; AL-Saboonchi *et al.*, (1986) studied the zooplankton of the Al-Hammar marshes and recorded seven species of cladocera; Mohammad, (1986) made report on 13 species of cladocera in the Tigris River in Baghdad and recorded eighteen species in the Euphrates River in Al-Falluja; Lazim and Zeki, (1987) recorded *Scapholeberis kingi* in Al-Zoafaranyh and reported it also in Arbil; Mangalo and Akbar, (1988) found that the population density of cladocera in Diyala River is higher than that in the Tigris River; Sabri *et al.*, (1989) studied the vertical distribution of zooplankton in Samarra dam and found that most species of zooplankton avoid the surface layer, while the water fleas are distributed mainly in the upper water layers and recorded four species of cladocera; Salman *et al.*, (1990) encountered very few specimens of cladocera on a few occasions only in Khour Abdullah, north west Arabian Gulf; Al-Zubaidi, (1998) showed that the maximum abundance of cladocera in Al-Faw and Al-Sebah stations are 1802 and 1057 ind/m<sup>3</sup> respectively; Poltorak *et al.*, (2001) found 99

species of zooplankton in Al-Therthar, Al-Razzazah and Al-Habbaniya Lakes including 20 species of cladocera; AL-Zubaidi and Salman, (2001) studied the zooplankton in Shatt Al-Arab estuary, they referred to cladocera as comprising 58% of the zooplankton in Al-Sebah region; Ajeel, (2004) noted that cladocera formed 5.4-35 % of the total zooplankton in Shatt Al-Arab region; Al-Jizany, (2005) studied the impact of organic pollution on the diversity and abundance of the plankton in Shatt Al-Arab River, Al-Ashar and Al-Rabat channels, she found that the cladocera constituted the third important group; Al-Qarooni, (2005) studied the abundance and the presence of some zooplankton in the marshlands of southern Iraq and recorded 14 species of cladocera; Al-Nimrawi, (2006) recorded 15 species of cladocera in the Tigris and Euphrates Rivers; Ajeel *et al.*, (2006) found that the cladocera comprised 10.3% in the South of Hour Al-Hammar; Ajeel *et al.*, (2008) indicated that the cladocera comprised 8.8 % of the total zooplankton in Basrah district and Abbas, (2010) recorded 23 species of cladocera in the North of Shatt Al-Arab River.

In Hilla river, there are no previous detailed studies concerned with cladocera, So, in the present study we focusing on the identification and diversity of cladocera.

### **1-3-6-2 : Genetic diversity :**

In Iraq, the studies of genetic variation between and within species at molecular level are very few including Albhadly and Jawad, (2019) who studied the genetic diversity in three species belonging to the family of Daphniidae, one belongs to the genus *Daphnia* and two species belonging to the genus *Simocephalus* collected from different places in Baghdad-Iraq and showed that there are two species: *D. magna* and *S. vetulus* are

registered in Iraq, while the species *S.congener* recorded in Iraq for the first time. The genes COI, 16s rRNA, that of mtDNA, and 18s rRNA of the nucleus studied for the purpose of obtaining genetic diversity among species and for detecting the differences in the studied genes ; Hassan *et al.*, (2021) studied the molecular identification of *Brachionus calyciflorus* named HH1.

# *Chapter Two*

## *Materials and Methods*

## **2 : Material and Methods**

### **2 – 1 :Description of the study area**

Al-Hilla river is one of the main rivers branching of the Euphrates River. Where the Euphrates River reach Hindiya Dam after 602 km in Iraq it is branches off from the left side with a current length of 101km (according to the administrative borders of the Babylon province), Al-Hilla river feeds large areas of agricultural land in Babylon province reaches about 1,022,489 acres. In addition, water needs for civil and industrial consumption. Thirty six streams branch off from Al-Hilla river, reaching a total of its length is 511.6 km, in addition to 562 small outlets branching irregularly. The drainage of Al-Hilla river reaches 110 m<sup>3</sup>/sec in winter season and 150 m<sup>3</sup>/sec and in summer (Water Resources Department/Babylon).

Five stations are selected for collecting samples including three stations from Al-Hilla river and two stations branching from Al-Hilla river (Figure1). The first station (St1) is situated at the river's entrance to the village of Anana (32°33'6.61"N, 44°25'14.22"E) and classified as an agricultural area as well as having an animal barn close by. The second station (St2) represents Al-Hilla river before entering the city center (Betta Bridge) (32°31'1.35"N, 44°25'37.40"E) and categorized as an agricultural area. The third station (St3) depicts Al-Hilla river immediately following its departure from Hilla center in Al-Farsi region (32°28'7.31"N , 44°26'23.79"E). In addition to waste from Hilla market, sources of domestic waste from residential areas in Al-Farsi neighborhood were also noted. Additionally, it is observed that sewage water is discharged straight through water tanks or sewer outlets. The fourth station (St4), a branch that arises from Al-Hilla river and flows

through the village of Al-Ghalis ( $32^{\circ}26'15.59''\text{N}$ ,  $44^{\circ}26'53.56''\text{E}$ ) and distinguished by being an agricultural region where the region's agricultural fields are irrigated using drainage water. The fifth station (St5), branch that splits off from Al-Hilla river near Nazim Dora and travels through the region between Al- Dolab and Al-Dabla ( $32^{\circ}25'4.08''\text{N}$ ,  $44^{\circ}29'14.67''\text{E}$ ) and unique in that it is an agricultural area with drainage water used to irrigate the agricultural crops there.



**Figure (1): Map of Al-Hilla river show the study stations (Google Maps)**

## 2 – 2 :Collection of Samples

Water and zooplankton are collected seasonally during the period from spring 2021 to winter 2022. Three replicates for each sample are taken. Water samples collected for physical and chemical analysis using 5 liters polyethylene containers. Zooplankton samples are collected by zooplankton net with 30 cm diameter, 50 cm length and 50  $\mu\text{m}$  mesh size. The net rope is marked every meter and after the net plunges to the

bottom, pulled to the water surface then the net is washed from the outside using a sprayer. The samples concentrated in the receiver net then kept in plastic bottles then a little quantity of 4 % formalin is added immediately after sampling. Date, sample name and the depth are recorded on bottle (Harris *et al.*, 2000).

## **2 – 3 :Physical and Chemical Parameters**

### **2 –3- 1 :Air and Water Temperature**

The temperature of air and water are measured directly in the shade of the station using thermometer grading from 0–100 °C (Kamp England).

### **2 –3- 2 :Electrical Conductivity (EC), potential of Hydrogen (pH) , Total Dissolved Solid (TDS) and Salinity**

Electrical Conductivity ( $\mu\text{s}/\text{cm}$ ), pH, total dissolved solid (mg/l) and salinity (mg/l) are measured using multi-parameters type HANNA, model Romania, after the calibration each parameter.

### **2 –3- 3 : Dissolved Oxygen ( DO)**

Dissolved Oxygen is measured using Dissolved Oxygen meter type Oakton ,American made and the results are expressed in (mg/l) after the calibration.

## **2 – 4 : Enumeration of the Zooplankton**

In order to count the zooplankton per one liter, the amount of water filtered through the net is calculated according to the cylinder volume formula (Lind,1979) :

$$V = (\pi r^2 h) / 1000 , \text{ Whereas :}$$

V : volume of filtered water ( in L)

r : radius of the net ( in cm )

h : height of water column ( in cm )

A Sedgewick-Rafter Chamber, its capacity 1 ml characterized 1000 squares (50\*20) and the count is done using an inverted microscope. To calculate the number of individuals per liter used the method described by APHA (2005), all the results are defined by the number of individuals/liter

$$\text{No./L} = (C * V1) / (V2 * V3)$$

No./L = The number of individuals per liter .

C = The calculated number of individuals in the slide .

V1 = Intensive volume sample (ml)

V2 = The volume of sample in which the counting of zooplankton (1 ml )

V3 = Volume of filtered water in liter

## 2 – 5: Biodiversity Indices

### 2 – 5- 1: Shannon - Wiener diversity index

Biodiversity of zooplankton is studied using the formula of Shannon-Wiener diversity index (Shannon and Wiener, 1949) which denoted by

$$H' = - \sum_{i=1}^s P_i \ln P_i$$

Where,

H = the Shannon diversity index

P<sub>i</sub> = fraction of the entire population made up of species i

S = numbers of species encountered

Σ = sum from species 1 to species S

The value of this index can theoretically range from zero to infinity. However, values normally range from 0 to 4.

**2 – 5- 2: Margalef's richness index**

The richness is calculated according to (Margalef, 1956). This index is used to clarify the relationship between the number of species and the number of individuals and their distribution depending on their abundance.

$$R = (S-1) / \ln (N)$$

Where,

R = richness index

S = the total number of species

N = the total number of individuals observed in the sample.

This index commonly varies between 1 and 5, and larger the index indicates a more healthy body of water. When it tends towards 1 pollution is thought to increase and damage should be suspected.

**2 – 5- 3: Index of Dominance (Simpson Index)**

Within a major community there are species or groups which largely control the energy flow and strongly affect the environment of all other species. They are known as ecological dominance (Odum, 1971). The degree to which dominance is concentrated in one or many species can be expressed by an appropriate index of dominance that sums each species importance in relation to community as a whole (Simpson, 1949)

$$C = \sum (n_i / N)^2, \text{ where}$$

C = Index of dominance

$n_i$  = Number of individuals for each species

N = Total number of individuals in the sample

The value of 'C' varies between 0 and 1. Higher diversity values reflect diversified resources in the habitat available for components of the community. Decreased values indicate increase by an average species resulting in the lowering of the number of coexisting species in the community.

### 2 – 5- 4: Evenness index (Equitability index)

Another major component of diversity is "evenness" or "equitability" in the apportionment of individuals among the species. It is expressed according to (Pielou, 1966) as the following

$$E = \frac{-\sum_i \left( \frac{n_i}{N} - \ln \frac{n_i}{N} \right)}{\ln N}$$

### 2 – 5- 5: Menhinick Index

Another diversity index (Menhinick, 1964), which considers only the number of species (S) and the square root of the total number of individuals and is calculated by the equation:

$$R_2 = S / \sqrt{\sum_i n_i}$$

Where ,

$S$  = the total number of species in a sample/community

$n_i$  = the number of individuals of the  $i$ - species, where  $i = 1, 2, \dots, S$

### 2 – 5- 6: Berger-Parker Index

Dominance refers to the abundance of the most abundant species, the simplest measure of which is the abundance of the most abundant species (absolute dominance;  $n_{max}$ ) (Magurran and McGill, 2011). Relative

dominance is also a common measure, and it is known as the Berger–Parker index.

$$DBP = n_{max} / N$$

Where,

DBP = Berger–Parker index of dominance

$n_{max}$  = abundance of the most abundant species

N = Total abundance

### **2 – 5- 7: Jaccard's Index (%)**

It represents the degree of similarity in the number of species between two stations and calculated according (Southwood, 1978) as the following:

$$J = C / (A+B-C) \times 100 \quad \text{Where,}$$

A= Number of species at station A

B= Number of species at station B

C= Number of species present in both stations

### **2 – 5- 8: Alpha, Beta and Gamma Diversity**

The three terms for measuring biodiversity over spatial scales are alpha, beta, and gamma diversity (Whittaker, 1972). Alpha diversity refers to the diversity within a particular area or ecosystem, and is usually expressed by the number of species (species richness) in that ecosystem. Beta diversity is a comparison of diversity between ecosystems usually measured as the amount of species change between the ecosystems. Gamma diversity is a measure of the overall diversity for different ecosystems within a region or as "geographic-scale species diversity" (Hunter, 2002).

## **2 – 6 : Morphological Identification of the Zooplankton**

### **2 – 6-1 : Identification of Rotifera**

Numerous classification keys and studies are utilized to identify of rotifers including: (Koste, 1978; Koste and Shiel, 1980; Shiel, 1995; De Smet, 1997; Segers, 1995a,b; Segers, 2002a,b; Segers, 2003; Segers, 2007; Nogrady and Segers, 2002; Radwan *et al.*, 2004; Wallace *et al.*, 2006; Jersabek and Leitner, 2013 ; Abd Al- Razzaq, 2014).

### **2 – 6-2 : Identification of Cladocera**

Various classification keys and investigations are used to identify of cladocera including (Sweetman and Smol, 2006; Korosi and Smol, 2012a,b ; Błędzki and Rybak, 2016 ; Wojewodka *et al.*, 2020).

## **2 – 7 : Molecular Identification of the Zooplankton**

### **2-7-1: Modified method for Genomic DNA extraction**

Cetyl trimethyl ammonium bromide (CTAB) DNA Extraction Kit is used to extract genomic DNA from samples following the manufacturer's protocol:

#### **Step 1 : Preparation of sample and Lysis**

1- Transfer 20-25 mg of the sample or (100-200 uL for the liquid sample) to Eppendorf tube.

2- Immediately, add 800 µL of Pre-heated CTAB buffer, vortex to re-suspend the sample, then incubate in water bath for 30-60 min at 65°C. Mix by inverting tube every 5 min.

3- After incubation, Briefly spin down to get the drops clinging under the Lid. Allow the tubes for 3 min to cool to room temperature..

4- To each tube add 500  $\mu$ l of Chloroform : Iso Amyl Alcohol and mix the solution by inverting or on a rotating mixer for 5 min.

5- After mixing, spin the tubes at 14000 rpm for 5 min.

6- Transferring ALL the supernatant to new eppendorf tube.

### **Step 2 : DNA Binding**

1- Add Equal volume of binding buffer, mix well by inverting and vortex briefly for 10 sec.

2- Place a spin column in the collection tube. Transfer 700 uL of the lysate to the column.

3- Centrifuge for 1 min at 10.000 rpm at R.T. Save the column and collection tube. Discard the elute only.

Note : Ensure the entire lysate volume has passed through into the collection tube by inspecting the column. If the entire lysate volume has not passed, spin at 14,000 rpm for 1 minutes.

4- Add the remaining mixture (700 uL) to the column, then, repeat centrifuge in step 3.

### **Step 3 and 4 : DNA Washing 1 and DNA Washing 2**

1- Add 500  $\mu$ L of wash buffer-1, centrifuge 1 min at 10.000 rpm. Save the column and discard the elute only.

2- Add 500  $\mu$ L of wash buffer-2, centrifuge for 1 min at 13,000 rpm. Save the column and collection tube. Discard the eluate only.

3- Perform an additional centrifuge for 3 min at 13,000 rpm to remove ethanol residual.

### **Step 5 :DNA Elution**

1- Spin columns are inserted into new 1.5 ml tubes.

2- Add 100 - 120  $\mu$ L of pre-warmed (65 °C) Elution buffer to the column and incubate at R.T for 5 min.

- 3- Centrifuge for 2 min at 10.000 rpm at room temperature.
- 4- Discard the column and store extracted DNA at 2 to 8 °C.

### 2-7-2: Estimation of DNA concentration and purity

The purity and concentration of the DNA obtained are determined through 260/280 nm absorbance measures (Desjardins and Conklin,2011) using the NanoDrop spectrophotometer 2000 (Thermo Scientific) (Wilmington DE, USA (2009)).

### 2-7-3: Primer preparation

Primers are obtained as lyophilized form from macrogen company (South Korea) and design especially for this study (table 1) depending on sequence available at <https://www.ncbi.nlm.nih.gov/> using free online tool <https://www.bioinformatics.nl/cgi-bin/primer3plus/primer3plus.cgi/>. Each primer are dissolved in molecular grade distill water to reach 100 pmol as stock and then the work solution of primer are prepared by adding 10 ul from stock to 90 ul of D.W.

**Table (1): Primers designed for the current study**

Primer for characterization			
Primer	Sequence 5 to 3	Product	Annealing temperature
Rotifera,F	GACTCAACACGGGAAATCTC ACC	255	58 C°
Rotifera,R	CTAAGGGGCATCACAGACC		
Cladocera,F	GACCCGTCTTGAAACACGGA	393	60 C°
Cladocera,R	TCGGGGGGAACCAGCTACTA		

### 2-7-4: Polymerase chain reaction (PCR)

Conventional PCR are used to amplify the target DNA using specific primer pairs. It include three consecutive steps that repeated for specific

number of cycles to get PCR product (amplicon) which could be finally visualized after agarose gel electrophoresis. The thermal cycling conditions mentioned in the (table 2).

**Table (2): PCR Conditions of the current study**

Primer	Product	Conditions
Rotifera,F	255	Step 1: 95 C <sup>o</sup> , 2 min. Step 2: 95 C <sup>o</sup> , 30 sec. Step 3: 58 C <sup>o</sup> , 30 sec. Step 4: 72 C <sup>o</sup> , 20.0 sec. Step 6: 72 C <sup>o</sup> , 5 min. Step 7: 4 C <sup>o</sup> , forever
Rotifera,R		
Cladocera,F	393	Step 1: 95 C <sup>o</sup> , 2 min. Step 2: 95 C <sup>o</sup> , 30 sec. Step 3: 60 C <sup>o</sup> , 30 sec. Step 4: 72 C <sup>o</sup> , 20.0 sec. Step 6: 72 C <sup>o</sup> , 5 min. Step 7: 4 C <sup>o</sup> , forever
Cladocera,R		

### 2-7-5: Agarose Gel Electrophoresis

Agarose gel electrophoresis is the most effective way of separating DNA fragments. The concentration of agarose in a gel depends on the sizes of the DNA fragments need to be separated, ranging between 0.5%-2% (Lee *et al.*, 2012). A 0.7% gel was used to obtain good separation of genomic DNA (5-10 kb) after extraction while 1.5%-2% is used to gain good resolution for small fragments of PCR product (0.2-1 kb). However, the specific weight of agarose is added to 100 ml of 1×TBE buffer and then melted in microwave until the solution becomes clear. Once the agarose is cooled to 50-55°C, 5µl of simply safe dye (10 mg/ml) is added to 100 ml of melting agarose gel to get final concentration 0.5µg/ml (Sambrook and Russel, 2006). The agarose is poured in the gel tray with sealed ends, comb placed properly, and then left to dry. The samples are loaded in a separate well of the gel, with marker in one well. Electrodes are connected correctly and the run is applied according to the gel

percentage and size of gel, (The time of agarose gel electrophoresis is 45 minute for genomic DNA and 1 hour and 30 minute for PCR product).

- 1- Place the gel-casting tray in plastic tray, check that the teeth of the comb are approximately 0.5 mm above the gel bottom.
- 2- Prepare 500 ml of TBE (1X) by adding 50ml of TBE (10X) stock solution to a final volume of 450 ml of Deionized water.
- 3- Place 100 ml of the buffer into a 500 ml flask and add 1.5 g of agarose. Melt the agarose by heating (microwave) the solution on hot plate for approximately 10 min. Carefully swirl the agarose solution to ensure that the agarose is dissolved, that is no agarose particles are visible.
- 4- Cool the agarose solution to approximately 50 °C and added 2-3  $\mu$ l of ethidium bromide 10 mg/ml stock solution. Slowly pour the agarose into the gel-casting tray. Remove any air bubbles by yellow tip.
- 5- Position the comb approximately 1.5 cm from the edge of the gel. Let the agarose solidify for approximately 20–30 minutes. After the agarose has solidified remove the comb with a gentle back and forth motion, taking care not to tear the gel.
- 6- Remove the gel-casting tray and place the tray on the central supporting platform of the gel box.
- 7- Add electrophoresis buffer to the buffer chamber until it reaches a level of 0.5–1cm above the surface of the gel.
- 8- Load the samples into the wells using a yellow tip. Place the tip under the surface of the electrophoresis buffer just above the well. Expel the sample slowly, allowing it to sink to the bottom of the well. Take care not to spill the sample into a neighboring well. Note: samples must be loaded in sequential sample wells. When loading fewer samples than the number of wells it is preferable to leave the wells nearest the edge of the gel empty.

9- First load 5µl of ladder molecular weight marker to each side of the gel (flanking the sample line) and 20 µl of DNA specimen in the other well.

10- Place the lid on the gel box and connect the electrodes. DNA will travel towards the positive (red) electrode positioned away from the well. Turn on the power supply.

11- Continue electrophoresis until the tracking dye moves at least 10 cm of the gel length.

### **2-7-6: Sequencing of PCR product**

Fifty micro liters of PCR product for each sample are send to Macrogen/Korea for Sanger sequencing. After trimming of each sequence, the result of the trimmed sequence are blasted in NCBI to check the similarities and differences with data base. ([https://blast.ncbi.nlm.nih.gov/Blast.cgi?PROGRAM=blastn&PAGE\\_TY PE=BlastSearch&LINK\\_LOC=blasthome](https://blast.ncbi.nlm.nih.gov/Blast.cgi?PROGRAM=blastn&PAGE_TY PE=BlastSearch&LINK_LOC=blasthome)) software are used to check the similarities and differences.

### **2-7-7: Phylogenetic Tree**

A phylogenetic tree is an estimate of the relationships among taxa or sequences and their hypothetical common ancestors. Today, most phylogenetic trees are built from molecular data: DNA or protein sequences. Originally, the purpose of most molecular phylogenetic trees are estimating the relationships among the species represented by those sequences (Hall, 2013). The phylogenetic tree is designed by NCBI blast and MEGA X software.

### **2-8: Statistical Analysis**

SPSS V.23 program used for calculated the significant different by using Duncan test , also this program used for correlation coefficient at

level of significance ( $p < 0.05$ ). The CANOCO program (version 4.5) of multivariate analysis of ecological data used for calculates the Canonical correspondence analysis (CCA).

# *Chapter Three*

## *The Results*

### 3 – 1 :Physical and Chemical Parameters

#### 3 – 1-1 :Air and Water Temperature

Air temperature varied throughout the study period from 11.73 °C in station one in the winter to 46.01 °C in station five in the summer (Figure 2) , while water temperatures varied from 10.51 °C in station two in the winter to 35.21 °C in station five in the summer (Figure 3). The statistical analysis revealed significant differences across all seasons for both air and water temperature rates when comparing it during the study period and between the stations (tables 3-4).

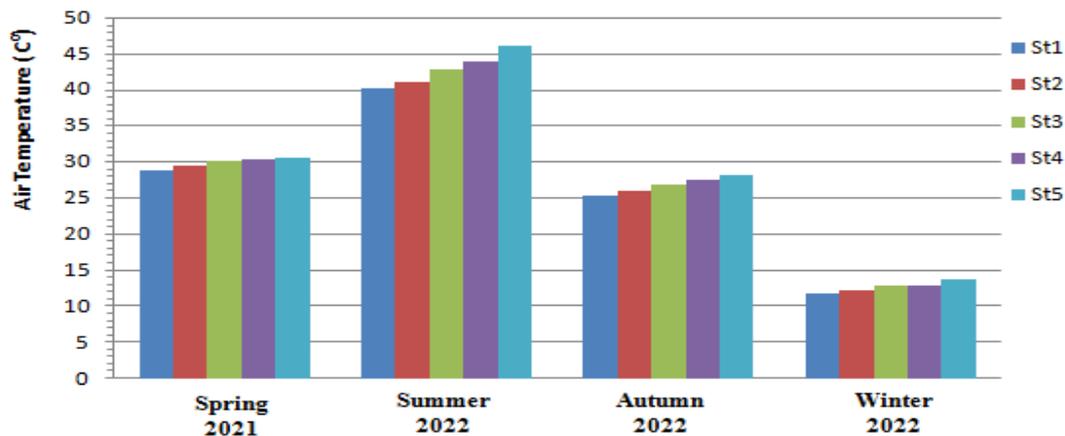


Figure (2): Seasonal changes of air temperature (°C) for the five stations during the study period from Spring 2021 to Winter 2022

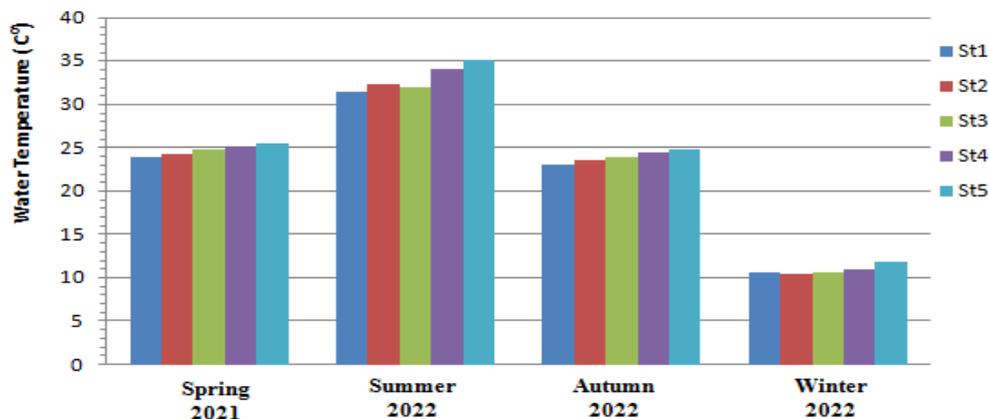


Figure (3) : Seasonal changes of water temperature (°C) for the five stations during the study period from spring 2021 to winter 2022

Table (3): The differences of physicochemical characters, total density and number of taxa among stations from Spring 2021 to Winter 2022

Seasons Parameters	Spring					Summer					Autumn					Winter				
	St1	St2	St3	St4	St5	St1	St2	St3	St4	St5	St1	St2	St3	St4	St5	St1	St2	St3	St4	St5
<b>Air Temperature (C°)</b>	28.86 ± 0.202 a	29.50 ± 0.0 b	30.13 ± 0.12 c	30.38 ± 0.12 d	30.61 ± 0.07 e	40.15 ± 0.21 a	41.18 ± 0.11 b	42.93 ± 0.16 c	43.91 ± 0.14 d	46.01 ± 0.175 e	25.33 ± 0.288 a	26.06 ± 0.11 b	26.83 ± 0.28 c	27.56 ± 0.208 d	28.16 ± 0.15 e	11.73 ± 0.02 a	12.25 ± 0.05 b	12.80 ± 0.08 c	12.96 ± 0.12 c	13.66 ± 0.25 d
<b>Water Temperature (C°)</b>	24.03 ± 0.17 a	24.36 ± 0.15 b	24.83 ± 0.15 c	25.18 ± 0.07 d	25.58 ± 0.208 e	31.41 ± 0.28 a	32.28 ± 0.37 b	31.96 ± 0.40 ab	34.05 ± 0.26 c	35.21 ± 0.202 d	23.00 ± 0.0 a	23.53 ± 0.25 b	24.00 ± 0.0 c	24.46 ± 0.05 d	24.76 ± 0.35 d	10.58 ± 0.20 a	10.51 ± 0.14 a	10.66 ± 0.24 a	11.03 ± 0.17 b	11.83 ± 0.12 c
<b>pH</b>	8.13 ± 0.07 a	8.01 ± 0.07 a	8.35 ± 0.17 b	8.38 ± 0.15 b	8.06 ± 0.05 a	7.85 ± 0.08 ab	7.73 ± 0.05 a	8.35 ± 0.13 c	7.73 ± 0.07 a	8.00 ± 0.18 b	7.40 ± 0.30 a	7.63 ± 0.28 abc	7.93 ± 0.15 bc	7.56 ± 0.05 ab	8.06 ± 0.251 c	8.01 ± 0.05 b	7.70 ± 0.13 a	7.50 ± 0.13 a	7.56 ± 0.20 a	7.55 ± 0.18 a
<b>Electrical Conductivity (µs/cm)</b>	582.66 ± 6.78 b	520.66 ± 13.53 a	640.83 ± 1.75 c	530.83 ± 7.57 a	540.5 ± 16.22 a	911.0 ± 15.4 b	897.5 ± 33.77 b	1055.0 ± 6.14 d	859.66 ± 15.06 a	999.0 ± 14.23 c	1064.0 ± 40.44 a	1063.66 ± 23.02 a	1140.0 ± 41.72 b	1041.33 ± 18.55 a	1183.66 ± 27.93 b	1159.5 ± 39.86 b	1093.66 ± 27.59 a	1375.66 ± 34.01 d	1075.16 ± 39.28 a	1291.5 ± 18.66 b
<b>TDS (mg/l)</b>	395.66 ± 13.05 b	396.33 ± 11.09 b	435.33 ± 4.93 c	362.66 ± 5.79 a	373.16 ± 16.91 a	662.5 ± 2.50 ab	650.16 ± 40.65 a	742.66 ± 5.79 c	676.5 ± 68.54 abc	721.16 ± 14.49 bc	814.0 ± 32.07 bc	792.0 ± 23.64 ab	859.66 ± 20.74 cd	761.33 ± 22.94 a	867.66 ± 32.53 d	853.33 ± 23.93 b	804.0 ± 20.42 a	964.0 ± 37.40 c	776.33 ± 13.65 a	880.83 ± 15.73 b
<b>Salinity (mg/l)</b>	347.5 ± 12.75 a	350.0 ± 6.87 a	433.66 ± 6.17 c	372.83 ± 2.84 b	377.0 ± 7.50 b	492.5 ± 8.35 b	491.33 ± 13.51 b	526.0 ± 4.35 c	466.66 ± 5.53 a	499.83 ± 10.56 b	566.0 ± 30.64 a	544.66 ± 14.50 a	624.33 ± 11.23 b	539.66 ± 11.50 a	638.33 ± 14.18 b	529.33 ± 10.77 a	525.16 ± 5.34 a	691.0 ± 28.73 c	516.66 ± 12.75 a	650.0 ± 14.08 b
<b>Dissolved Oxygen (mg/l)</b>	5.90 ± 0.21 bc	5.53 ± 0.03 a	5.80 ± 0.15 ab	5.90 ± 0.17 bc	6.15 ± 0.15 c	5.02 ± 0.03 a	5.10 ± 0.03 a	4.94 ± 0.08 a	5.08 ± 0.19 a	4.96 ± 0.09 a	6.10 ± 0.16 a	6.09 ± 0.10 a	5.94 ± 0.07 a	6.06 ± 0.11 a	5.79 ± 0.28 a	10.04 ± 0.02 b	10.25 ± 0.22 b	9.68 ± 0.06 a	10.04 ± 0.11 b	10.17 ± 0.22 b
<b>Density of Zooplankton (Ind/l)</b>	0.916 ± 0.11 a	1.026 ± 0.07 a	1.254 ± 0.33 a	1.223 ± 0.21 a	1.115 ± 0.12 a	0.912 ± 0.07 a	0.956 ± 0.11 a	0.924 ± 0.03 a	1.386 ± 0.15 b	1.014 ± 0.12 a	0.597 ± 0.1 b	0.522 ± 0.05 b	0.524 ± 0.06 b	0.917 ± 0.05 c	0.32 ± 0.05 a	0.447 ± 0.04 a	0.472 ± 0.08 a	0.429 ± 0.02 a	1.264 ± 0.28 b	0.637 ± 0.08 a
<b>No of Taxa</b>	13.33 ± 9.01 a	11.00 ± 5.29 a	9.66 ± 3.78 a	8.66 ± 2.88 a	12.66 ± 7.23 a	14.00 ± 6.92 a	14.00 ± 9.16 a	12.33 ± 9.23 a	12.00 ± 5.56 a	14.00 ± 8.88 a	10.00 ± 6.08 a	7.66 ± 4.72 a	6.00 ± 2.64 a	9.00 ± 5.56 a	5.33 ± 4.93 a	7.00 ± 4.35 a	7.33 ± 4.16 a	6.00 ± 5.29 a	12.66 ± 6.65 a	7.00 ± 3.60 a

- The values represents ( mean ± SD)

- Similar adjacent letters indicate a non significant differences  $P \leq 0.05$  for each season

Table (4): The differences of physicochemical characters, total density and number of taxa among seasons within station from Spring 2021 to Winter 2022

Stations	St1				St2				St3				St4				St5			
	Spring	Summer	Autumn	Winter	Spring	Summer	Autumn	Winter	Spring	Summer	Autumn	Winter	Spring	Summer	Autumn	Winter	Spring	Summer	Autumn	Winter
Air Temperature (°C)	28.68 ± 0.202 c	40.15 ± 0.217 d	25.33 ± 0.288 b	11.73 ± 0.02 a	29.50 ± 0.0 c	41.18 ± 0.11 d	26.06 ± 0.11 b	12.25 ± 0.05 a	30.13 ± 0.12 c	42.93 ± 0.16 d	26.83 ± 0.28 b	12.80 ± 0.08 a	30.38 ± 0.12 c	43.91 ± 0.14 d	27.56 ± 0.208 b	12.96 ± 0.12 a	30.61 ± 0.07 c	46.01 ± 0.175 d	28.16 ± 0.15 b	13.66 ± 0.25 a
Water Temperature (°C)	24.03 ± 0.17 c	31.41 ± 0.28 d	23.00 ± 0.0 b	10.58 ± 0.20 a	24.36 ± 0.15 c	32.28 ± 0.37 d	23.53 ± 0.25 b	10.51 ± 0.14 a	24.83 ± 0.15 c	31.96 ± 0.40 d	24.00 ± 0.0 b	10.66 ± 0.24 a	25.18 ± 0.07 c	34.05 ± 0.26 d	24.46 ± 0.05 b	11.03 ± 0.17 a	25.58 ± 0.208 c	35.21 ± 0.202 d	24.76 ± 0.35 b	11.83 ± 0.12 a
pH	8.13 ± 0.07 b	7.85 ± 0.08 b	7.40 ± 0.30 a	8.01 ± 0.05 b	8.01 ± 0.07 a	7.73 ± 0.05 ab	7.63 ± 0.28 a	7.70 ± 0.13 ab	8.35 ± 0.17 c	8.35 ± 0.13 c	7.93 ± 0.15 b	7.50 ± 0.13 a	8.38 ± 0.15 a	7.73 ± 0.07 b	7.56 ± 0.05 b	7.56 ± 0.20 b	8.06 ± 0.05 b	8.00 ± 0.18 b	8.06 ± 0.251 b	7.55 ± 0.18 a
Electrical Conductivity (µs/cm)	582.66 ± 6.78 a	911.0 ± 15.40 b	1064.0 ± 40.44 c	1159.5 ± 39.86 d	520.66 ± 13.53 a	897.5 ± 33.77 b	1063.6 ± 23.02 c	1093.6 ± 27.59 c	640.83 ± 1.75 a	1055.0 ± 6.14 b	1140.0 ± 41.72 c	1375.66 ± 34.01 d	530.83 ± 7.57 a	859.66 ± 15.06 b	1041.3 ± 18.55 c	1075.1 ± 39.28 c	540.5 ± 16.22 a	999.0 ± 14.23 b	1183.6 ± 27.93 c	1291.5 ± 18.66 d
TDS (mg/l)	395.66 ± 13.05 a	662.5 ± 2.50 b	814.0 ± 32.07 c	853.33 ± 23.93 c	396.33 ± 11.09 a	650.16 ± 40.65 b	792.0 ± 23.64 c	804.0 ± 20.42 c	435.33 ± 4.93 a	742.66 ± 5.79 b	859.66 ± 20.74 c	964.0 ± 37.40 d	362.66 ± 5.79 a	676.5 ± 68.54 b	761.33 ± 22.94 c	776.33 ± 13.65 c	373.16 ± 16.91 a	721.16 ± 14.49 b	867.66 ± 32.53 c	880.83 ± 15.73 c
Salinity (mg/l)	347.5 ± 12.75 a	492.5 ± 8.35 b	566.0 ± 30.64 d	529.33 ± 10.77 c	350.0 ± 6.87 a	491.33 ± 13.51 b	544.66 ± 14.50 c	525.16 ± 5.34 c	433.66 ± 6.17 a	526.0 ± 4.35 b	624.33 ± 11.23 c	691.0 ± 28.73 d	372.83 ± 2.84 a	466.66 ± 5.53 b	539.66 ± 11.50 d	516.66 ± 12.75 c	377.0 ± 7.50 a	499.83 ± 10.56 b	638.33 ± 14.18 c	650.0 ± 14.08 c
Dissolved Oxygen (mg/l)	5.90 ± 0.21 b	5.02 ± 0.03 a	6.10 ± 0.16 b	10.04 ± 0.02 c	5.53 ± 0.03 b	5.10 ± 0.03 a	6.09 ± 0.10 c	10.25 ± 0.22 d	5.80 ± 0.15 b	4.94 ± 0.08 a	5.94 ± 0.07 b	9.68 ± 0.06 c	5.90 ± 0.17 b	5.08 ± 0.19 a	6.06 ± 0.11 b	10.04 ± 0.11 c	6.15 ± 0.15 b	4.96 ± 0.09 a	5.79 ± 0.28 b	10.17 ± 0.22 c
Density of zooplankton (Ind/l)	0.916 ± 0.11 b	0.912 ± 0.07 b	0.597 ± 0.107 a	0.447 ± 0.04 a	1.026 ± 0.07 b	0.956 ± 0.11 b	0.522 ± 0.05 a	0.472 ± 0.08 a	1.254 ± 0.33 c	0.924± 0.03 b	0.524± 0.06 a	0.429 ± 0.02 a	1.223 ± 0.21 ab	1.386 ± 0.15 b	0.917 ± 0.05 a	1.264 ± 0.28 ab	1.115 ± 0.12 c	1.014 ± 0.12 c	0.32 ± 0.05 a	0.637 ± 0.08 b
No of Taxa	13.33 ± 9.01 a	14.00 ± 6.92 a	10.00 ± 6.08 a	7.00 ± 4.35 a	11.00 ± 5.29 a	14.0 ± 9.16 a	7.66 ± 4.72 a	7.33 ± 4.16 a	9.66 ± 3.78 a	12.33 ± 9.23 a	6.00 ± 2.64 a	6.00 ± 5.29 a	8.66 ± 2.88 a	12.00 ± 5.56 a	9.00 ± 5.56 a	12.66 ± 6.65 a	12.66 ± 7.23 a	14.00 ± 8.88 a	5.33 ± 4.93 a	7.00 ± 3.60 a

- The values represents ( mean ± SD)

- Similar adjacent letters indicate a non significant differences  $P \leq 0.05$  for each station

### 3 – 1-2 : The Potential of Hydrogen (pH)

According to the current study, station one's pH is lowest in the autumn (7.4) while station four's pH was highest in the spring (8.38) (Figure 4). When comparing pH rates during the study period and between the stations, statistical analysis showed that there are significant seasonal changes for pH rates (tables 3-4).

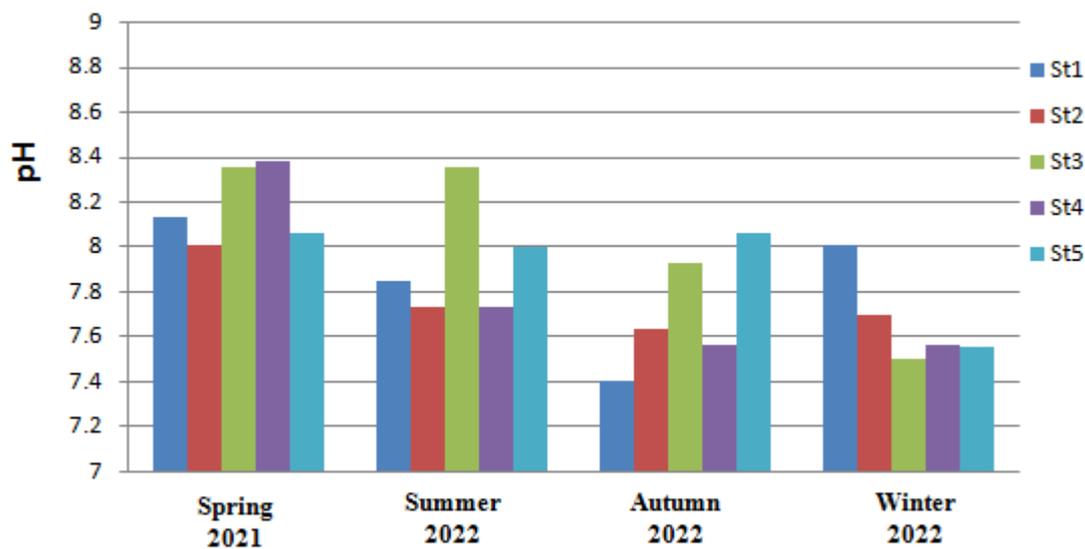


Figure (4): Seasonal changes of pH for the five stations during the study period from Spring 2021 to Winter 2022

### 3–1-3 : Electrical conductivity, Total dissolved solids and Salinity

In the present study, electrical connectivity, total dissolved solids and salinity values varied from the lowest values (520.66)  $\mu\text{s}/\text{cm}$  in station two, (362.66)  $\text{mg}/\text{l}$  in station four and (347.5)  $\text{mg}/\text{l}$  in station one during the spring to the highest values (1375.6)  $\mu\text{s}/\text{cm}$ , (964)  $\text{mg}/\text{l}$  and (691)  $\text{mg}/\text{l}$  respectively in station three during the winter (Figures 5-7). Significant fluctuations are discovered by statistical examination of EC, TDS and salinity during the study period and between the sites revealed that there are significant variations (tables 3-4).

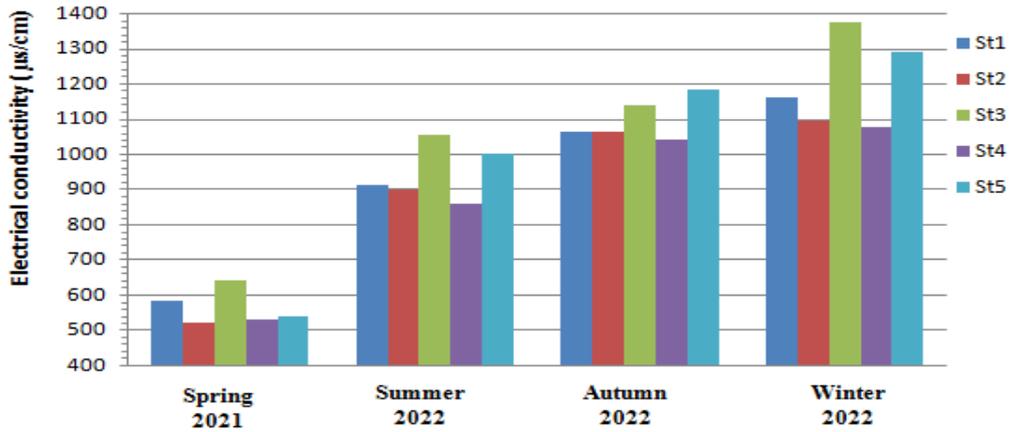


Figure (5): Seasonal changes of Electrical conductivity (µs/cm) for the five stations during the study period from Spring 2021 to Winter 2022

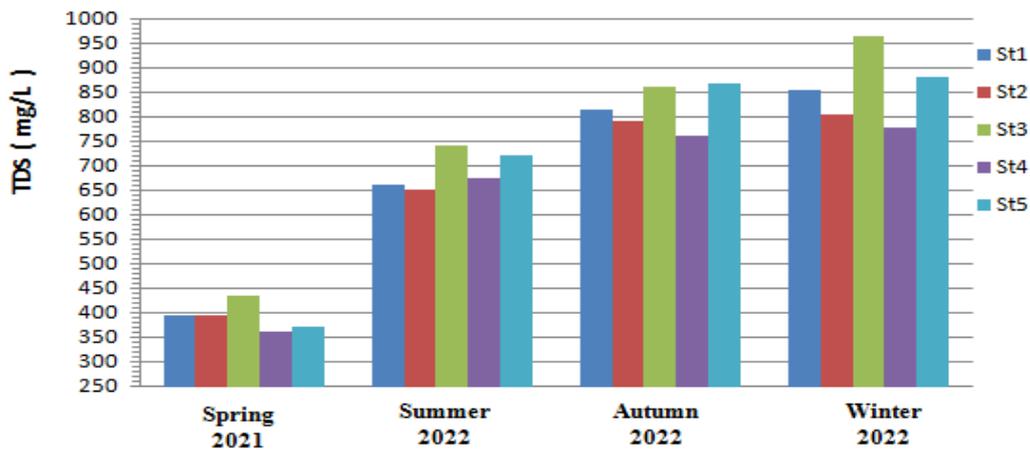


Figure (6): Seasonal changes of TDS (mg/l) for the five stations during the study period from Spring 2021 to Winter 2022

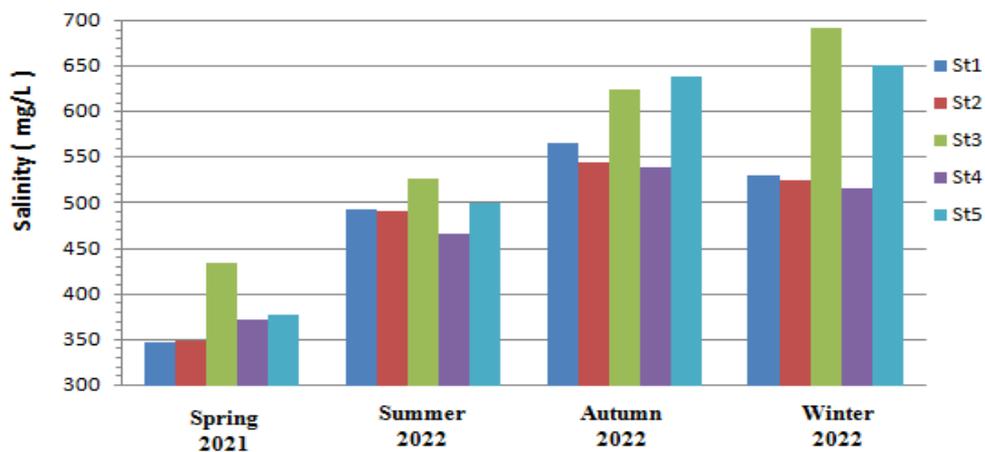
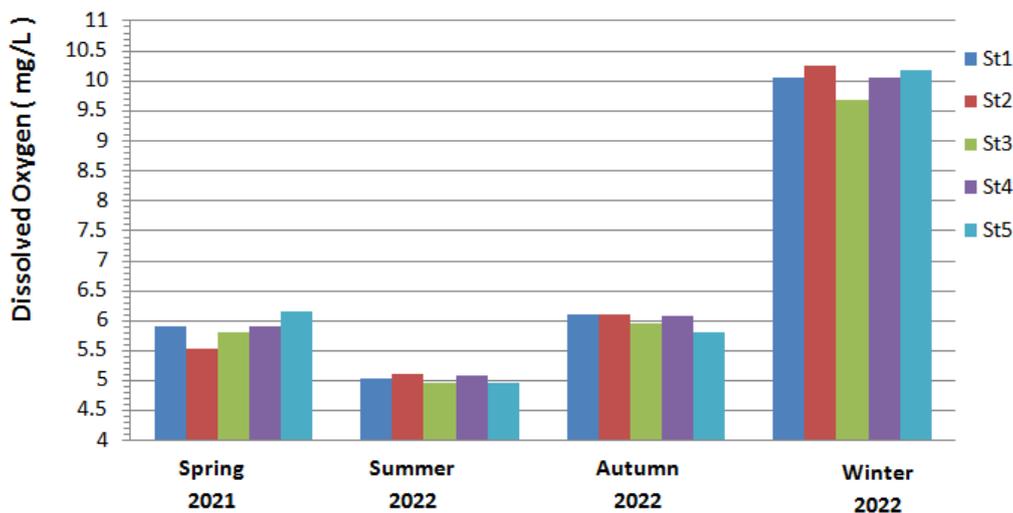


Figure (7): Seasonal changes of Salinity (mg/l) for the five stations during the study period from Spring 2021 to Winter 2022

### 3 – 1-4 : Dissolved Oxygen

Dissolved oxygen levels are found to be greatest in the winter at (10.25) mg/l in station two and lowest in the summer at (4.94) mg/l in station three according to the current investigation (Figure 8). A statistical investigation revealed significant seasonal fluctuations in the concentration of dissolved oxygen. The statistical analysis revealed that there are significant differences between the rates of dissolved oxygen during the spring and winter when comparing the five stations over the period of the study (table 3), but there are not significant differences during the summer or autumn. While the dissolved oxygen rates during the study seasons (table 4) indicated distinct changes that are statistically significant.

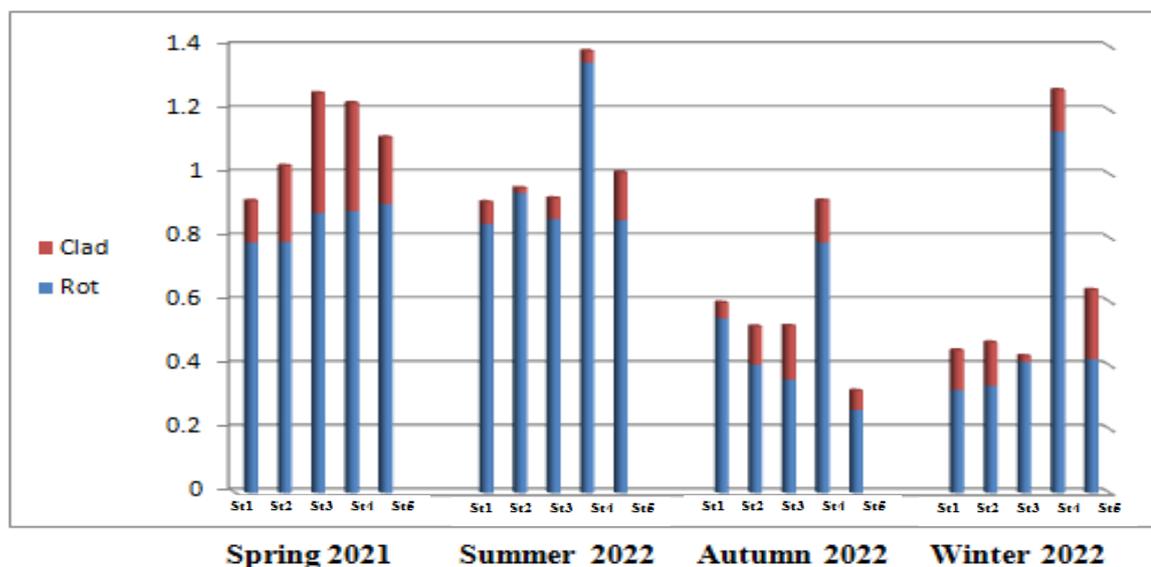


**Figure (8): Seasonal changes of Dissolved Oxygen (mg/l) for the five stations during the study period from spring 2021 to winter 2022**

### 3 -2: Distribution of Zooplankton

The distribution of zooplankton (Ind/l) through study period (Spring 2021 to Winter 2022) at five stations along the Hilla River displayed in (Figure 9). The total number of individuals fluctuated over the period of

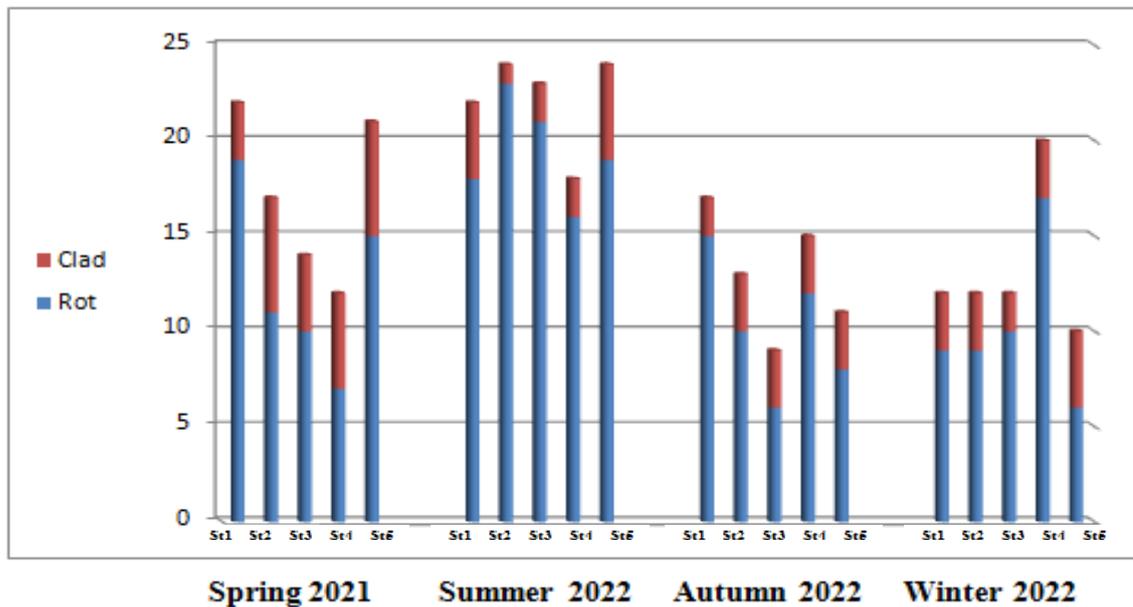
the study with the lowest value at station five in the autumn being (0.32 Ind/l) and the maximum value at station four in the summer being (1.386 Ind/l). In general, the Findings showed that the density is high during summer regarding Rotifera while the density of Cladocera is high during the spring. When comparing the five sites during the the study period, the statistical analysis showed significant differences in the number of individuals during the summer, autumn, and winter (table 3), but not during the spring. While the number of individuals during the period of the research seasons (table 4) showed noticeable variations that were statistically significant.



**Figure (9): Seasonal changes of Zooplankton total density (Ind/l) for the five stations during the study period from Spring 2021 to Winter 2022**

The mean number of taxa in the five stations during the study period is also shown in (Figure 10), the lowest mean number (5.33 taxa) is recorded in the autumn at station five, while the largest mean number (14 taxa) is reported in the summer at stations one, two, and five. Generally, the current results recorded the high number of taxa of rotifera and cladocera is during summer. According to the statistical analysis, there

are no significant differences in the mean number of taxa among the stations and seasons (tables 3-4).



**Figure (10): Seasonal changes in number of Zooplankton taxa for the five stations during the study period from Spring 2021 to Winter 2022**

The seasonal density of Rotifera and Cladocera in station one over the duration of the study is shown in (table 5). The highest mean is observed by Bdelloidea in the winter (0.144 Ind/l), followed by *T.rattus* (0.132 Ind/l) in the spring, *E.dilatata* (0.125 Ind/l) in the spring. While Naupilus, *L.closterocerca* and *T.tetractis* recorded (0.1 Ind/l) during the corresponding seasons of spring and summer, summer and autumn. For cladocera, *E.longispina* had the highest density (0.075 Ind/l) in the winter and *A.angustatus* had the second-highest density (0.056 Ind/l) in the spring.

**Table (5):**The seasonal densities of Rotifera and Cladocera (Ind/l) in station one from Spring 2021 to Winter 2022.

<b>Rotifera Taxa</b>		<b>Spring</b>	<b>Summer</b>	<b>Autumn</b>	<b>Winter</b>	<b>Sum</b>
<i>Ascomorpha ovalis</i>	<b>Sum</b>			<b>0.018</b>		<b>0.018</b>
	<b>mean</b>			<b>0.006</b>		<b>0.006</b>
<i>Ascomorpha saltans</i>	<b>Sum</b>		<b>0.018</b>	<b>0.018</b>		<b>0.036</b>
	<b>mean</b>		<b>0.006</b>	<b>0.006</b>		<b>0.012</b>
<i>Asplanchna priodonta</i>	<b>Sum</b>				<b>0.018</b>	<b>0.018</b>
	<b>mean</b>				<b>0.006</b>	<b>0.006</b>
Bdelloidea	<b>Sum</b>	<b>0.245</b>	<b>0.358</b>	<b>0.339</b>	<b>0.434</b>	<b>1.376</b>
	<b>mean</b>	<b>0.081</b>	<b>0.119</b>	<b>0.113</b>	<b>0.144</b>	<b>0.457</b>
<i>Brachionus forficula</i>	<b>Sum</b>		<b>0.132</b>			<b>0.132</b>
	<b>mean</b>		<b>0.044</b>			<b>0.044</b>
<i>Brachionus rubens</i>	<b>Sum</b>		<b>0.018</b>			<b>0.018</b>
	<b>mean</b>		<b>0.006</b>			<b>0.006</b>
<i>Brachionus</i> spp	<b>Sum</b>			<b>0.018</b>		<b>0.018</b>
	<b>mean</b>			<b>0.006</b>		<b>0.006</b>
<i>Brachionus angularis</i>	<b>Sum</b>				<b>0.018</b>	<b>0.018</b>
	<b>mean</b>				<b>0.006</b>	<b>0.006</b>
<i>Brachionus falcatus</i>	<b>Sum</b>			<b>0.018</b>		<b>0.018</b>
	<b>mean</b>			<b>0.006</b>		<b>0.006</b>
<i>Cephalodella gibba</i>	<b>Sum</b>	<b>0.132</b>	<b>0.207</b>		<b>0.226</b>	<b>0.565</b>
	<b>mean</b>	<b>0.044</b>	<b>0.069</b>		<b>0.075</b>	<b>0.188</b>
<i>Cephalodella</i> sp.	<b>Sum</b>	<b>0.018</b>				<b>0.018</b>
	<b>mean</b>	<b>0.006</b>				<b>0.006</b>
<i>Collotheca</i> spp.	<b>Sum</b>	<b>0.018</b>				<b>0.018</b>
	<b>mean</b>	<b>0.006</b>				<b>0.006</b>
<i>Colurella adriatica</i>	<b>Sum</b>			<b>0.018</b>		<b>0.018</b>
	<b>mean</b>			<b>0.006</b>		<b>0.006</b>
<i>Colurella</i> sp.	<b>Sum</b>	<b>0.150</b>				<b>0.150</b>
	<b>mean</b>	<b>0.050</b>				<b>0.05</b>
<i>Encentrum porsildi</i>	<b>Sum</b>			<b>0.018</b>		<b>0.018</b>
	<b>mean</b>			<b>0.006</b>		<b>0.006</b>
<i>Euchlanis dilatata</i>	<b>Sum</b>	<b>0.377</b>	<b>0.132</b>			<b>0.509</b>
	<b>mean</b>	<b>0.125</b>	<b>0.044</b>			<b>0.169</b>
<i>Horaella brehmi</i>	<b>Sum</b>	<b>0.018</b>				<b>0.018</b>
	<b>mean</b>	<b>0.006</b>				<b>0.006</b>
<i>Keratella tecta</i>	<b>Sum</b>			<b>0.113</b>	<b>0.113</b>	<b>0.226</b>
	<b>mean</b>			<b>0.037</b>	<b>0.037</b>	<b>0.074</b>
<i>Lecane bulla</i>	<b>Sum</b>		<b>0.207</b>	<b>0.207</b>		<b>0.414</b>
	<b>mean</b>		<b>0.069</b>	<b>0.069</b>		<b>0.138</b>
<i>Lecane closterocerca</i>	<b>Sum</b>		<b>0.301</b>			<b>0.301</b>
	<b>mean</b>		<b>0.100</b>			<b>0.100</b>
<i>Lecane elsa</i>	<b>Sum</b>	<b>0.132</b>				<b>0.132</b>
	<b>mean</b>	<b>0.044</b>				<b>0.044</b>
<i>Lecane hamata</i>	<b>Sum</b>	<b>0.018</b>				<b>0.018</b>
	<b>mean</b>	<b>0.006</b>				<b>0.006</b>

<i>Lecane lunaris</i>	Sum		0.207			0.207
	mean		0.069			0.069
<i>Lecane nana</i>	Sum		0.018			0.018
	mean		0.006			0.006
<i>Lecane quadridentata</i>	Sum		0.207			0.207
	mean		0.069			0.069
<i>Lecane</i> spp.	Sum		0.018	0.113		0.131
	mean		0.006	0.037		0.043
<i>Lepadella patella</i>	Sum	0.018	0.018			0.036
	mean	0.006	0.006			0.012
<i>Lepadella</i> spp.	Sum			0.018		0.018
	mean			0.006		0.006
<i>Macrochaetus sericus</i>	Sum				0.018	0.018
	mean				0.006	0.006
<i>Macrotrachela angusta</i>	Sum		0.018			0.018
	mean		0.006			0.006
<i>Mytilina ventralis</i>	Sum			0.037		0.037
	mean			0.012		0.012
Nauplius	Sum	0.301	0.301	0.207		0.809
	mean	0.100	0.100	0.069		0.269
<i>Platyias quadricornis</i>	Sum	0.113				0.113
	mean	0.037				0.037
<i>Polyarthra dolichoptera</i>	Sum	0.132				0.132
	mean	0.044				0.044
<i>Polyarthra major</i>	Sum				0.113	0.113
	mean				0.037	0.037
<i>Rotaria</i> spp.	Sum	0.018				0.018
	mean	0.006				0.006
Other Rotifera spp	Sum	0.018				0.018
	mean	0.006				0.006
<i>Scaridium longicaudum</i>	Sum	0.018				0.018
	mean	0.006				0.006
<i>Synchaeta lakowitziana</i>	Sum				0.018	0.018
	mean				0.006	0.006
<i>Synchaeta</i> spp.	Sum				0.018	0.018
	mean				0.006	0.006
<i>Trichocerca rattus</i>	Sum	0.396				0.396
	mean	0.132				0.132
<i>Testudinella patina</i>	Sum		0.132	0.207		0.339
	mean		0.044	0.069		0.113
<i>Testudinella reflexa</i>	Sum		0.018			0.018
	mean		0.006			0.006
<i>Trichotria pocillum</i>	Sum	0.113				0.113
	mean	0.037				0.037
<i>Trichotria tetractis</i>	Sum	0.132	0.226	0.301		0.659
	mean	0.044	0.075	0.100		0.219
<b>Cladocera Taxa</b>						
<i>Acantholeberis curvirostris</i>	Sum		0.150			0.150

	<b>mean</b>		<b>0.050</b>			<b>0.050</b>
<i>Acroperus angustatus</i>	<b>Sum</b>	<b>0.169</b>	<b>0.018</b>			<b>0.187</b>
	<b>mean</b>	<b>0.056</b>	<b>0.006</b>			<b>0.062</b>
Bosminidae	<b>Sum</b>			<b>0.037</b>		<b>0.037</b>
	<b>mean</b>			<b>0.012</b>		<b>0.012</b>
Chydoridae	<b>Sum</b>		<b>0.018</b>			<b>0.018</b>
	<b>mean</b>		<b>0.006</b>			<b>0.006</b>
<i>Daphnia magna</i>	<b>Sum</b>	<b>0.113</b>			<b>0.113</b>	<b>0.226</b>
	<b>mean</b>	<b>0.037</b>			<b>0.037</b>	<b>0.074</b>
Daphniidae	<b>Sum</b>				<b>0.037</b>	<b>0.037</b>
	<b>mean</b>				<b>0.012</b>	<b>0.012</b>
<i>Eubosmina longispina</i>	<b>Sum</b>	<b>0.113</b>		<b>0.113</b>	<b>0.226</b>	<b>0.452</b>
	<b>mean</b>	<b>0.037</b>		<b>0.037</b>	<b>0.075</b>	<b>0.149</b>
Other Cladocera spp	<b>Sum</b>		<b>0.018</b>			<b>0.018</b>
	<b>mean</b>		<b>0.006</b>			<b>0.006</b>
Total density (Ind/l)		<b>0.916</b>	<b>0.912</b>	<b>0.597</b>	<b>0.447</b>	<b>2.872</b>
No of Taxa recorded in each season		<b>22</b>	<b>22</b>	<b>17</b>	<b>12</b>	
Total No. of Taxa	<b>53</b>					

The highest density among Rotifera in station two is recorded by *T.rattus* (0.158 Ind/l) followed by Nauplius (0.150 Ind/l) during spring (table 6). Then *E.dilatata* (0.128 Ind/l) during spring and summer followed by Bdelloidea (0.105 Ind/l) during spring. While *K.cochlearis* and *T.patina* (0.090 Ind/l) are recorded during spring and summer, respectively. *P.major* and *T.tetractis* had mean of (0.083 Ind/l) in the winter and spring, respectively. For Cladocera, *E.longispina* had the maximum density (0.120 Ind/l) followed by *D.magna* (0.045 Ind/l) during the spring.

**Table (6):**The seasonal densities of rotifera and cladocera (Ind/l) in station two from Spring 2021 to Winter 2022.

<b>Rotifera Taxa</b>		<b>Spring</b>	<b>Summer</b>	<b>Autumn</b>	<b>Winter</b>	<b>Sum</b>
Bdelloidea	<b>Sum</b>	<b>0.317</b>	<b>0.271</b>	<b>0.294</b>		<b>0.882</b>
	<b>mean</b>	<b>0.105</b>	<b>0.090</b>	<b>0.098</b>		<b>0.293</b>
<i>Brachionus forficula</i>	<b>Sum</b>		<b>0.045</b>			<b>0.045</b>
	<b>mean</b>		<b>0.015</b>			<b>0.015</b>
<i>Brachionus calyciflorus</i>	<b>Sum</b>				<b>0.135</b>	<b>0.135</b>
	<b>mean</b>				<b>0.045</b>	<b>0.045</b>
<i>Cephalodella gibba</i>	<b>Sum</b>	<b>0.135</b>			<b>0.135</b>	<b>0.270</b>
	<b>mean</b>	<b>0.045</b>			<b>0.045</b>	<b>0.090</b>
<i>Cephalodella</i> sp.	<b>Sum</b>	<b>0.022</b>				<b>0.022</b>
	<b>mean</b>	<b>0.007</b>				<b>0.007</b>
<i>Encentrum porsildi</i>	<b>Sum</b>		<b>0.022</b>			<b>0.022</b>
	<b>mean</b>		<b>0.007</b>			<b>0.007</b>
<i>Euchlanis dilatata</i>	<b>Sum</b>	<b>0.384</b>	<b>0.384</b>			<b>0.768</b>
	<b>mean</b>	<b>0.128</b>	<b>0.128</b>			<b>0.256</b>
<i>Euchlanis</i> spp	<b>Sum</b>		<b>0.022</b>			<b>0.022</b>
	<b>mean</b>		<b>0.007</b>			<b>0.007</b>
<i>Filinia opoliensis</i>	<b>Sum</b>		<b>0.158</b>			<b>0.158</b>
	<b>mean</b>		<b>0.052</b>			<b>0.052</b>
<i>Keratella cochlearis</i>	<b>Sum</b>	<b>0.271</b>	<b>0.249</b>	<b>0.158</b>		<b>0.678</b>
	<b>mean</b>	<b>0.090</b>	<b>0.083</b>	<b>0.052</b>		<b>0.225</b>
<i>Keratella quadrata</i>	<b>Sum</b>				<b>0.135</b>	<b>0.135</b>
	<b>mean</b>				<b>0.045</b>	<b>0.045</b>
<i>Keratella</i> spp.	<b>Sum</b>	<b>0.022</b>		<b>0.022</b>		<b>0.044</b>
	<b>mean</b>	<b>0.007</b>		<b>0.007</b>		<b>0.014</b>
<i>Keratella tecta</i>	<b>Sum</b>				<b>0.022</b>	<b>0.022</b>
	<b>mean</b>				<b>0.007</b>	<b>0.007</b>
<i>Keratella tropica</i>	<b>Sum</b>		<b>0.135</b>	<b>0.135</b>		<b>0.270</b>
	<b>mean</b>		<b>0.045</b>	<b>0.045</b>		<b>0.090</b>
<i>Lecane bulla</i>	<b>Sum</b>		<b>0.135</b>	<b>0.022</b>		<b>0.157</b>
	<b>mean</b>		<b>0.045</b>	<b>0.007</b>		<b>0.052</b>
<i>Lecane closterocerca</i>	<b>Sum</b>			<b>0.135</b>		<b>0.135</b>
	<b>mean</b>			<b>0.045</b>		<b>0.045</b>
<i>Lecane hamata</i>	<b>Sum</b>		<b>0.022</b>			<b>0.022</b>
	<b>mean</b>		<b>0.007</b>			<b>0.007</b>
<i>Lecane ludwigii</i>	<b>Sum</b>		<b>0.022</b>			<b>0.022</b>
	<b>mean</b>		<b>0.007</b>			<b>0.007</b>
<i>Lecane lunaris</i>	<b>Sum</b>		<b>0.135</b>			<b>0.135</b>
	<b>mean</b>		<b>0.045</b>			<b>0.045</b>
<i>Lecane nana</i>	<b>Sum</b>		<b>0.022</b>			<b>0.022</b>
	<b>mean</b>		<b>0.007</b>			<b>0.007</b>
<i>Lecane quadridentata</i>	<b>Sum</b>		<b>0.135</b>			<b>0.135</b>
	<b>mean</b>		<b>0.045</b>			<b>0.045</b>
<i>Lecane</i> spp.	<b>Sum</b>		<b>0.022</b>			<b>0.022</b>
	<b>mean</b>		<b>0.007</b>			<b>0.007</b>

<i>Lecane tenuiseta</i>	Sum		0.022			0.022
	mean		0.007			0.007
<i>Lecane thienemanni</i>	Sum			0.022		0.022
	mean			0.007		0.007
<i>Lecane ungulata</i>	Sum		0.022			0.022
	mean		0.007			0.007
<i>Macrotrachela angusta</i>	Sum		0.135			0.135
	mean		0.045			0.045
Nauplius	Sum	0.452	0.181			0.633
	mean	0.150	0.060			0.21
<i>Platyias quadricornis</i>	Sum		0.158			0.158
	mean		0.052			0.052
<i>Polyarthra major</i>	Sum				0.249	0.249
	mean				0.083	0.083
Other Rotifera spp	Sum		0.022			0.022
	mean		0.007			0.007
<i>Scaridium longicaudum</i>	Sum				0.022	0.022
	mean				0.007	0.007
<i>Synchaeta</i> spp.	Sum	0.022			0.045	0.067
	mean	0.007			0.015	0.022
<i>Trichocerca rattus</i>	Sum	0.475		0.271		0.746
	mean	0.158		0.090		0.248
<i>Trichocerca</i> spp	Sum	0.022				0.022
	mean	0.007				0.007
<i>Testudinella patina</i>	Sum		0.271	0.135	0.135	0.541
	mean		0.090	0.045	0.045	0.18
<i>Trichotria tetractis</i>	Sum	0.249	0.249	0.022	0.135	0.655
	mean	0.083	0.083	0.007	0.045	0.218
<b>Cladocera Taxa</b>						
<i>Acantholeberis curvirostris</i>	Sum	0.135	0.045	0.135	0.249	0.564
	mean	0.045	0.015	0.045	0.083	0.188
<i>Camptocercus</i> spp	Sum	0.022				0.022
	mean	0.007				0.007
<i>Daphnia magna</i>	Sum	0.135				0.135
	mean	0.045				0.045
Daphniidae	Sum	0.045				0.045
	mean	0.015				0.015
<i>Eubosmina longispina</i>	Sum	0.362		0.158	0.135	0.655
	mean	0.120		0.052	0.045	0.217
Other Cladocera spp	Sum	0.022		0.067	0.022	0.111
	mean	0.007		0.022	0.007	0.036
Total density (Ind/l)		1.026	0.956	0.522	0.472	2.976
No of Taxa recorded in each season		17	24	13	12	
Total No.of Taxa	42					

In station three, during the summer and spring respectively, Bdelloidea and Nauplius reported the highest Rotifera density (0.179 Ind/l). It is followed by *E.dilatata* (0.155 Ind/l) during spring, *T.tetractis* (0.113 Ind/l) during summer, *P.dolichoptera* (0.103 Ind/l) during spring and *Synchaeta* spp (0.075 Ind/l) during winter. As for Cladocera, *A. angustatus* recorded the highest mean (0.122 Ind/l) followed by *E.longispina* (0.113 Ind/l) during spring (table 7).

**Table (7):The seasonal densities of rotifera and cladocera (Ind/l) in station three from Spring 2021 to Winter 2022.**

<b>Rotifera Taxa</b>		<b>Spring</b>	<b>Summer</b>	<b>Autumn</b>	<b>Winter</b>	<b>Sum</b>
Bdelloidea	<b>Sum</b>	<b>0.368</b>	<b>0.537</b>	<b>0.339</b>	<b>0.368</b>	<b>1.612</b>
	<b>mean</b>	<b>0.122</b>	<b>0.179</b>	<b>0.113</b>	<b>0.122</b>	<b>0.536</b>
<i>Brachionus forficula</i>	<b>Sum</b>		<b>0.028</b>			<b>0.028</b>
	<b>mean</b>		<b>0.009</b>			<b>0.009</b>
<i>Brachionus calyciflorus</i>	<b>Sum</b>		<b>0.169</b>			<b>0.169</b>
	<b>mean</b>		<b>0.056</b>			<b>0.056</b>
<i>Brachionus falcatus</i>	<b>Sum</b>		<b>0.198</b>			
	<b>mean</b>		<b>0.066</b>			
<i>Cephalodella gibba</i>	<b>Sum</b>	<b>0.198</b>	<b>0.169</b>			<b>0.367</b>
	<b>mean</b>	<b>0.066</b>	<b>0.056</b>			<b>0.122</b>
<i>Collotheca</i> spp.	<b>Sum</b>			<b>0.198</b>		<b>0.198</b>
	<b>mean</b>			<b>0.066</b>		<b>0.066</b>
<i>Colurella adriatica</i>	<b>Sum</b>				<b>0.028</b>	<b>0.028</b>
	<b>mean</b>				<b>0.009</b>	<b>0.009</b>
<i>Dicranophorus prionacis</i>	<b>Sum</b>		<b>0.056</b>			<b>0.056</b>
	<b>mean</b>		<b>0.018</b>			<b>0.018</b>
<i>Encentrum porsildi</i>	<b>Sum</b>		<b>0.028</b>		<b>0.028</b>	<b>0.056</b>
	<b>mean</b>		<b>0.009</b>		<b>0.009</b>	<b>0.018</b>
<i>Euchlanis dilatata</i>	<b>Sum</b>	<b>0.467</b>	<b>0.028</b>			<b>0.495</b>
	<b>mean</b>	<b>0.155</b>	<b>0.009</b>			<b>0.164</b>
<i>Filinia opoliensis</i>	<b>Sum</b>		<b>0.028</b>	<b>0.169</b>		<b>0.197</b>
	<b>mean</b>		<b>0.009</b>	<b>0.056</b>		<b>0.065</b>
<i>Keratella tecta</i>	<b>Sum</b>				<b>0.169</b>	<b>0.169</b>
	<b>mean</b>				<b>0.056</b>	<b>0.056</b>
<i>Keratella tropica</i>	<b>Sum</b>		<b>0.198</b>			<b>0.198</b>
	<b>mean</b>		<b>0.066</b>			<b>0.066</b>
<i>Lecane arcula</i>	<b>Sum</b>		<b>0.028</b>			<b>0.028</b>
	<b>mean</b>		<b>0.009</b>			<b>0.009</b>
<i>Lecane bulla</i>	<b>Sum</b>	<b>0.198</b>				<b>0.198</b>
	<b>mean</b>	<b>0.066</b>				<b>0.066</b>
<i>Lecane hamata</i>	<b>Sum</b>			<b>0.169</b>		<b>0.169</b>
	<b>mean</b>			<b>0.056</b>		<b>0.056</b>

<i>Lecane luna</i>	Sum		0.028			0.028
	mean		0.009			0.009
<i>Lecane lunaris</i>	Sum		0.169			0.169
	mean		0.056			0.056
<i>Lecane quadridentata</i>	Sum		0.169			0.169
	mean		0.056			0.056
<i>Lecane</i> spp.	Sum			0.028		0.028
	mean			0.009		0.009
<i>Lecane ungulata</i>	Sum		0.028			0.028
	mean		0.009			0.009
<i>Macrochaetus</i> spp	Sum	0.028				0.028
	mean	0.009				0.009
Nauplius	Sum	0.537	0.198		0.028	0.763
	mean	0.179	0.066		0.009	0.254
<i>Platylabus quadricornis</i>	Sum		0.169			0.169
	mean		0.056			0.056
<i>Polyarthra dolichoptera</i>	Sum	0.311		0.169		0.48
	mean	0.103		0.056		0.159
<i>Polyarthra major</i>	Sum				0.028	0.028
	mean				0.009	0.009
<i>Polyarthra minor</i>	Sum		0.028			0.028
	mean		0.009			0.009
<i>Polyarthra remata</i>	Sum	0.028			0.028	0.056
	mean	0.009			0.009	0.018
Other Rotifera spp	Sum	0.028				0.028
	mean	0.009				0.009
<i>Synchaeta</i> spp.	Sum				0.226	0.226
	mean				0.075	0.075
<i>Trichocerca rattus</i>	Sum	0.481				0.481
	mean	0.160				0.160
<i>Trichocerca similis</i>	Sum		0.028			0.028
	mean		0.009			0.009
<i>Testudinella patina</i>	Sum		0.169		0.169	0.338
	mean		0.056		0.056	0.112
<i>Trichotria tetractis</i>	Sum		0.339		0.198	0.537
	mean		0.113		0.066	0.179
<b>Cladocera Taxa</b>						
<i>Acantholeberis curvirostris</i>	Sum		0.028		0.028	0.056
	mean		0.009		0.009	0.018
<i>Acroperus angustatus</i>	Sum	0.368		0.169	0.028	0.565
	mean	0.122		0.056	0.009	0.187
<i>Camptocercus</i> spp	Sum	0.028				0.028
	mean	0.009				0.009
<i>Eubosmina longispina</i>	Sum	0.339	0.169	0.311		0.819
	mean	0.113	0.056	0.103		0.272
Other Cladocera spp	Sum	0.396		0.028		0.424
	mean	0.132		0.009		0.141
Total density (Ind/l)		1.254	0.924	0.524	0.429	3.131

No of Taxa recorded in each season		14	23	9	12	
Total No of Taxa	39					

According to (table 8), Bdelloidea, *E. dilatata* and *T. rattus* each had the maximum density of rotifers in the spring (0.217 Ind/l), followed by *T. tetractis* (0.169 Ind/l) in the winter, Nauplius (0.160 Ind/l) in the spring, and *L. bulla* (0.150 Ind/l) in the summer. *A. angustatus* had the highest rate of Cladocera during the spring (0.132 Ind/l) followed by *E. longispina* (0.113 Ind/l) in station four.

**Table (8):**The seasonal densities of rotifera and cladocera (Ind/l) in station four from Spring 2021 to Winter 2022.

Rotifera Taxa		Spring	Summer	Autumn	Winter	Sum
Bdelloidea	Sum	0.651	0.594	0.339	0.651	2.235
	mean	0.217	0.198	0.113	0.217	0.745
<i>Brachionus forficula</i>	Sum		0.169			0.169
	mean		0.056			0.056
<i>Brachionus calyciflorus</i>	Sum				0.169	0.169
	mean				0.056	0.056
<i>Cephalodella gibba</i>	Sum	0.028	0.424		0.198	0.65
	mean	0.009	0.141		0.066	0.216
<i>Collotheca</i> spp.	Sum	0.169				0.169
	mean	0.056				0.056
<i>Colurella adriatica</i>	Sum				0.028	0.028
	mean				0.009	0.009
<i>Euchlanis dilatata</i>	Sum	0.651	0.481			1.132
	mean	0.217	0.160			0.377
<i>Filinia opoliensis</i>	Sum			0.028		0.028
	mean			0.009		0.009
<i>Filinia terminalis</i>	Sum			0.028		0.028
	mean			0.009		0.009
<i>Keratella cochlearis</i>	Sum			0.311	0.056	0.367
	mean			0.103	0.018	0.121
<i>Keratella tecta</i>	Sum		0.169			0.169
	mean		0.056			0.056
<i>Keratella valga</i>	Sum			0.028		0.028
	mean			0.009		0.009
<i>Lecane bulla</i>	Sum		0.452	0.169	0.198	0.819
	mean		0.150	0.056	0.066	0.272
<i>Lecane closterocerca</i>	Sum				0.339	0.339

	mean				0.113	0.113
<i>Lecane elsa</i>	Sum		0.028			0.028
	mean		0.009			0.009
<i>Lecane hamata</i>	Sum	0.028				0.028
	mean	0.009				0.009
<i>Lecane lunaris</i>	Sum				0.169	0.169
	mean				0.056	0.056
<i>Lecane spp.</i>	Sum		0.028		0.028	0.056
	mean		0.009		0.009	0.018
<i>Lecane ungulata</i>	Sum		0.028			0.028
	mean		0.009			0.009
<i>Lepadella spp.</i>	Sum				0.028	0.028
	mean				0.009	0.009
<i>Macrotrachela angusta</i>	Sum		0.056			0.056
	mean		0.018			0.018
Nauplius	Sum	0.481	0.028		0.169	0.678
	mean	0.160	0.009		0.056	0.225
<i>Platytas quadricornis</i>	Sum		0.311	0.311		0.622
	mean		0.103	0.103		0.206
<i>Polyarthra dolichoptera</i>	Sum		0.311	0.169	0.311	0.791
	mean		0.103	0.056	0.103	0.262
<i>Polyarthra major</i>	Sum				0.169	0.169
	mean				0.056	0.056
Other Rotifera spp	Sum			0.028	0.169	0.197
	mean			0.009	0.056	0.065
<i>Synchaeta spp.</i>	Sum				0.028	0.028
	mean				0.009	0.009
<i>Trichocerca rattus</i>	Sum	0.651	0.481	0.622		1.754
	mean	0.217	0.160	0.207		0.584
<i>Testudinella patina</i>	Sum		0.339	0.311	0.198	0.848
	mean		0.113	0.103	0.066	0.282
<i>Trichotria tetractis</i>	Sum		0.169	0.028	0.509	0.706
	mean		0.056	0.009	0.169	0.234
<b>Cladocera Taxa</b>						
<i>Acroperus angustatus</i>	Sum	0.396				0.396
	mean	0.132				0.132
<i>Alona guttata</i>	Sum		0.056			0.056
	mean		0.018			0.018
<i>Alonella exigua</i>	Sum			0.028		0.028
	mean			0.009		0.009
<i>Camptocercus uncinatus</i>	Sum			0.169		0.169
	mean			0.056		0.056
<i>Daphnia magna</i>	Sum	0.169			0.028	0.197
	mean	0.056			0.009	0.065
Daphniidae	Sum	0.028				0.028
	mean	0.009				0.009
<i>Eubosmina longispina</i>	Sum	0.339	0.056	0.198	0.311	0.904
	mean	0.113	0.018	0.066	0.103	0.3

Other Cladocera spp	Sum	0.084			0.056	0.14
	mean	0.028			0.018	0.046
Total density (Ind/l)		1.223	1.386	0.917	1.264	4.79
No of Taxa recorded in each season		12	18	15	20	
Total No.of Taxa	38					

With regard to station five, the (table 9) revealed that Bdelloidea had the highest density of rotifera (0.211 Ind/l) during the winter, followed by *E.dilatata* (0.166 Ind/l) during the summer, Nauplius (0.143 Ind/l) in the spring, *T.rattus* (0.128 Ind/l) during the winter, and *T.patina* (0.120 Ind/l) during the summer. The greatest mean values for Cladocera are obtained by *A.angustatus* and *E.longispina* (0.090 Ind/l) in the winter and spring, respectively followed by *A.curvirostris* in the summer (0.083 Ind/l).

**Table (9): The seasonal densities of rotifera and cladocera (Ind/l) in station five from Spring 2021 to Winter 2022.**

Rotifera Taxa		Spring	Summer	Autumn	Winter	Sum
Bdelloidea	Sum	0.543	0.271	0.249	0.643	1.706
	mean	0.181	0.090	0.083	0.211	0.565
<i>Brachionus forficula</i>	Sum		0.022			0.022
	mean		0.007			0.007
<i>Brachionus rubens</i>	Sum		0.022			0.022
	mean		0.007			0.007
<i>Brachionus angularis</i>	Sum		0.022			0.022
	mean		0.007			0.007
<i>Brachionus budapestinensis</i>	Sum		0.022			0.022
	mean		0.007			0.007
<i>Brachionus calyciflorus</i>	Sum	0.158	0.158			0.316
	mean	0.052	0.052			0.104
<i>Brachionus quadridentatus</i>	Sum	0.022				0.022
	mean	0.007				0.007
<i>Colurella adriatica</i>	Sum			0.022		0.022
	mean			0.007		0.007
<i>Dicranophorus prionacis</i>	Sum		0.022	0.022		0.044
	mean		0.007	0.007		0.014
<i>Encentrum porsildi</i>	Sum			0.045		0.045
	mean			0.015		0.015
<i>Euchlanis dilatata</i>	Sum	0.362	0.498			0.86
	mean	0.120	0.166			0.286
<i>Filinia opoliensis</i>	Sum		0.158			0.158

	mean		0.052			0.052
<i>Horaella brehmi</i>	Sum	0.045				0.045
	mean	0.015				0.015
<i>Keratella cochlearis</i>	Sum	0.249				0.249
	mean	0.083				0.083
<i>Keratella tecta</i>	Sum	0.022				0.022
	mean	0.007				0.007
<i>Lecane bulla</i>	Sum	0.022	0.271			0.293
	mean	0.007	0.090			0.097
<i>Lecane closterocerca</i>	Sum	0.135			0.271	0.406
	mean	0.045			0.090	0.135
<i>Lecane elsa</i>	Sum		0.022		0.022	0.044
	mean		0.007		0.007	0.014
<i>Lecane inermis</i>	Sum			0.022		0.022
	mean			0.007		0.007
<i>Lecane lunaris</i>	Sum	0.022				0.022
	mean	0.007				0.007
<i>Lecane nana</i>	Sum		0.135			0.135
	mean		0.045			0.045
Nauplius	Sum	0.430	0.022			0.452
	mean	0.143	0.007			0.15
<i>Platytias quadricornis</i>	Sum		0.271			0.271
	mean		0.090			0.09
<i>Polyarthra major</i>	Sum	0.158	0.022		0.045	0.225
	mean	0.052	0.007		0.015	0.074
<i>Polyarthra minor</i>	Sum				0.135	0.135
	mean				0.045	0.045
Other Rotifera spp	Sum		0.022			0.022
	mean		0.007			0.007
<i>Synchaeta</i> spp.	Sum	0.022		0.022		0.044
	mean	0.007		0.007		0.014
<i>Trichocerca rattus</i>	Sum	0.271	0.158	0.384		0.813
	mean	0.090	0.052	0.128		0.27
<i>Testudinella patina</i>	Sum	0.271	0.362	0.022	0.158	0.813
	mean	0.090	0.120	0.007	0.052	0.269
<i>Trichotria tetractis</i>	Sum		0.135			0.135
	mean		0.045			0.045
<b>Cladocera Taxa</b>						
<i>Acantholeberis curvirostris</i>	Sum	0.022	0.249			0.271
	mean	0.007	0.083			0.09
<i>Acroperus angustatus</i>	Sum	0.249	0.022		0.271	0.542
	mean	0.083	0.007		0.090	0.18
<i>Alona</i> spp	Sum			0.022	0.022	0.044
	mean			0.007	0.007	0.014
<i>Anthalona verrucosa</i>	Sum			0.022		0.022
	mean			0.007		0.007
<i>Camptocercus</i> spp	Sum		0.022			0.022
	mean		0.007			0.007

<i>Daphnia magna</i>	Sum	0.022	0.135			0.157
	mean	0.007	0.045			0.052
Daphniidae	Sum	0.022				0.022
	mean	0.007				0.007
<i>Eubosmina longispina</i>	Sum	0.271		0.135	0.249	0.655
	mean	0.090		0.045	0.083	0.218
Other Cladocera spp	Sum	0.045	0.022		0.113	0.18
	mean	0.015	0.007		0.037	0.059
Total density (Ind/l)		1.115	1.014	0.32	0.637	3.086
No of Taxa recorded in each season		21	24	11	10	
Total No.of Taxa	39					

Finally, the results of the current study demonstrated that, when compared to other rotifera species, Bdelloidea, Nauplius, *E.dilatata*, *T.rattus* and *T.tetractis* species are the most abundant during the study period and at all stations. When compared to the other Cladocera, the species *A.angustatus* and *E.longispina* are the most dense.

### **3–3: The correlation between the environmental factors, rotifera and cladocera**

The correlation between the environmental factors and the species of rotifera and cladocera, as well as the environmental factors among them and the species of rotifera and cladocera among them using SPSS (Appendixes 1-5 ) and CCA (Figures 11-13) and (table 10).

According to the statistical analysis, the air and water temperatures were positively correlated ( $p \leq 0.05$ ) at stations one ( $r=0.991$ ), station two ( $r=0.991$ ), station three ( $r=0.982$ ), stations four ( $r=0.992$ ) and station five ( $r=0.992$ ).

The statistical analysis indicated a negative significant correlation ( $p \leq 0.05$ ) between water temperature and dissolved oxygen respectively in

stations one, two, three and four ( $r = -0.972$ ,  $r = -0.951$ ,  $r = -0.980$  and  $r = -0.960$ ).

In station two, pH exhibited a negative significant correlation ( $p \leq 0.05$ ) with EC, TDS and salinity respectively ( $r = -0.966$ ,  $r = -0.963$  and  $r = -0.995$ ). While in station four, the pH had a negative correlation  $p \leq 0.05$  with EC, salinity and TDS respectively ( $r = -0.983$ ,  $r = -0.972$  and  $r = -0.999$ ).

EC showed a positive significant correlation with TDS ( $p \leq 0.05$ ,  $r = 0.997$ ) in station one. Meanwhile, the E.C showed a positive significant correlation ( $p \leq 0.05$ ) with TDS and salinity respectively ( $r = 0.999$  and  $r = 0.986$ ) in station two. Additionally, EC demonstrated a positive significant correlation ( $p \leq 0.05$ ) with TDS and salinity in stations three and four respectively ( $r = 0.991$  and  $r = 0.964$ ), ( $r = 0.987$  and  $r = 0.983$ ). In station five, there is also a positive significant correlation ( $r = 0.993$ ) between EC and TDS and with salinity ( $r = 0.974$ ).

In stations one, two, three, four and five respectively, TDS revealed a positive significant correlation ( $p \leq 0.05$ ) with salinity ( $r = 0.965$ ,  $r = 0.983$ ,  $r = 0.971$ ,  $r = 0.972$  and  $r = 0.970$ ).

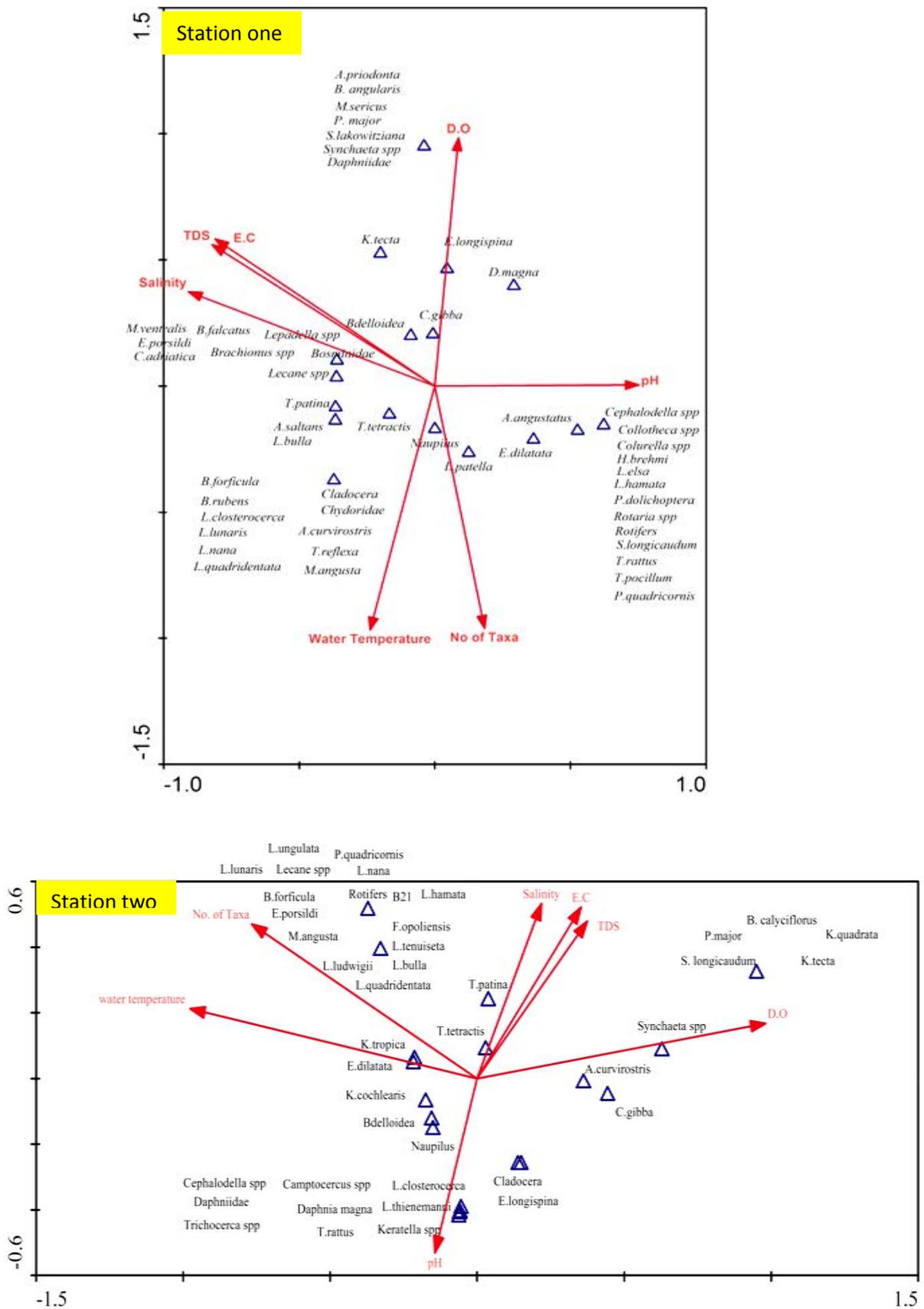


Figure (11): Canonical correspondence analysis (CCA) analysis of the correlation coefficients between rotifera and cladocera taxa and the environmental factors for stations one and two during the study period.

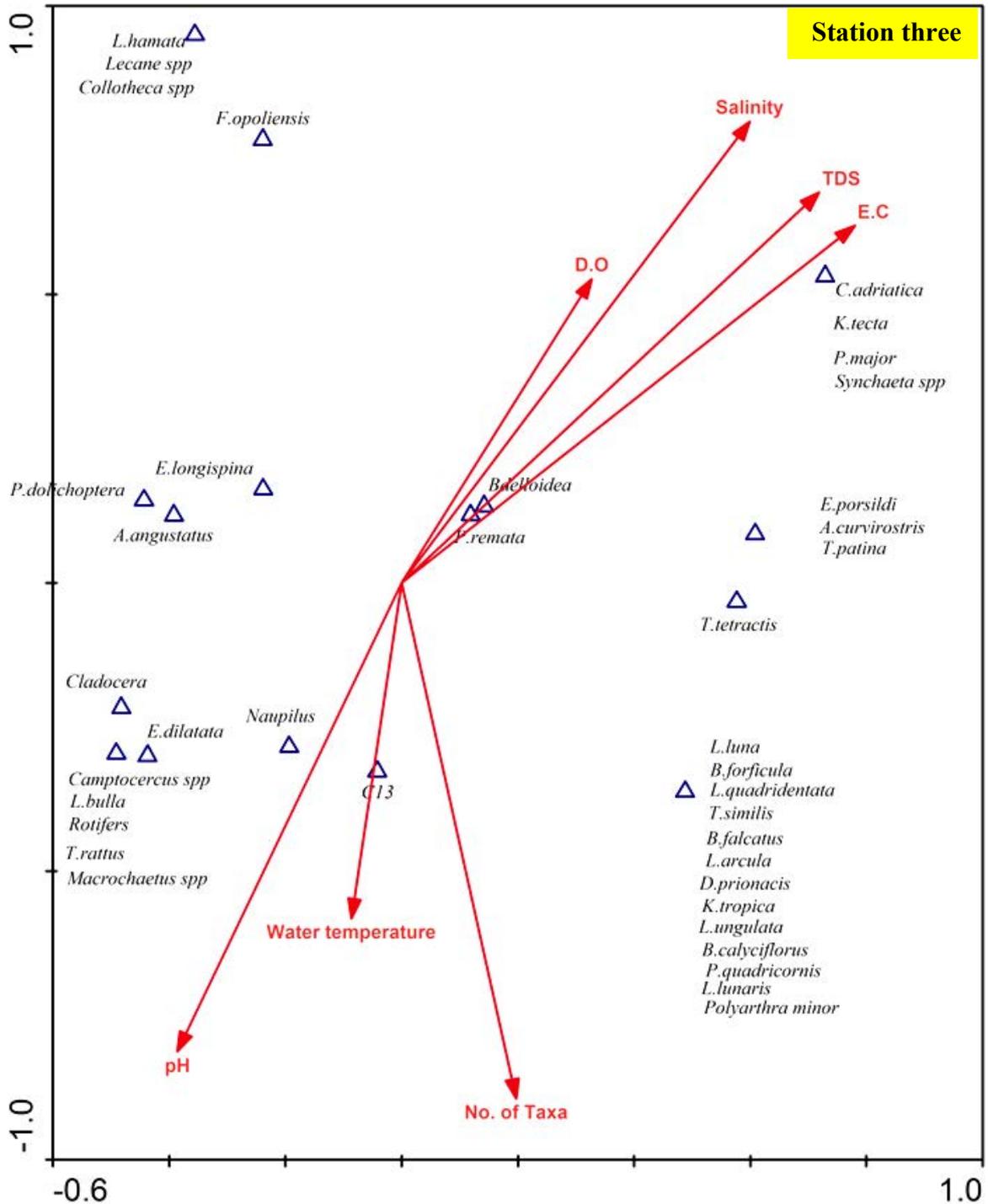


Figure (12): Canonical correspondence analysis (CCA) analysis of the correlation coefficients between rotifera and cladocera taxa and the environmental factors for station three during the study period.

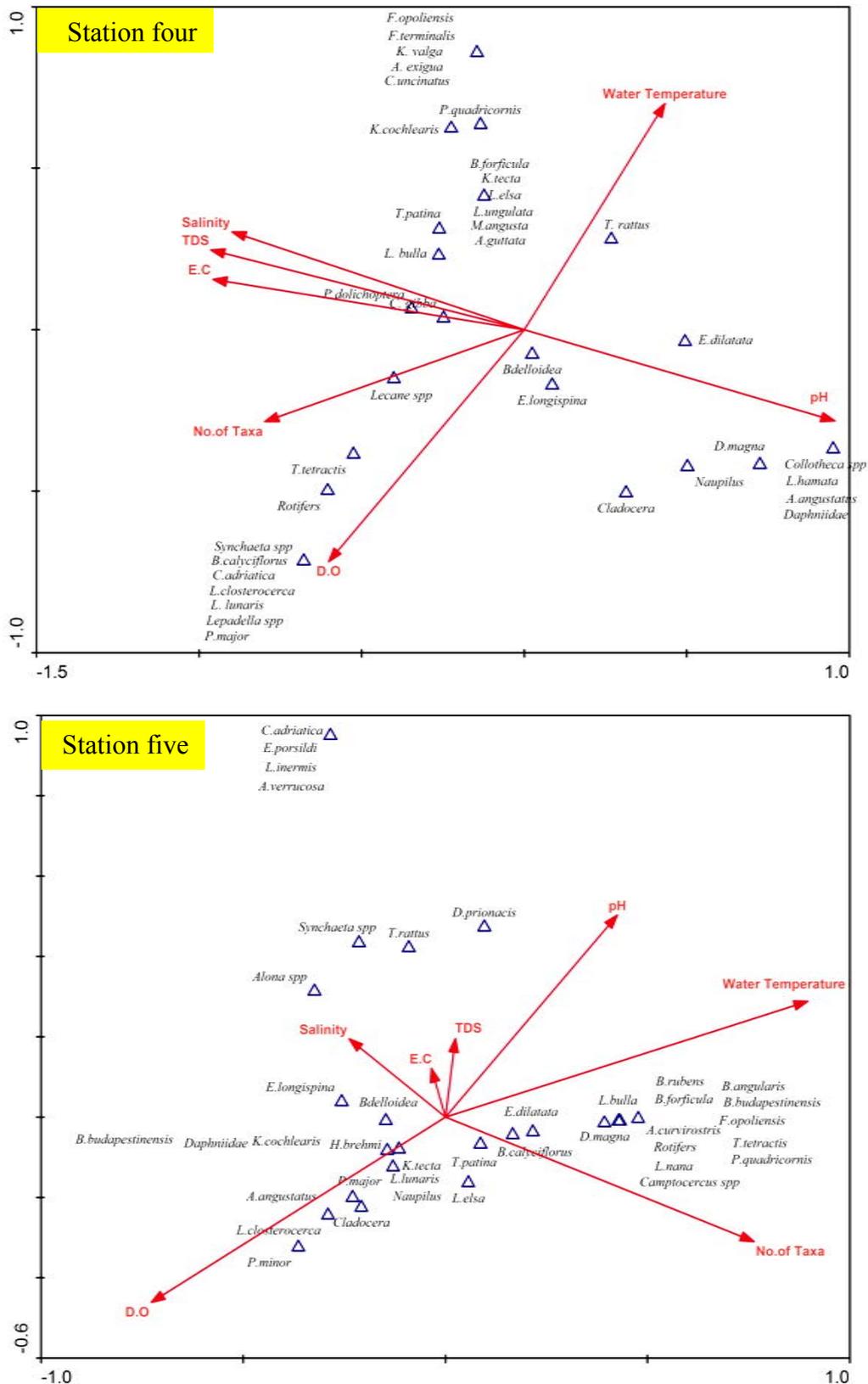


Figure (13): Canonical correspondence analysis (CCA) analysis of the correlation coefficients between rotifera and cladocera taxa and the environmental factors for stations four and five during the study period.

According to the statistical analysis, some species reported a positive and negative correlation with the environmental factors (Appendixes 1-5) and (Figures 11-13) as following :

In station one, *A. priodonta* showed a positive significant correlation ( $p \leq 0.05$ ) with dissolved oxygen ( $r=0.978$ ). As well as, *B. calyciflorus* observed a positive significant correlation with dissolved oxygen respectively ( $r=0.985$ ,  $r=0.987$ ) in station two and four. Likely, *C. adriatica* found a positive significant correlation with D.O respectively ( $p \leq 0.05$ ,  $r=0.978$  and  $r=0.987$ ) in station three and four. *L.closterocerca* exhibited also a positive significant correlation ( $r=0.981$ ) with DO at station four. *Lepadella spp* demonstrated a positive significant correlation ( $r=0.981$ ) with dissolved oxygen in station four. Also, *M.sericus* recorded a positive significant correlation ( $r=0.978$ ) with DO in station one. While in station two, Bdelloidea displays a negative significant correlation ( $p \leq 0.05$ ,  $r= - 0.968$ ) with DO.

*Cephalodella spp* reported a positive significant correlation ( $p \leq 0.05$ ) with pH ( $r=0.968$ ) while a negative significant correlation ( $r= - 0.968$ ) with salinity in station two. *Collotheca spp* indicated a negative significant correlation ( $r= - 0.974$ ) with TDS and a positive significant correlation ( $r=0.979$ ) with pH in station four. Also, *E.diltata* showed a negative significant correlation with EC ( $r= - 0.988$ ), salinity ( $r= - 0.984$ ) and TDS ( $r= -0.966$ ) within station one. As well as, *B. quadridentatus* showed a negative significant correlation ( $r= - 0.952$ ) with TDS in station five. *H.brehmi* display a negative significant correlation with TDS ( $r= - 0.952$ ) in station five.

*K.cochlearis* had a negative significant correlation ( $p \leq 0.05$ ) with DO ( $r = -0.958$ ) at station two. As well as, it recorded a negative significant correlation ( $r = -0.952$ ) with TDS in station five.

*K.tecta* showed a positive significant correlation ( $p \leq 0.05$ ) with DO ( $r = 0.985$ ) within station two. While in station five, it observed a negative significant correlation with TDS ( $r = -0.952$ ).

The data from station four showed a negative significant correlation ( $p \leq 0.05$ ) between *L.hamata* and TDS ( $r = -0.974$ ) while a positive significant correlation ( $r = 0.979$ ) with pH.

*L.lunaris* discovered a positive significant correlation ( $p \leq 0.05$ ) with DO ( $r = 0.981$ ), rotifera ( $r = 0.987$ ), *Lepadella* spp ( $r = 1.000$ ), *P.major* ( $r = 1.000$ ) and *Synchaeta* spp ( $r = 1.000$ ) at station four. But there was a negative significant correlation ( $r = -0.952$ ) with TDS within station five.

Nauplius recorded a negative significant correlation ( $p \leq 0.05$ ) ( $r = -0.971$ ) with DO in station one. As well as, with EC ( $r = -0.996$ ), TDS ( $r = -0.999$ ), salinity ( $r = -0.981$ ) but there is a positive significant correlation with pH ( $r = 0.961$ ) in station two. Within station three, there is negative significant correlation ( $r = -0.973$ ) with TDS. Finally, in station five there is negative significant correlation ( $r = -0.965$ ) with TDS.

*P.major* recorded a positive significant correlation ( $p \leq 0.05$ ) with DO ( $r = 0.987$ ) in station one, ( $r = 0.985$ ) in station two, ( $r = 0.987$ ) in station three and ( $r = 0.981$ ) within station four. While *P.minor* recorded a positive significant correlation with DO ( $r = 0.977$ ) and negative significant correlation with pH ( $r = -0.993$ ) in station five.

Different species recorded a positive significant correlation ( $p \leq 0.05$ ) with DO such as *S.longicaudum* ( $r = 0.985$ ) in station two, *S.lakowitziana*

( $r=0.978$ ) in station one. Also, *Synchaeta* spp recorded a positive significant correlation ( $r=0.978$ ) in station one, ( $r=0.978$ ) in station three and ( $r=0.981$ ) in station four.

*Trichocerca* spp showed a negative significant correlation ( $p \leq 0.05$ ) with salinity ( $r = - 0.968$ ) while a positive significant correlation ( $r = - 0.968$ ) with pH in station two.

*A. curvirostris* recorded a negative significant correlation ( $p \leq 0.05$ ) with air temperature ( $r = - 0.993$ ) and water temperature ( $r = - 0.996$ ) in station two.

*A. angustatus* recorded a negative significant correlation ( $p \leq 0.05$ ) with TDS ( $r = - 0.954$ ) and salinity ( $r = - 0.973$ ) in station one. As well as a negative significant correlation with TDS ( $r = - 0.974$ ) while a positive significant correlation with pH ( $r = 0.979$ ) in station four. *Camptocercus* spp observed a negative significant correlation with salinity ( $r = - 0.968$ ) and positive significant correlation ( $r = 0.968$ ) with pH in station two.

*D. magna* recorded a negative significant correlation ( $p \leq 0.05$ ) with salinity ( $r = - 0.968$ ) and positive significant correlation with pH ( $r = 0.968$ ) in station two. As well as, a positive significant correlation ( $r = 0.950$ ) with pH in station four.

Daphniidae showed a positive significant correlation ( $p \leq 0.05$ ) with DO ( $r = 0.978$ ) in station one. Within station two, it showed a negative significant correlation with salinity ( $r = - 0.968$ ) and positive significant correlation ( $r = 0.968$ ) with pH. As well as, it showed a negative significant correlation with TDS ( $r = -0.974$ ) and positive significant correlation ( $r = 0.979$ ) with pH in station four. While in station five, it showed a negative significant correlation with TDS ( $r = - 0.952$ ).

Finally, the statistical analysis indicated that most species recorded a positive significant correlation with both of DO and pH while it reported a negative significant correlation with EC, TDS and Salinity .

As well as, the statistical analysis showed that some species reported a positive or negative correlation with other species within study stations (Appendixes 1-5) and as following :

*A. ovalis* reported a positive significant correlation ( $p \leq 0.05$ ) with *Brachionus* spp, *C. adriatica*, *E. porsildi*, *Lepadella* spp., *M. ventralis* and *Bosminidae* ( $r=1.000$ ), as well as with *Lecane* spp ( $r=0.987$ ). *A. saltans* recorded a positive significant correlation with *L.bulla* ( $r=1.000$ ) and with *T. patina* ( $r=0.954$ ), while it recorded a negative significant correlation with *D.magna* ( $r= - 1.000$ ). *A. priodonta* showed a positive significant correlation with *B. angularis*, *M.sericus*, *P.major*, *S.lakowitziana.*, *Synchaeta* spp, and *Daphniidae* ( $r=1.000$ ). While it reported a negative significant correlation with *Nauplius* ( $r= - 0.951$ ).

*B. forficula* reported a positive significant correlation ( $p \leq 0.05$ ) with *B.rubens*, *L.closterocerca*, *L.lunaris*, *L.nana*, *L.quadridentata*, *M.angusta*, *T. reflexa*, *A.curvirostris*, *Chydoridae*, *cladocera*, *E.porsildi*, *Euchlanis* spp, *F.opoliensis*, *L.hamata*, *L.ludwigii*, *Lecane* spp, *L.tenuiseta*, *L.ungulata*, *P.quadricornis* and *Rotifera* spp, *B. calyciflorus*, *B. falcatus*, *D. prionacis*, *K.tropica*, *L.arcula*, *L.luna*, *P.minor*, *T.similis*, *K tecta*, *L.elsa*, *A.guttata*, *B.angularis*, *B.budapestinensis*, *T.tetractis*, *Camptocercus* spp ( $r=1.000$ ), *L.bulla* ( $r=0.988$ ) and *D.magna* ( $r=0.988$ ).

*B. calyciflorus* observed a positive significant correlation ( $p \leq 0.05$ ) with *K.quadrata*, *K.tecta*, *P.major*, *S.longicaudum*, *C.adriatica*, *L.closterocerca*, *Synchaeta* spp , *Alona* spp ( $r=1.000$ ) and *E.diltata*

( $r=0.975$ ). While it showed a negative significant correlation with *T. rattus* ( $r= - 0.969$ ).

*B. quadridentatus* showed a positive significant correlation ( $p \leq 0.05$ ) with *H.brehmi*, *K.cochlearis*, *K.tecta*, *L.lunaris* and Daphniidae ( $r=1.000$ ) and Nauplius ( $r=0.999$ ) and *P.major* ( $r=0.964$ ).

Bdelloidea displays a negative significant correlation ( $p \leq 0.05$ ) with *B. calyciflorus*, *K.quadrata*, *K.tecta*, *P.major* and *S.longicaudum* ( $r= - 0.992$ ). Additionally, a positive significant correlation with *B.forficula*, *B.falcatus*, *D.prionacis*, *K.tropica*, *L. arcula*, *L.luna*, *L.lunaris*, *L.quadridentata*, *L.ungulata*, *P.quadricornis*, *P.minor* and *T. similis* ( $r=0.990$ ) is seen. While it showed a negative significant correlation with *F.opoliensis*, *F.terminalis*, *K.valga*, *A.exigua* and *C.uncinatus* ( $r= - 0.984$ ) and with *K. cochlearis* ( $r= - 0.953$ ). Finally, it is shown that a negative significant correlation with *D.prionacis* ( $r= -0.981$ ) and positive significant correlation with *L. closterocerca* ( $r=0.968$ ) as well as with *A. angustatus* ( $r=0.991$ ).

*Cephalodella* spp reported a positive significant correlation ( $p \leq 0.05$ ) with *Collotheca* spp, *Colurella* spp, *H.brehmi*, *L.elsa*, *L.hamata*, *P.quadricornis*, *P. dolichoptera*, *Rotaria* spp, Rotifera spp, *S. longicaudum*, *T. rattus* and *T.pocillum* ( $r=1.000$ ) as well as *M.angusta* ( $r=0.995$ ). Additionally, it revealed a positive significant correlation *Trichocerca* spp, *Camptocercus* spp, *D.magna* and Daphniidae ( $r=1.000$ ). *C.gibba* exhibit a negative significant correlation only with *K.tropica* ( $r= -1.000$ ).

*Collotheca* spp had a positive significant correlation ( $p \leq 0.05$ ) with *L.hamata*, *Lecane* spp, *A. angustatus*, Daphniidae ( $r=1.000$ ) and *F. opoliensis* ( $r=0.987$ ). *C. adriatica* found a positive significant correlation

with *K.tecta*, *L.closterocerca*, *L.lunaris*, *E. porsildi*, *L.inermis*, *A.verrucosa* ( $r=1.000$ ) and rotifera spp ( $p \leq 0.05$ ,  $r=0.987$ ). *D.prionacis* exhibits a negative significant correlation only with *A. angustatus* ( $r= - 0.996$ ). With respect to *Lepadella* spp, it demonstrated a positive significant correlation ( $p \leq 0.05$ ) with *T.patina*, *A.curvirostris*, *E porsildi*, *L.inermis* and *Camptocercus* spp ( $r=1.000$ ).

*E.diltata* showed a positive significant correlation ( $p \leq 0.05$ ) with *A. angustatus* ( $r=0.968$ ) and with *L.bulla*, *Macrochaetus* spp, Rotifera spp, *T.rattus* and *Camptocercus* spp ( $r=0.998$ ), as well as with cladocera ( $r=0.994$ ) and Naupilus ( $r=0.953$ ). In contrast, it reported a negative significant correlation with *Alona* spp ( $r= - 0.975$ ).

*F.opoliensis* discovered a positive significant correlation ( $p \leq 0.05$ ) with *F.terminalis*, *K.valga*, *A.exigua*, *C.uncinatus*, *L.nana*, *P.quadricornis*, Rotifers, *T.tetractis*, *Camptocercus* spp and *L.bulla* ( $r=1.000$ ), as well as with *K.cochlearis* ( $r=0.985$ ), *A. curvirostris* ( $r=0.997$ ) and *D.magna* ( $r=0.988$ ).

*H.brehmi* display a positive significant correlation ( $p \leq 0.05$ ) with *K.cochlearis*, *K.tecta*, *L.lunaris*, Daphniidae ( $r=1.000$ ) and Nauplius ( $r=0.999$ ) as well as *P.major* ( $r= 0.964$ ).

*K.cochlearis* had a positive significant correlation ( $p \leq 0.05$ ) with *K.valga*, *A.exigua* and *C.uncinatus* ( $r= 0.985$ ). *K.tecta* showed a positive significant correlation with *L.patella*, *Synchaeta* spp, *L.lunaris*, Daphniidae ( $r= 1.000$ ) and Naupilus ( $r=0.999$ ). *K.valga* revealed a positive significant correlation with *A.exigua* and *C.uncinatus* ( $r=1.000$ ).

*L.bulla* reported a positive significant correlation ( $p \leq 0.05$ ) with *Macrochaetus* spp, Rotifera spp, *T.rattus*, *Camptocercus* spp and *A.curvirostris* ( $r=1.000$ ), as well as with cladocera ( $r=0.998$ ), *L.nana*,

*P.quadricornis*, Rotifera spp, *T.tetractis*, *Camptocercus* spp and *D.magna* ( $r=0.997$ ).

*L.closterocerca* exhibited a positive significant correlation ( $p \leq 0.05$ ) with *L.thienemanni*, *Lepadella* spp, *P.major* and *Synchaeta* spp ( $r=1.000$ ) but there was a negative significant correlation with *T.rattus* ( $r=- 0.969$ ).

*L.elsa* showed a positive significant correlation ( $p \leq 0.05$ ) with *L.hamata*, *P.quadricornis*, *P.dolichoptera*, *Rotaria* spp, Rotifera spp, *S.longicaudum*, *T. rattus* and *T. pocillum*, *L.ungulata*, *M.angusta*, *A.guttata*, *Synchaeta* spp and *A.angustatus* ( $r=0.955$ ).

*L.hamata* is shown a positive significant correlation ( $p \leq 0.05$ ) with *L.ludwigii*, *L.lunaris*, *L.nana*, *L. quadridentata*, *Lecane* spp, *L.tenuiseta*, *A.angustatus* and Daphniidae ( $r=1.000$ ) and *D.magna* ( $r=0.987$ ). As well as, *L.inermis* reported a positive significant correlation with *A.curvirostris* ( $r=1.000$ ).

*L.lunaris* recorded a positive significant correlation ( $p \leq 0.05$ ) with *Lepadella* spp, *P.major*, *Synchaeta* spp, Daphniidae ( $r=1.000$ ) and Nauplius ( $r=0.999$ ). But there is a negative significant correlation with *T.rattus* ( $r=- 0.969$ ). *L.nana* showed reported a positive significant correlation with *T.tetractis* and *Camptocercus* spp ( $r=1.000$ ) and with *A.curvirostris* ( $r=0.997$ ) as well as with *D.magna* ( $r=0.988$ ). *Lecane* spp reported a positive significant correlation with *Lepadella* spp, *M.ventralis* and Bosminidae ( $r=0.987$ ).

*Lepadella* spp recorded a positive significant correlation ( $p \leq 0.05$ ) with *M.ventralis*, *P.major*, *Synchaeta* spp and Bosminidae ( $r=1.000$ ). However, it revealed a negative significant correlation with *T.rattus* ( $r= - 0.969$ ).

*M.sericus* recorded a positive significant correlation ( $p \leq 0.05$ ) with *P.major*, *S.lakowitziana*, *Synchaeta* spp and Daphniidae ( $r=1.000$ ). But it demonstrated a negative significant correlation with Nauplius ( $r= -0.951$ ). *Macrochaetus* spp recorded a positive significant correlation with Rotifera spp, *T.rattus* and *Camptocercus* spp ( $r=1.000$ ) and cladocera ( $r=0.998$ ).

*P.quadricornis* recorded a positive significant correlation ( $p \leq 0.05$ ) with Rotifera spp, *T.tetractis*, *Camptocercus* spp ( $r=1.000$ ) and *A.curvirostris* ( $r=0.997$ ) as well as *D.magna* ( $r=0.988$ ).

*P.dolichoptera* exhibits a positive significant correlation ( $p \leq 0.05$ ) with *Rotaria* spp, Rotifers, *S.longicaudum*, *T.rattus* and *T.pocillum* ( $r=1.000$ ) as well as *A.angustatus* ( $r=0.995$ ).

*P.major* recorded a positive significant correlation *S.lakowitziana*, *Synchaeta* spp, Daphniidae, *S.longicaudum* and *Synchaeta* spp ( $r=1.000$ ). While a negative significant correlation with *T.rattus* ( $r= -0.969$ ). *P.minor* recorded a positive significant correlation with *T.similis* ( $r= 1.000$ ).

*T.rattus* observed a positive significant correlation ( $p \leq 0.05$ ) with *T.pocillum* ( $r=1.000$ ), *A.angustatus* ( $r=0.995$ ), *Camptocercus* spp and cladocera ( $r=0.998$ ). Whereas it showed a negative significant correlation with *T.tetractis* ( $r=- 0.997$ ).

*Trichocerca* spp showed a positive significant correlation ( $p \leq 0.05$ ) with *Camptocercus* spp, *D.magna* and Daphniidae ( $r=1.000$ ).

*T.patina* exhibits a negative significant correlation ( $p \leq 0.05$ ) with *D.magna* ( $r= -0.954$ ), *E.longispina* ( $r= -0.989$ ) and cladocera ( $r= -0.964$ ). While it recorded a positive significant correlation with *A.curvirostris* ( $r=$

1.000). *T.reflexa* recorded a positive significant correlation with *A.curvirostris*, Chydoridae and cladocera spp (r=1.000).

*T.pocillum* reported a positive significant correlation ( $p \leq 0.05$ ) with *A.angustatus* (r= 0.995). *T.tetractis* demonstrated a positive significant correlation with *Camptocercus* spp (r=1.000), *A.curvirostris* (r=0.997) and with *D.magna* (r=0.988).

**Table (10): Canonical correspondence analysis (CCA) analysis of the correlation coefficients between rotifera and cladocera species and the environmental factors at the stations during the study period.**

Station one					
Axes	1	2	3	4	Total inertia
Eigenvalues	0.550	0.443	0.296	0.000	1.288
Species-environment correlations	1	1	1	1	
Cumulative percentage variance					
of species data	42.7	77.1	100	0.000	
of species-environment relation	42.7	77.1	100	0.000	
Sum of all eigenvalues					1.288
Sum of all canonical eigenvalues					1.288
Station two					
Axes	1	2	3	4	Total inertia
Eigenvalues	0.552	0.388	0.234	0.000	1.175
Species-environment correlations	1	1	1	1	
Cumulative percentage variance					
of species data	47.0	80.0	100	0.000	
of species-environment relation	47.0	80.0	100	0.000	
Sum of all eigenvalues					1.175
Sum of all canonical eigenvalues					1.175
Station three					
Axes	1	2	3	4	Total inertia
Eigenvalues	0.642	0.425	0.406	0.000	1.473
Species-environment correlations	1	1	1	1	
Cumulative percentage variance					
of species data	43.6	72.4	100	0.000	
of species-environment relation	43.6	72.4	100	0.000	
Sum of all eigenvalues					1.473
Sum of all canonical eigenvalues					1.473
Station four					
Axes	1	2	3	4	Total inertia
Eigenvalues	0.433	0.365	0.237	0.000	1.035
Species-environment correlations	1	1	1	1	
Cumulative percentage variance					
of species data	41.9	77.1	100	0.000	
of species-environment relation	41.9	77.1	100	0.000	
Sum of all eigenvalues					1.035
Sum of all canonical eigenvalues					1.035
Station five					
Axes	1	2	3	4	Total inertia
Eigenvalues	0.529	0.320	0.235	0.000	1.084
Species-environment correlations	1	1	1	1	
Cumulative percentage variance					
of species data	48.8	78.3	100	0.000	
of species-environment relation	48.8	78.3	100	0.000	
Sum of all eigenvalues					1.084
Sum of all canonical eigenvalues					1.084

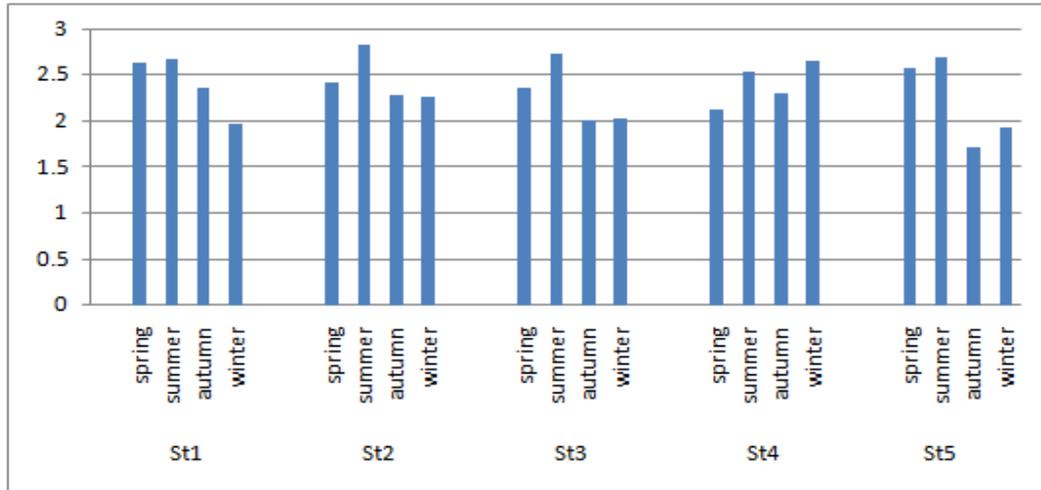
### 3 – 4: Biodiversity Indices

#### 3 -4-1 : Shannon-Wiener diversity index

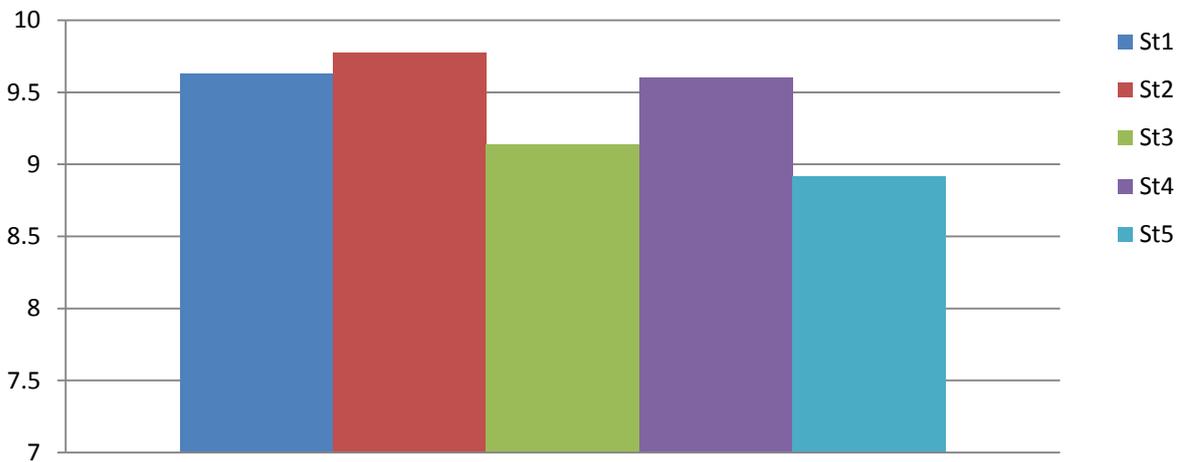
According to (Figure 14) which displays the five stations' quarterly Shannon-Wiener index values during the study period. The second station recorded the highest value (2.83) in the summer and the fifth station recorded the lowest value (1.721) in the fall. While according to (Figure 15) which shows Shannon-Wiener annual values for the five stations. The lowest value is recorded at the fifth station (8.913) and the highest value is recorded at the second station (9.777). According to our results, Station two was the most divers during summer and characterized by moderate status of the diversity.

**Table (11): Seasonal changes in biodiversity indices for the five stations during the study period from Spring 2021 to Winter 2022**

Indices	St1				St2				St3				St4				St5			
	Spring	Summer	Autumn	Winter																
Simpson Index	0.09	0.08	0.11	0.18	0.10	0.07	0.11	0.11	0.10	0.08	0.14	0.16	0.135	0.09	0.12	0.08	0.09	0.08	0.250	0.18
Shanon Index	2.63	2.66	2.36	1.96	2.40	2.83	2.28	2.25	2.36	2.73	2.00	2.02	2.12	2.52	2.30	2.64	2.58	2.68	1.72	1.92
Menhinick Index	0.69	0.72	0.69	0.56	0.53	0.77	0.56	0.55	0.39	0.73	0.39	0.57	0.34	0.48	0.49	0.56	0.62	0.75	0.61	0.39
Evenness Index	0.85	0.86	0.83	0.79	0.84	0.88	0.89	0.90	0.89	0.87	0.91	0.81	0.85	0.87	0.85	0.88	0.84	0.84	0.71	0.83
Berger-Parker Index	0.17	0.13	0.18	0.32	0.15	0.13	0.18	0.17	0.14	0.18	0.21	0.27	0.17	0.14	0.22	0.17	0.16	0.16	0.4	0.33
Margalef Index	3.033	3.081	2.503	1.803	2.308	3.351	1.918	1.787	1.822	3.189	1.278	1.809	1.547	2.35	2.052	2.66	2.85	3.323	1.734	1.394



**Figure (14):**Seasonal changes of Shannon-Wiener Index for the five stations during the study period from Spring 2021 to Winter 2022

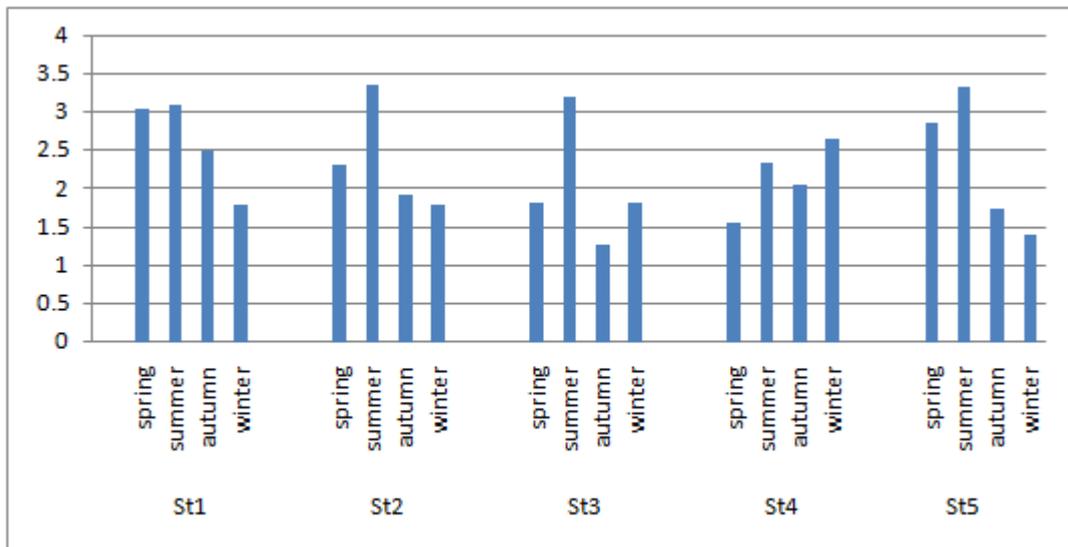


**Figure (15):** Annual changes in Shannon-Wiener for the five stations during the study period from Spring 2021 to Winter 2022

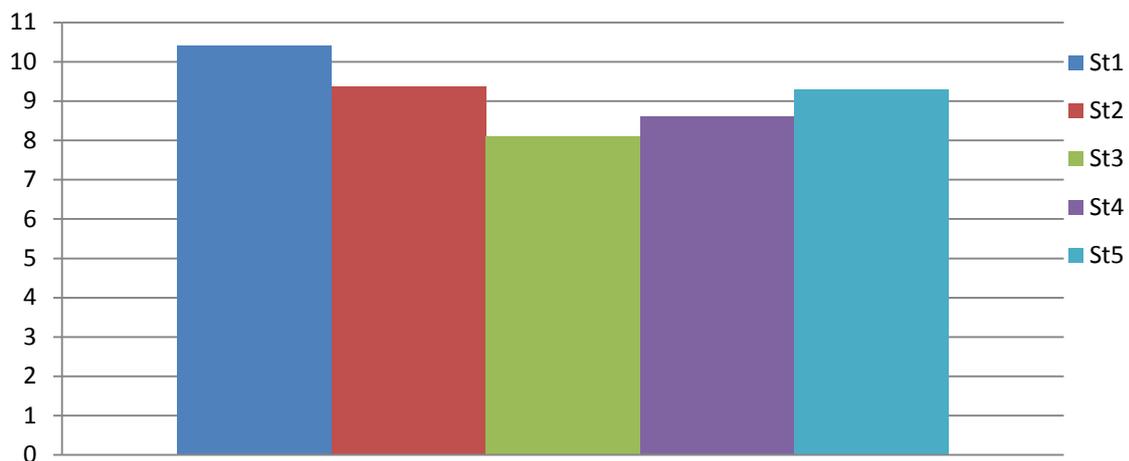
### 3 -4-2 : Margalef’s richness index

The seasonally Margalef index values for the five stations over the period of the study are shown in (Figure 16). The third station recorded the lowest value during the autumn (1.278) and the second station recorded the greatest value during the summer (3.351). According to (Figure 17), which displays Margalef yearly values for the five stations, the third station recorded the lowest value (8.098) and the first site

recorded the greatest value (10.42). According to current results, Station two was the most richness during summer and characterized as semi-disturbed of the diversity.



**Figure (16): Seasonal changes of Margalef Index for the five stations during the study period from Spring 2021 to Winter 2022**



**Figure (17): Annual changes of Margalef Index for the five stations during the study period from Spring 2021 to Winter 2022**

### 3 -4-3 : Index of Dominance (Simpson Index)

The quarterly Simpson index values for the five stations during the study period are shown in (Figure 18). In the summer, the second station recorded the lowest value (0.071) but in the autumn, the fifth station recorded the greatest value (0.250). While (Figure 19) displays Simpson's annual values for the five stations with the lowest value being recorded at the fourth station (0.432) and the greatest value recorded at the fifth station (0.605). According to present results, station five was the most richness during autumn and characterized by low degree of diversity (Heterogeneity).

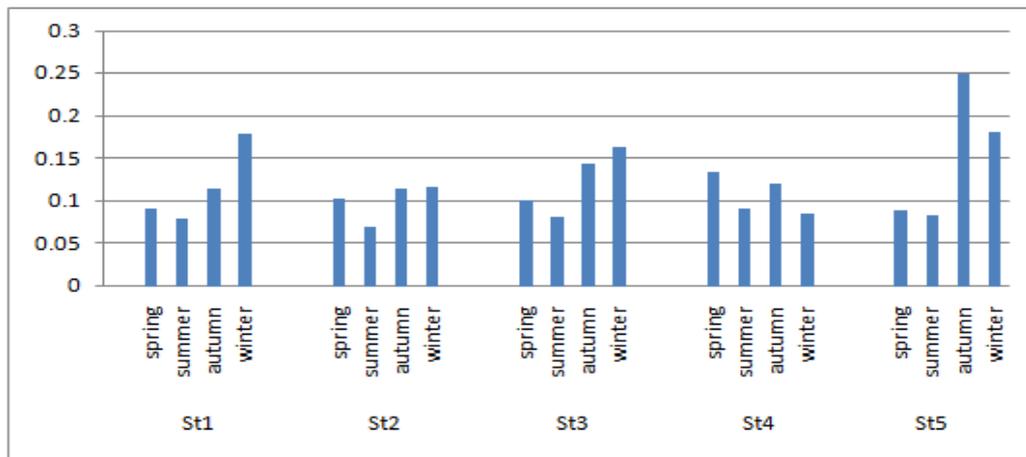


Figure (18): Seasonal changes of Simpson Index for the five stations during the study period from Spring 2021 to Winter 2022

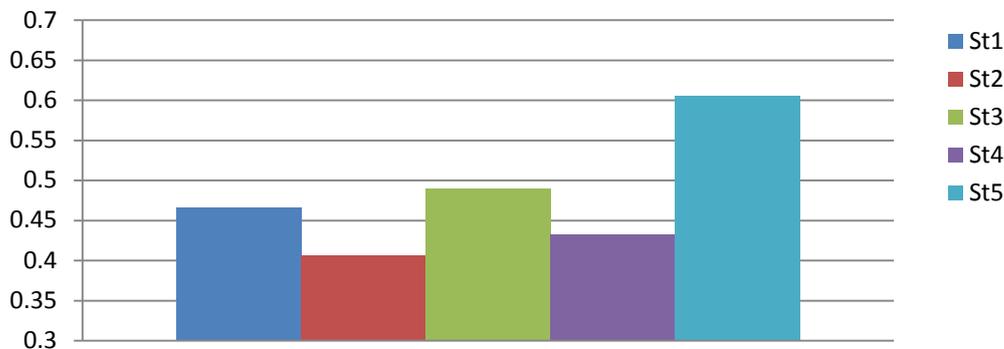
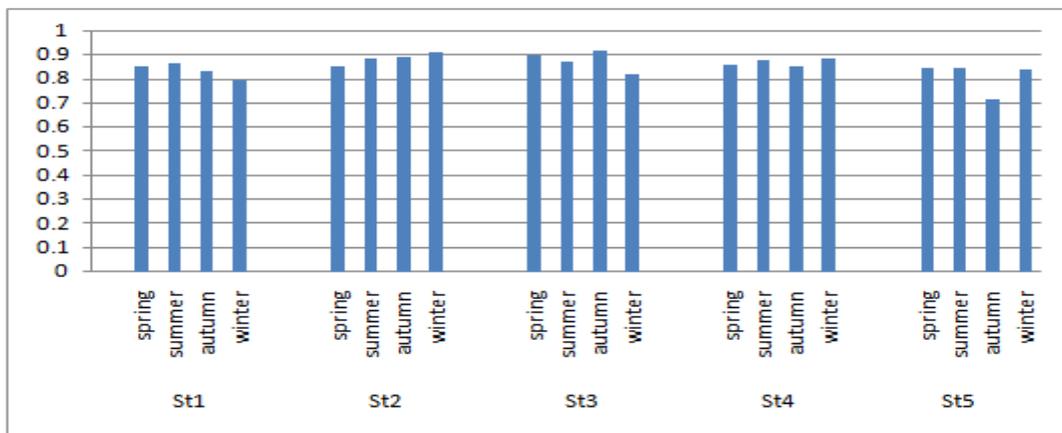


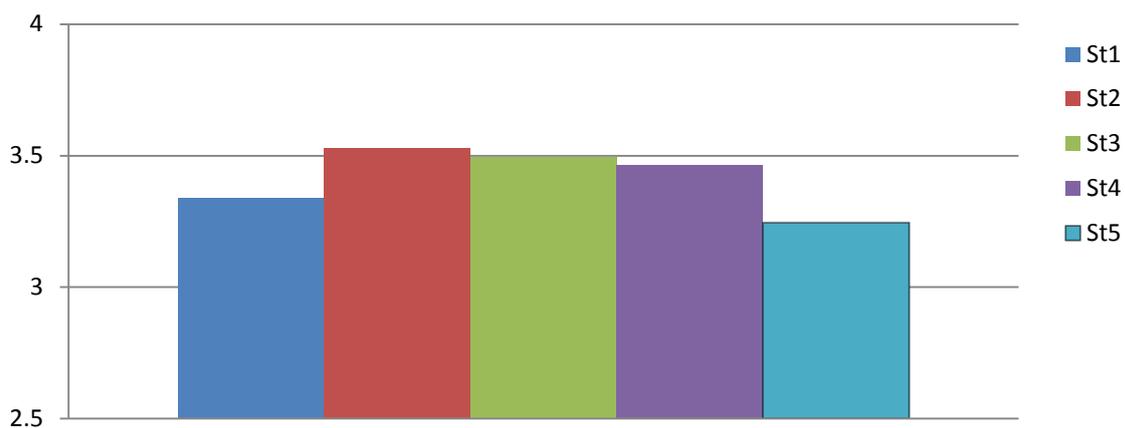
Figure (19): Annual changes in Simpson Index for the five stations during the period from spring 2021 to winter 2022

**3 -4-4 : Evenness index**

Based on (Figure 20), the Evenness seasonal values for the five stations during the study indicated that third station reported the highest value (0.913) and the fifth station recorded the lowest value (0.717) both in the autumn. While (Figure 21) shows Evenness yearly values for the five stations and reported that lowest value is recorded at the fifth station (3.245) and the highest value is recorded at the second station (3.527). According to present findings, Station three was the most evenness during autumn and characterized as balanced station.



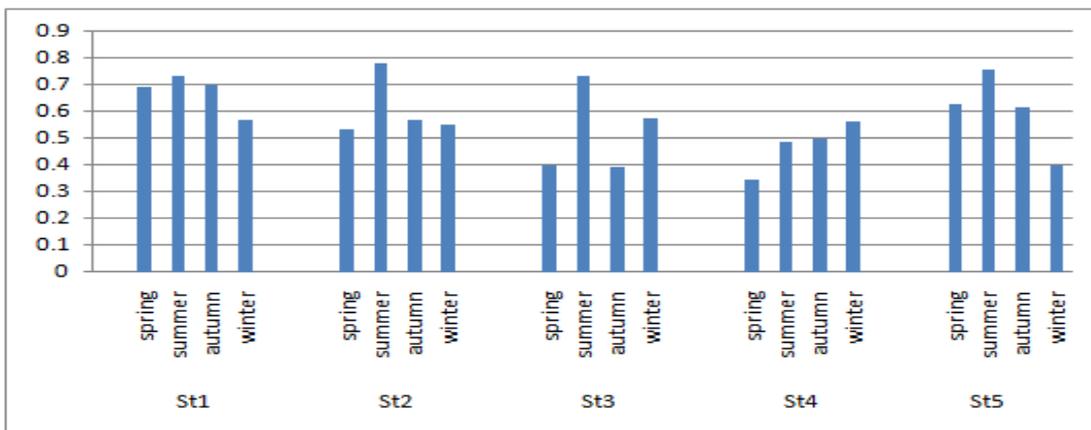
**Figure (20): Seasonal changes of Evenness Index for the five stations during the study period from Spring 2021 to Winter 2022**



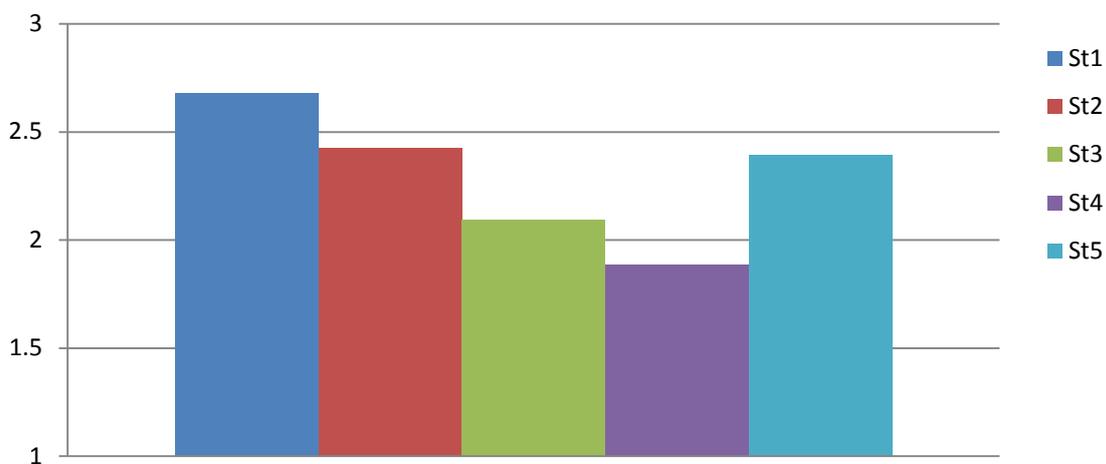
**Figure (21): Annual changes of Evenness Index for the five stations during the study period from Spring 2021 to Winter 2022**

### 3 -4-5 : Menhinick Index

During the study period, the Menhinick seasonal values for the five stations are shown in (Figure 22). The lowest value (0.343) was recorded in the spring at the fourth station and the highest value (0.776) is reported in the summer at the second station. According to (Figure 23), which displays Menhinick yearly values for the five stations, the lowest value is recorded at the fourth station (1.883) while the highest value is recorded at the first station (2.68). According to current results, Station two was the most richness during summer.



**Figure (22): Seasonal changes of Menhinick Index for the five stations during the study period from Spring 2021 to Winter 2022**



**Figure (23): Annual changes of Menhinick Index for the five stations during the study period from Spring 2021 to Winter 2022**

3 – 4-6: Berger-Parker Index

Depending on (Figure 24), the seasonally Berger-Parker index values for the five locations over the period of the study demonstrated that the first station recorded the lowest value (0.13) during the summer and the highest value (0.4) during the fall at the fifth site. While according to (Figure 25) which shows Berger-Parker annual values for the five stations, it is clear that the lowest value (0.649) is obtained at the second station and the highest value (1.056) is recorded at the fifth site.

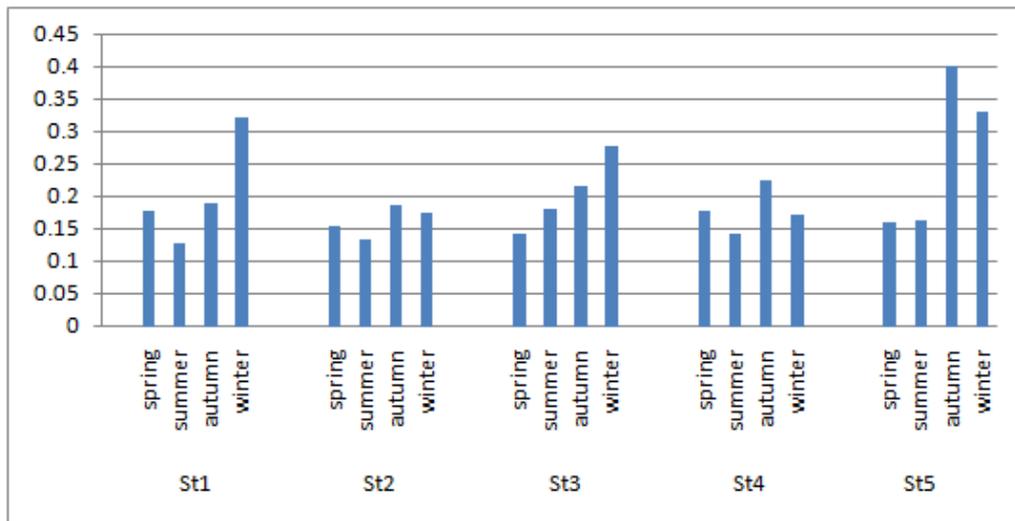


Figure (24): Seasonal changes of Berger-Parker Index for the five stations during the study period from Spring 2021 to Winter 2022

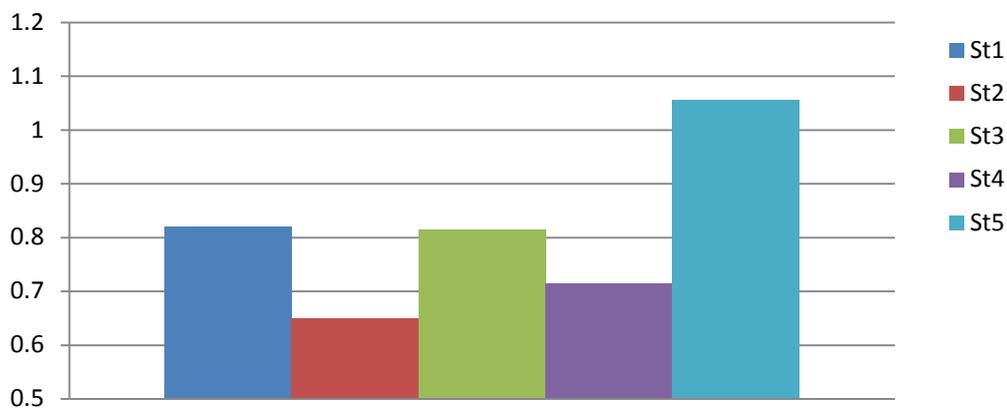


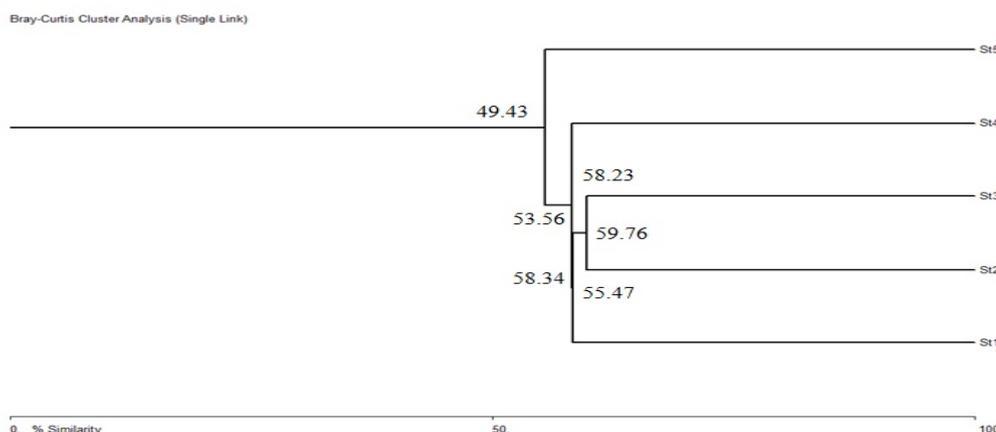
Figure (25): Annual changes of Berger-Parker Index for the five stations during the study period from Spring 2021 to Winter 2022

### 3 – 4-7: Jaccard's Index (%)

The Jaccard values for the five stations are shown in (Figure 26) (table 12). The lowest similarity value (49 %) was observed between the first and fifth stations, and the highest similarity value (59 %) is between the second and third stations and these coefficients indicated that stations 2 and 3 were very much similar to each other.

**Table (12): Jaccard's Index values for similarity of zooplankton among five stations from Spring 2021 to Winter 2022**

Similarity matrix	St1	St2	St3	St4	St5
St1	*	56.38	58.34	53.31	49.43
St2	*	*	59.76	57.203	55.47
St3	*	*	*	58.23	50.52
St4	*	*	*	*	53.56
St5	*	*	*	*	*



**Figure (26): Cluster distribution of Jaccard's Index for similarity of zooplankton among five stations from Spring 2021 to Winter 2022**

### 3 – 4-8: Alpha , Beta and Gamma Diversity

The values of alpha, beta and gamma diversity for the five stations over the period of the investigation are displayed in (table 13). According to the findings, the second station had high alpha diversity value (103)

while the third station had low alpha diversity value (84). The findings also revealed that the lowest value of beta diversity is (59) between the second and fifth stations and the maximum value is (83) between the second and third stations. As well as, during the study period, gamma diversity is (178).

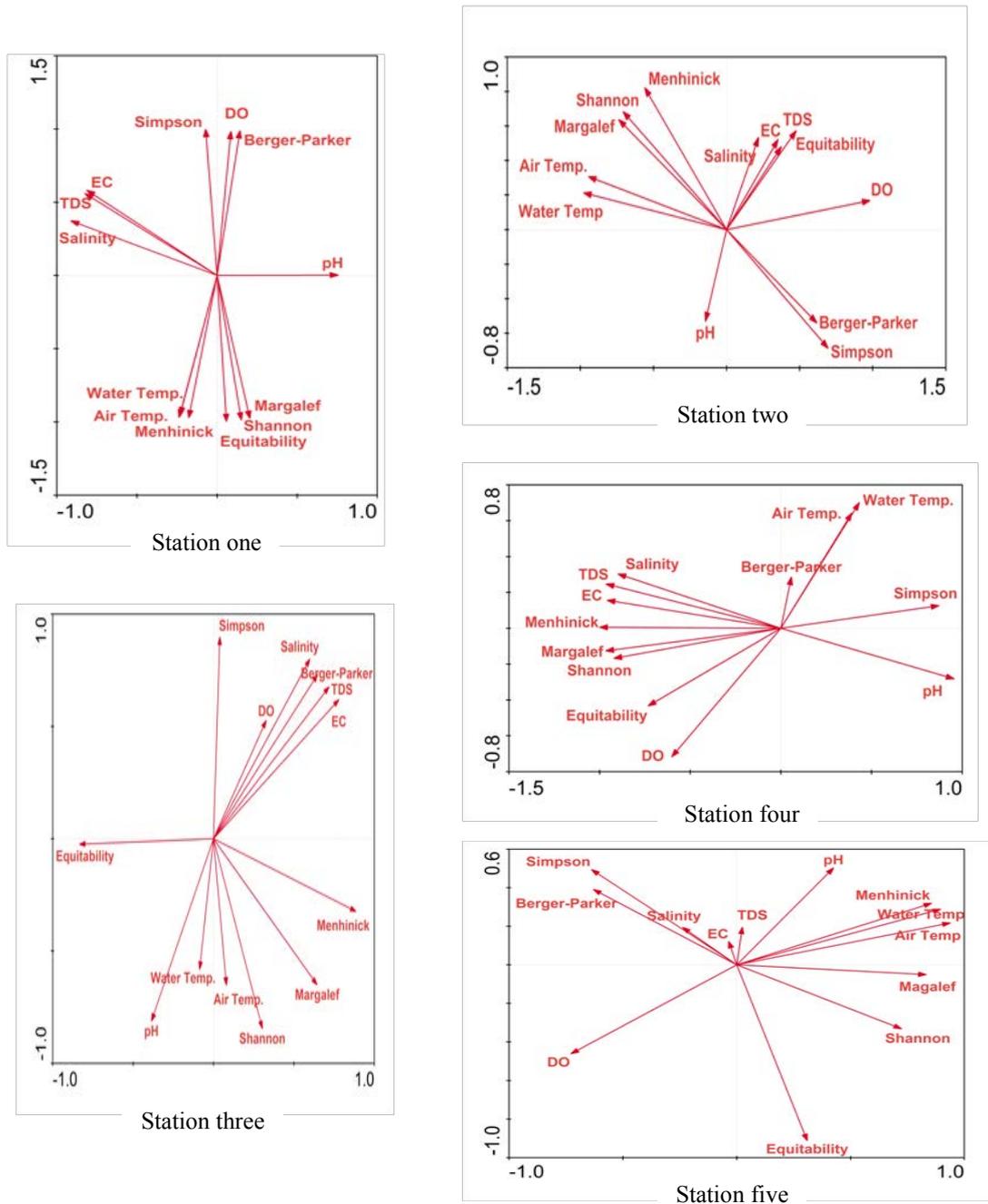
**Table (13): Alpha, Beta and Gamma Diversity values according to morphological and genetic identification among five stations from Spring 2021 to Winter 2022**

Type of diversity	St 1	St 2	St 3	St 4	St 5
Alpha Diversity	89	103	84	85	86
Beta Diversity		St1 & St 2 = 76	St1 & St 3 = 77	St1 & St 4 = 60	St1 & St 5 = 71
			St2 & St 3 = 83	St2 & St 4 = 60	St2 & St 5 = 59
				St3 & St 4 = 75	St3 & St 5 = 72
					St4 & St 5 = 65
Gamma Diversity	178				

### 3–5: The correlation between the environmental factors and biodiversity indices

The statistical analysis demonstrated that the indices reported a positive or negative correlation with each other as well as with environmental factors within study stations (Appendixes 6-10) and (Figure 27) as following :

During station one, Index of Dominance (Simpson Index) had a positive significant correlation ( $p \leq 0.05$ ) with DO ( $r=0.982$ ) but there was a negative significant correlation with water temperature ( $r= - 0.967$ ) and total number of taxa ( $r= -0.969$ ). In station two, it recorded a negative significant correlation with Margalef's richness index ( $r= - 0.997$ ) and



**Figure (27): Canonical correspondence analysis (CCA) analysis of the correlation coefficients between biodiversity indices and the environmental factors at the stations during the study period.**

total number of taxa ( $r = -0.957$ ). Likely, it showed a negative significant correlation with total number of taxa ( $r = -0.971$ ) within station three. As well as, it reported a negative significant correlation with total number of taxa ( $r = -0.980$ ) in station four. Also, it showed a negative significant

correlation with total number of taxa ( $r = -0.975$ ) and Shannon-Wiener index ( $r = -0.982$ ) while it display a positive significant correlation with Berger-Parker Index ( $r = 0.991$ ) in station five.

Shannon-Wiener index had a positive significant correlation ( $p \leq 0.05$ ) with the total number of taxa ( $r = 0.991$ ) while it recorded a negative significant correlation with Simpson Index ( $r = -0.991$ ) during station one. In station two, it showed a positive significant correlation with the total number of taxa ( $r = 0.953$ ) and with Margalef's richness index ( $r = -0.998$ ) but it exhibited a negative significant correlation with Simpson Index ( $r = -0.999$ ). Also, it showed a positive significant correlation with number of taxa ( $r = 0.995$ ) in station three. During station four, it also recorded a positive significant correlation with number of taxa ( $r = 0.964$ ) and a negative significant correlation with Simpson Index ( $r = -0.991$ ). Likely, it has positive significant correlation with number of taxa ( $r = 0.999$ ) and a negative significant correlation with Berger-Parker Index ( $r = -0.994$ ) in station five.

Menhinick Index demonstrated a positive significant correlation ( $p \leq 0.05$ ) with water temperature ( $r = 0.976$ ) but it showed a negative significant correlation with Simpson Index ( $r = -0.969$ ) and with DO ( $r = -0.997$ ) in station one. Within station four, it display a a negative significant correlation with pH ( $r = -0.954$ ) while it showed a positive significant correlation with EC ( $r = 0.961$ ) and TDS ( $r = 0.964$ ). Likely, it reported a positive significant correlation with water temperature ( $r = 0.997$ ) and air temperature ( $r = 0.979$ ) but it showed a negative significant correlation ( $r = -0.969$ ) with DO in station five.

Evenness index display a positive significant correlation ( $p \leq 0.05$ ) with water temperature ( $r = 0.970$ ), air temperature ( $r = 0.963$ ), total number of

taxa ( $r=0.982$ ), Menhinick Index ( $r=0.958$ ) and Shannon-Wiener index ( $r=0.994$ ) while it showed a negative significant correlation with Simpson Index ( $r= - 0.998$ ) and with DO ( $r= -0.971$ ) in station one. Within station two, it showed a negative significant correlation with pH ( $r=-0.895$ ) and EC ( $r=-0.973$ ). As well as, it showed a positive significant correlation with pH ( $r=0.978$ ) and total number of taxa ( $r=0.978$ ) in station four.

Berger-Parker Index showed a negative significant correlation ( $p \leq 0.05$ ) with water temperature ( $r= -0.991$ ), air temperature ( $r= -0.968$ ), Shannon index ( $r= - 0.954$ ), Menhinick Index ( $r= -0.994$ ) and Evenness index ( $r= - 0.980$ ) but it showed a positive significant correlation with DO ( $r=0.994$ ) and with Simpson index ( $r=0.984$ ) in station one. However, it recorded a negative significant correlation with total number of taxa ( $r= - 0.969$ ) within station two. Among station three, it display a negative significant correlation with pH ( $r=-0.963$ ) but positive significant correlation with salinity ( $r= 0.975$ ). Also, it showed a negative significant correlation with total number of taxa ( $r= -0.989$ ) in station five.

Margalef's richness index recorded a positive significant correlation ( $p \leq 0.05$ ) with total number of taxa ( $r=0.994$ ), Shannon index ( $r=1.000$ ) and Evenness index ( $r=0.992$ ) while it showed a negative significant correlation with Simpson index ( $r= -0.988$ ) in station one. It also showed a negative significant correlation with Simpson index ( $r= -0.997$ ) but there was a positive significant correlation with total number of taxa ( $r= 0.970$ ) in station two. As well as, it showed a negative significant correlation with Simpson index ( $r= -0.964$ ) and a positive significant correlation with Shannon index ( $r= 0.989$ ) in station four.

Regarding to the total density, it showed a positive significant correlation ( $p \leq 0.05$ ) with total number of taxa ( $r = 0.982$ ), Shannon index ( $r = 0.956$ ) and Margalef's index ( $r = 0.960$ ) in station one. During station three, it demonstrated a negative significant correlation with TDS ( $r = -0.971$ ) and salinity ( $r = -0.990$ ). Also, it showed negative significant correlation with Berger-Parker Index ( $r = -0.994$ ) in station four. Whereas in station five, it recorded a positive significant correlation with total number of taxa ( $r = 0.970$ ) and Shannon index ( $r = 0.963$ ) but it showed a negative significant correlation with Simpson index ( $r = -0.990$ ) and Berger-Parker Index ( $r = -0.986$ ).

Finally, the statistical analysis indicated that the species richness indices (Menhinick index and Margalef index) recorded a positive significant correlation with total number of taxa, total density, air and water temperature, EC, TDS and Evenness indices (Shannon index and Evenness index), but it recorded a negative significant correlation with DO, pH and Dominance indices (Simpson index and Berger-Parker index). As well as, Evenness indices (Shannon index and Evenness index) demonstrated a positive significant correlation with total number of taxa, total density, air and water temperature and pH but it showed a negative significant correlation with DO and Dominance indices (Simpson index and Berger-Parker Index). As for Dominance indices (Simpson index and Berger-Parker Index), it showed a positive significant correlation with DO and salinity but it display a negative significant correlation with total number of taxa, total density, air and water temperature and pH.

### 3 – 6 : Morphological Identification of the Zooplankton

In the current study, 69 taxa of rotifera comprising (54 species and 12 genus) following to (17) families and 14 taxa of Cladocera comprising (8 species and 2 genus) following to (4) families are identified. In the current study, three species of rotifera (*Ascomorpha ovalis*, *Horaella brehmi* and *Lecane arcula*) and five species of Cladocera (*Acantholeberis curvirostris*, *Acroperus angustatus*, *Anthalona verrucosa*, *Alonella exigua* and *Eubosmina longispina*) are recorded for the first time in Iraq, as well as four species of rotifera (*Lecane quadridentata*, *Lecane tenuiseta*, *Lecane unguolata* and *Scaridium longicaudum*) are recorded in the Hilla River for the first time. It is difficult to identify class Bdelloidea because the sample is kept in formalin, therefore only the class Monogononta was recognized.

#### Rotifera :

Kingdom: Animalia

--Phylum : Rotifera

----Super class: Eurotatoria

-----Class Bdelloidea

-----Order : Philodinida

-----Family : Philodinidae

-----Genus: *Macrotrachela*

*Macrotrachela angusta* (Bryce 1894)

-----Genus: *Rotaria*

-----Class: Monogononta

-----Order : Collothecaceae

-----Family : Collothecidae

-----Genus: *Collotheca*

*Collotheca* sp.

----- Order : Flosculariaceae

-----Family : Testudinellidae

-----Genus: *Testudinella*

*T. patina* (Hermann, 1783)

*T. reflexa* (Gosse, 1887)

-----Family: Trochosphaeridae

-----Genus: *Filinia*

*F. opoliensis* ( Zacharias, 1898 )

*F. terminalis* ( Plate ,1886 )

-----Genus: *Horaella*

*H. brehmi* Donner , 1949

-----Order : Ploima

-----Family : Asplanchnidae

-----Genus: *Asplanchna*

*A. priodonta* Gosse, 1850

-----Family : Scaridiidae

-----Genus: *Scaridium*

*S. longicauda* (Müller,1786)

-----Family: Brachionidae

-----Genus: *Brachionus*

*B. angularis* Gosse ,1851

*B. budapestinensis* Daday, 1885

*B. calyciflorus* Pallas, 1766

*B. falcatus* Zacharias, 1898

*B. forficula* Wierzejski, 1891

*B. quadridentatus* Hermann, 1783

*B. rubens* Ehrenberg, 1838

*Brachionus* spp.

-----Genus: *Keratella*

*K. cochlearis* ( Gosse, 1851 )

*K. quadrata* ( Müller, 1786 )

*K. tecta* (Gosse, 1851)

*K. tropica* ( Apstein, 1907 )

*K. valga* (Ehrenberg, 1834)

*Keratella* spp.

-----Genus : *Platyias*

*Platyias quadricornis* (Ehrenberg, 1832) .

-----Family : Dicranophoridae

-----Genus: *Dicranophorus*

*Dicranophorus prionacis* Haring & Myers, 1928 .

-----Genus: *Encentrum*

*Encentrum porsildi* Sørensen, 1998 .

-----Family: Euchlanidae

-----Genus: *Euchlanis*

*E. dilatata* Ehrenberg, 1832

*Euchlanis* sp.

-----Family: Gastropodidae

-----Genus: *Ascomorpha*

*A. saltans* Bartsch ,1870

*A. ovalis* Bergendahl, 1892

-----Family : Lecanidae

-----Genus: *Lecane*

*L. arcula* Haring,1914

*L. bulla* (Gosse, 1851)

*L. closterocerca* (Schmarda, 1859)

*L. elsa* Hauer, 1931

*L. hamata* (Stokes, 1896)

*L. inermis* Bryce, 1892

*L. ludwigii* (Eckstein, 1883).

*L. luna* (Müller, 1776)

*L. lunaris* (Ehrenberg, 1832)

*L. nana* (Murray, 1913)

*L. quadridentata* Ehrenberg, 1832

*L. tenuiseta* Haring, 1914

*L. thienemanni* (Hauer, 1938)

*L. unguate* Gosse, 1887

*Lecane* spp.

-----Family : Lepadellidae

-----Genus: *Colurella*

*C. adriatica* Ehrenberg, 1831

*Colurella* sp.

-----Genus: *Lepadella*

*L. patella* (Müller, 1773)

*Lepadella* sp.

-----Family : Mytilinidae

-----Genus: *Mytilina*

*M. ventralis* (Ehrenberg, 1830)

- Family : Notommatidae  
 -----Genus: *Cephalodella*  
                   *C. gibba* (Ehrenberg, 1832)  
                   *Cephalodella* sp.
- Family : Synchaetidae  
 -----Genus: *Polyarthra*  
                   *P. dolichoptera* Idelson, 1925  
                   *P. minor* Carlin, 1943  
                   *P. major* Burckhardt, 1900  
                   *P. remata* Skorikov, 1896
- Genus : *Synchaeta*  
                   *S. lakowitziana* Lucks, 1930  
                   *Synchaeta* spp.
- Family : Trichocercidae  
 -----Genus: *Trichocerca*  
                   *T. rattus* (Müller, 1776)  
                   *T. similis* (Wierzejski, 1893)  
                   *Trichocerca* sp.
- Family : Trichotriidae  
 -----Genus: *Macrochaetus*  
                   *M. sericus* (Thorpe, 1893)  
                   *Macrochaetus* sp.
- Genus: *Trichotria*  
                   *T. pocillum* (Müller, 1776)  
                   *T. tetractis* (Ehrenberg, 1830)

**Cladocera :**

- Kingdom: Animalia  
 --Phylum : Arthropoda  
 ---- Subphylum : Crustacea  
 ----- Class : Branchiopoda  
 -----Order : Anomopoda  
 ----- Suborder: Cladocera  
 -----Family : Acantholeberidae  
 -----Genus: *Acantholeberis*  
                   *Acantholeberis curvirostris* (Müller, 1776)

-----Family : Chydoridae  
 -----Genus : *Acroperus*  
                   *Acroperus angustatus* (Sars, 1863 )

-----Genus : *Alonella*  
                   *Alonella exigua* (Lilljeborg, 1853)

-----Genus : *Alona*  
                   *Alona guttata* (Sars,1862 )  
                   *Anthalona verrucosa* (Sars, 1901)  
                   *Alona* spp.

-----Genus : *Camptocercus*  
                   *Camptocercus uncinatus* (Smirnov, 1971)

-----Family : Daphniidae  
 -----Genus : *Daphnia*  
                   *Daphnia magna* (Straus, 1820)

-----Family : Bosminidae  
 -----Genus : *Eubosmina*  
                   *Eubosmina longispina* (Leydig, 1860)

### 3 – 7 : Key to the rotifera and cladocera in Al-Hilla river

#### 3 – 7-1 : Key to the Rotifera

1 -Rotifers with paired ovaries; trophi ramate ..... subclass **Bdelloidea** (2)

-Rotifers with a single ovary; trophi other than ramate

..... subclass **Monogononta** (4)

2-(1) Corona with paired trochal disks on raised pedicles

.....family **Philodinidae** (3)

- Not above .....other **bdelloid**.

3-(2) Rostrum, spurs, toes and dorsal antenna prominent;  
 ovoviviparous..... **Rotaria**

- Rostrum, spurs, toes and dorsal antenna not prominent; oviparous, The mouth-corners form squarebuilt protuberances ..... *Macrotrachela angusta*
- 4-(1) Trophi uncinata; corona a modified funnel with or without long setae, often without cilia, usually sessile..... Order **Collothecaceae (5)**
- Trophi malloramate ..... Order **Flosculariaceae (6)**
- Trophi not uncinata or malloramate ..... Order **Ploima (10)**
- 5- (4) a very large, circular, lobed or pointed. Setae not arranged in whorls. In a gelatinous tube. Foot terminated by a long, non-retractile peduncle, ending in an adhesive disc..... genus *Collotheca*
- Not above ..... other **Collothecaceae.**
- 6- (4) Body loricate ..... **Testudinellidae (7)**
- Body illoricate Body more or less a sphere... **Trochosphaeridae (8)**
- 7- (6) Foot opening central or slightly posterior to midline. Digestive tract with large lobed gastric glands, vitellarium horseshoe-shaped, antero-dorsal edge of lorica contain is wide medium pocket, antero-abdominal edge of lorica contain is clearly convex..... *T. patina*
- Ventral spin truncate, Head Aperture full width of anterior margin, Body length to 150  $\mu\text{m}$  ..... *T. reflexa*
- 8- (6) Body without appendages, Corona set off from body, antenna not on papillae ..... *Horaella brehmi*
- Body with elongate, occasionally moveable appendages ..... ( 9 )
- 9- (8) Body with one an immovable caudal seta inserted terminally. Lateral setae approximately 2 times longer than body, The body is oval to

- spherical shape ..... *F. terminalis*
- Body with two caudal seta, elongated, spindle shaped..... *F. opoliensis*
- 10-(4)** Trophi forcipate without alulae or with differently shaped alulae: stomach and intestine rarely with zoochlorellae .... **Dicranophoridae (11)**
- Trophi not forcipate.....**(12)**
- 11- (10)** Toes parallel –sided, foot a single pseudosegment, head long 1/3 total length, trophi large with 8-12 shearing teeth, contain alulae elongate – triangular ..... *Dicranophorus prionacis*
- Trophi with intramalleus between manubrium and uncus, foot short, less than 1/5 of the total length ..... *Encentrum porsildi*
- 12-(10)** Trophi incudate: illoricate: saccate body shape Vitellarium rounded. Trophicharacteristic; inner edge of each ramus with 4-6 teeth at its anterior ... ..... **Asplanchnidae, Asplanchna priodonta**
- Trophi not incudate; loricate or illoricated .....**(13)**
- 13-(12)** Foot and toes elongate, combined > than body length .....**(14)**
- Foot less elongate, toes may be long ..... **(15)**
- 14- (13)** Trophivirgate family **Scandiidae**, Alula rounded in lateral view. Manubrium relatively broad at articulation with uncus, anterior margin nearly straight ..... *Scaridium longicauda*
- Trophi malleate, Foot and toes slender, toes 1/3 length of dorsal plate ..... **Family Eucnianidae** ..... *Euchlanis dilatata*
- 15-(13)** Corona with four prominent setae (sensory bristles) and auricles (earlike structures); or possessing 12 movable, flattened, sword or feather shaped appendages (paddles); or sculptured lorica with ridges, grooves, or areolations..... Family **Synchaetidae (16)**
- Not above ..... **( 20)**
- 16-(15)** Body illoricate, square or somewhat rectangular in dorsal aspect, dorsoventrally compressed, with four bundles of serrated blade-shaped

- appendages at the anterior dorsal and ventral corners  
 ..... *Polyarthra* (17)
- Body illoricate, conical, bell- or vase-shaped; head usually wider than trunk, with lateral auricles and sensory setae. Foot retractable, up to about 1/2 total length, one or two small toes ..... *Synchaeta* (19)
- 17- (16) All fins equal of length..... (18)
- One dorsal fin visually longer than three others fins, fins width < 5 µm, small body, with ventral appendages. Vitellarium with 4 nuclei  
 ..... *p. minor*
- Fins as long as body, leaf or feather shaped with striation, width/length 1/5. Vitellarium with 8 nuclei .Four ramal teeth anterior to main tooth..... *p. major*
- 18-(17) Main fins longer than body, width 1/ 10 length or less, lateral antennae on or near postero–lateral corners of body, with ventral appendages ..... *P. dolichoptera*
- Main fins slightly longer than total body length, width /length ratio 1/10, without ventral appendages ..... *P. remata*
- 19 - Foot at least 1/5 of total length, Head differentiated from trunk by constriction below auricles ..... *Synchaet alakowitziana*
- Not above ..... other *Synchaeta*
- 20- (15) Loricata ..... 21
- Illoricate, Trophi virgate ,Toes dorsally bent, medium toes, total length /toe length = 3 - 5, total length > 250 µm, frontal eyespot(s), Family **Notommatidae**, ..... *Cephalodella gibba*
- 21-(20) Sulci present: lorica with 1 or more longitudinal sulci, ventral, dorsal, &/orlateral..... 22
- Sulci absent ..... (52)
- 22-(21) Lorica possessing a ventral and a dorsal plate: sharp-edged dorsal keel may be present: sulci laterally only .....23

- Lorica not as above .....(50)
- 23- (22) Foot absent .....24
- Foot present, but may be difficult to see in contracted specimens ....( 36)
- 24- (23) Trophi malleate ..... **Brachionidae (25)**
- Trophi virgate ..... Family: Gastropodidae , *Ascomorpha (35)*
- 25-(24) Foot present ..... (26)
- Foot absent. Lorica marked on dorsal surface with polygonal facets.  
..... Genus *Keratella (32)*
- 26- (25) Foot annulated, retractile within body.... Genus *Brachionus (27)*
- Foot jointed, not retractile within body.....*Platytias quadricornis*
- 27- (26) With 6 anterior spines ..... (31)
- With 2 or 4 anterior spines ..... (28)
- 28- (28) Median occipital spines short ..... (29)
- Median occipital spines not short ..... (30)
- 29- (28) Two median occipital spines present, submedian spines and  
anterior spines absent ..... *Brachionus angularis*
- Anterior lateral spines not elongated, median spines short...*B.forficula*
- 30-(28) Lorica thick, with dorsal plaques, median spines bent vertically  
and longer than lateral spines, without caudal spines ...*B.budapestinensis*
- Lorica thin, without plaques, variable length spines, with or without  
caudal spines ..... *B. calyciflorus*
- 31-(27) Submedian anterior spines very long compared with other  
spines..... *B. falcatus*
- anterior median spines usually longer than other..... *B. quadridentatus*
- Median and intermediate spines showing a narrow anteriorpart,  
rounding outward and forming a broadbase. Ventral foot aperture nearly  
parallel-sided.....*B. rubens*
- 32- Dorsal plate with median row of plaques .....(33)
- Dorsal plate with median ridge or keel ..... (34)

- 33-(32) Lorica broader at posterior end, nearly rectangular .. *K. quadrata*
- Posteromedian remnant under posteromedian plaque, lorica prolonged distinctly, last facets small, posterior spines usually long to short, parallel or diverging, sometimes only one spine or none ..... *K.tropica*
  - Posteromedian remnant absent, lorica broadest in middle third of length ..... *K. valga*
- 34-(32) Median frontal plaque pentagonal, plaques nearly symmetrical, with median caudal spines ..... *K. cochlearis*
- Median frontal plaque pentagonal, plaques nearly symmetrical, small body, without caudal spines ..... *K. tecta*
- 35- lorica formed by a dorsal and a shield-like ventral plate ..... *Ascomorpha ovalis*
- plates, cuticle one piece and not contain palpar organ..... *Ascomorpha saltans*
- 36- Foot short, inserted ventrally on ventral plate ..... **Lecanidae (37)**
- Foot short or elongate, inserted caudally: foot and toes project from betweenventral and dorsal plate ..... **Euchlanidae (49)**
- 37- (36) Toes separate, mostly spread ..... (38)
- Toes partly or completely fused, mostly held together ..... (45)
- 38 -(37) Lorica with dorsal and ventral plate, these may be soft ..... (39)
- Body illoricate: dorsal or ventral plate not distinguishable toes with relatively long claw ..... *L. inermis*
- 39-(38) Toes bearing claws or pseudoclaws .....(40)
- Toes ending in acute points, no claws ..... (41)
- 40 - (39) Antero-lateral corners angulate or rounded. Transverse fold on ventral plate complete, strong. Toes relatively short ..... *L. elsa*
- Antero-lateral corners with acute projections ..... (42)

- 41-(39)** Toes ending in acute points, no claws nor pseudoclaws, toe tips may be asymmetrical. Antero – lateral corners angulate. Foot pseudosegment not parallel – sided ..... *L. nana*
- Dorsal plate medially wider than ventral plate . Lorica elongate, mostly ornamented .....*L. ludwigii*
- 42-(40)** Footpseudosegment scarcely projecting .Lorica distinctly longer than wide , mostly smooth. .... *L. tenuiseta*
- Transver fold on ventral plate complete, lunate or incomplete, no longitudinal folds. Prepedal fold rounded posteriorly. Mostly small species.Pseudoclaws 20 µm or longer .....(43)
- 43- (42)** Pseudoclaws 20 µm or longer ..... *L. unguate*
- Pseudoclaws shorter than 20 µm ..... ( 44 )
- 44-(43)** Anterior margin of dorsal plate much narrower than that of ventral plate. Head aperture margins strongly lunate, antero-lateral spines broad-based ..... *L.luna*
- 45-** Lorica egg-shaped, occasionally soft. Ventral and dorsal head aperture margins with deep sinuses. Toe either with terminal fissure and long pseudoclaws or with fused pseudoclaws, accessory claws present ..... *L. bulla*
- Lorica dorso – ventrally flattened, not egg-shaped ..... (46)
- 46-(45)** Head aperture margin dorsally with pair of long , curved median projections, ventrally concave ..... *L. quadridentata*
- Dorsal plate medially wider than ventral . Lateral margins of dorsal plate reach head aperture margins .....(47)
- 47- (46)** Ventral and dorsal head aperture margins variable: straight to strongly concave ..... *L. lunaris*
- not above .....(48)

- 48-(47) Ventral and dorsal head aperture margins straight or smoothly concave ..... *L. closteroerca*
- Dorsal head aperture margin concave or nearly straight, broad, foot pseudosegment not projecting..... *L. hamata*
- Dorsal head aperture margin nearly straight, narrow, foot pseudosegment projecting ..... *L.thienemanni*
- 49-Dorsal plate arched, wider than the flat ventral plate..Genus *Euchlanis*
- Foot and toes slender, toes 1/3 length of dorsal plate ..... *E. dilatata*
- 50- Lorica possessing a distinct dorsal suicus, Family : Mytilinidae ..... *Mytilina ventralis*
- Lorica possessing a ventral suicus Trophi malleate .....Family: ..... **Lepadellidae (51)**
- 51-(50)Lorica valve posteriorly with curved spines... *Colurella adriatica*
- Lorica oval, egg- or pear-shaped or elongate, 1 piece; anterior opening narrow, semicircular, two toes pointed Cross-section of dorsal lorica 1/3 circle to hemispherical ..... *Lepadella patella*
- 52- Foot inserted terminally, consisting of a single, short pseudosegment and bearing elongate, occasionally curved and/or asymmetrical spine-like toes.....**Trichocercidae (53)**
- Lorica relatively flexible. covering head, trunk, and foot; toes sometimes long ..... **Trichotriidae (54)**
- 53-(52) Lorica anterior margin with spines (projections). Two spines long and slender of about equal in length; body shaped as elongated cone, right manubrium is larger than left manubrium and the oval cavity at its upper end ..... *Trichocerca similis*
- Left toe length more or less equal to lorica length, With one dorsal keel, two manubrium long and curved at the end towards the inside, left manubrium is much larger than right manubrium, fulcrum is long, thin, straight and ends with a bulge ..... *Trichocerca rattus*

54-(52) Dorsal surface of lorica with pairs of long spines placed symmetrically to mid-line Two pair of spines on dorsal plate..... *Macrochaetu ssericus*

-Dorsal surface without spines, or at most with 1 or 2 spines on mid-line, first joint of foot with heavy dorsal spines ..... *Trichotria (55)*

55-(54) Last foot joint with dorsal minute spine between the toe bases (Short spine located between toes) ..... *Trichotria pocillum*

- Elongated triangle projection at foot-body join ..... *Trichotria tetractis*

### 3 – 7-2: Key to the Cladocera (Branchiopoda)

1- First antennae fused with the rostrum to form two long and pointed tusk-like structures family Bosminidae, Mucro present- Carapace elongated - Antennules no longer than body length..... *Eubosmina longispina*

- First antennae not fused with the rostrum.....2

2-(1) One branch of the second antenna with three segments, the other (dorsal) branch with four segments ,Thoracic legs 3 and 4 with more than 40 setae total, mostly a group of closely set setae forming a filter-like comb family **Daphniidae**, postabdominal dorsal margin with an indentation or slightly concave - Fornix pointed and laterally extended, helmet low and broad with ridges, lateral rib present, postabdominal dorsal margin with a deep indentation..... *Daphnia magna*

- Both branches of the second antenna not three segments, First antenna is attached just ventral to the anterior tip of the head, there is no rostrum families **Acantholeberidae** , Rear bottom part of carapace valve with a group of long setae .....*Acantholeberis curvirostris*

- Both branches of the second antenna with three segments..... (3)

- 3-(2) Posterior margin of the valve much lower than the maximal height of the carapace. Exopodite of the 4th pair of thoracic limbs with 7 setae, Postabdomen relatively shorter. Dorsal-distal margin of the postabdomen with several single teeth, not associated with smaller secondary teeth..... *Alonella exigua*
- Posterior margin of the valve slightly lower than the maximal height of the carapace. Exopodite of the 4th pair of thoracic limbs with 6 setae.... 4
- 4-(3) Postabdomen long and narrowing distally, postabdomen without distal embayment ..... *Camptocercus uncinatus*
- Postabdomen not long and narrowing distally ..... 5
- 5- Postabdomen long and narrow, Dorsal margin of the postabdomen without teeth ,Head large, with concave anterior margin..... *Acroperus angustatus*
- Postabdomen not long and narrow .....6
- 6- The preanal anal and postanal margins of the postabdomen equal in length, the dorsal margin of the postabdomen with serrated teeth..... *Alona guttata*
- Sacs underneath lateral head pores “Cosmarium-like” or lobed ..... *Anthalona verrucosa*

### 3 – 8: New Record Species of Rotifera and Cladocera

#### 3-8-1: New Record Species of Rotifera

##### 1 – *Ascomorpha ovalis* ( Bergendahl, 1892)

#### Diagnosis :

Lorica consist of dorsal and ventral plates covered with a thin membrane. Besides the single main apical tentacle two palpal organs present. Cerebral eye placed medially, dorsal antenna small. Unci thin long rods, manubria crutched, rami somewhat asymmetric with long alulae and suprarami . Stomach with four blind sacs, rectum and anus absent (Figure 28) .

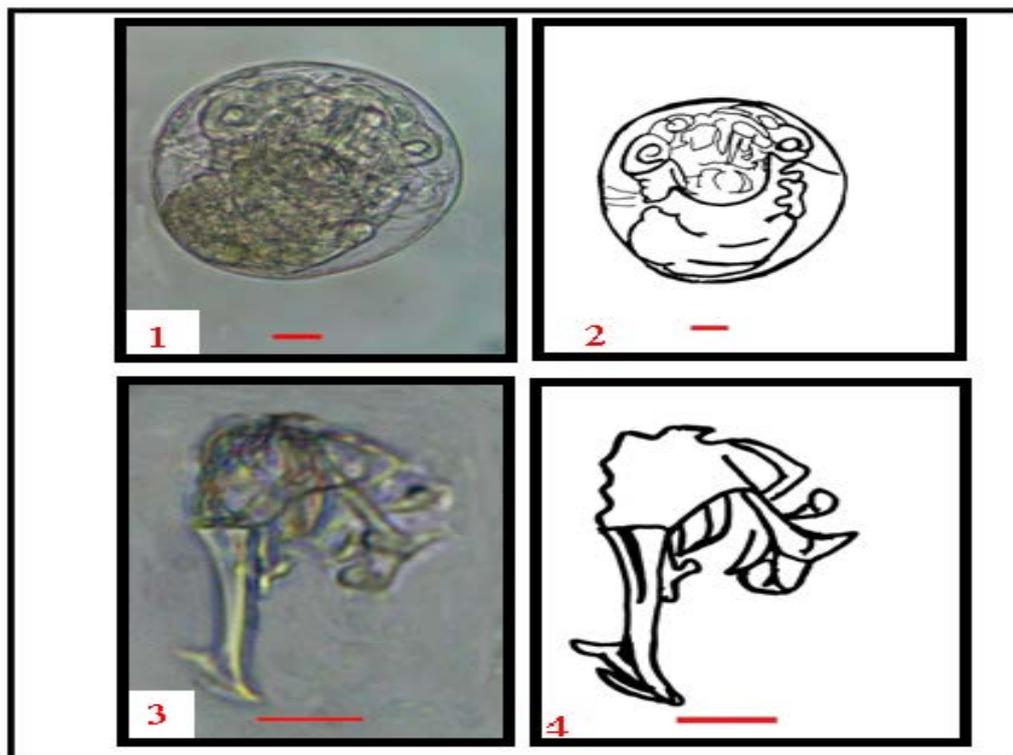


Figure 28: (1-2) *Ascomorpha ovalis*, (3-4) Trophus of *Ascomorpha ovalis* Scale bar 50  $\mu$ m for all

##### 2 - *Horaella brehmi* ( Donner, 1949)

#### Diagnosis :

Body saccate, broadly elliptical, transparent. Apical field bare, arched. Corona simple, circumapical band with small interruption dorsally

followed by a collar-like constriction. Ventral mouth distinct. Lateral antennas in anterior third of body is simple. Stomach with large, flat epithelial cells, a single pair of flat stomach glands lays against the stomach. Stomach and intestine separated by a constriction, intestine round, large. Cloaca without caeca, bladder large, anus terminal. Vitellarium rounded with 8 nuclei, ovarium part discernible. Ovoviviparous, developing eggs commonly present in the genital tract of the female, the young emerges as the egg is laid. Trophi malleoramate, symmetrical with two large and 14 – 15 small unci teeth, proximal tooth with anterior projection (Figure 29).

Body length 280 – 340  $\mu\text{m}$ , width 200  $\mu\text{m}$ , corona diameter 66  $\mu\text{m}$ , trophi width 37 – 45  $\mu\text{m}$ , trophi length 23 – 26  $\mu\text{m}$ , unci plate length 14 – 16  $\mu\text{m}$ , unci plate width 13.6 – 15.2  $\mu\text{m}$ , rami length 19 – 20  $\mu\text{m}$ , fulcrum 7  $\mu\text{m}$ .

### **3 - *Lecane arcula* (Harring, 1914)**

#### **Diagnosis :**

Lorica stiff, Dorsal plate anteriorly narrower, medially wider than ventral, ornamented. Head aperture margins nearly coincident, straight with acutely pointed antero-lateral spines emerging from between the dorsal and ventral plate. Ventral plate longer than wide with incomplete transverse and longitudinal folds, ornamented. Lateral margins irregularly undulate, straight, parallel or diverging to anteriorly. Anterior notches

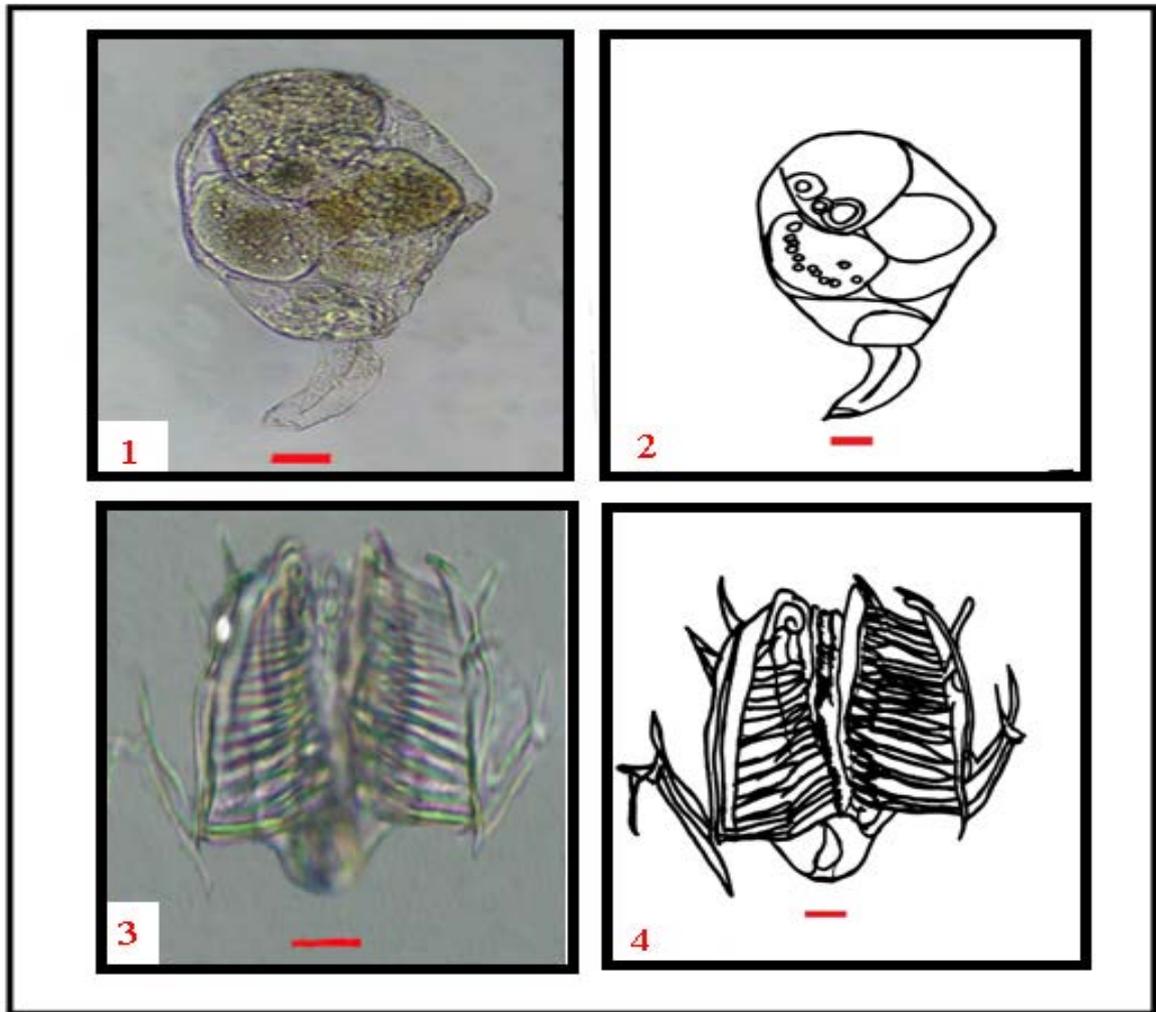


Figure 29: (1-2) *Horaella brehmi*, (3-4) Trophus of *Horaella brehmi* Scale bar 50  $\mu$ m for all

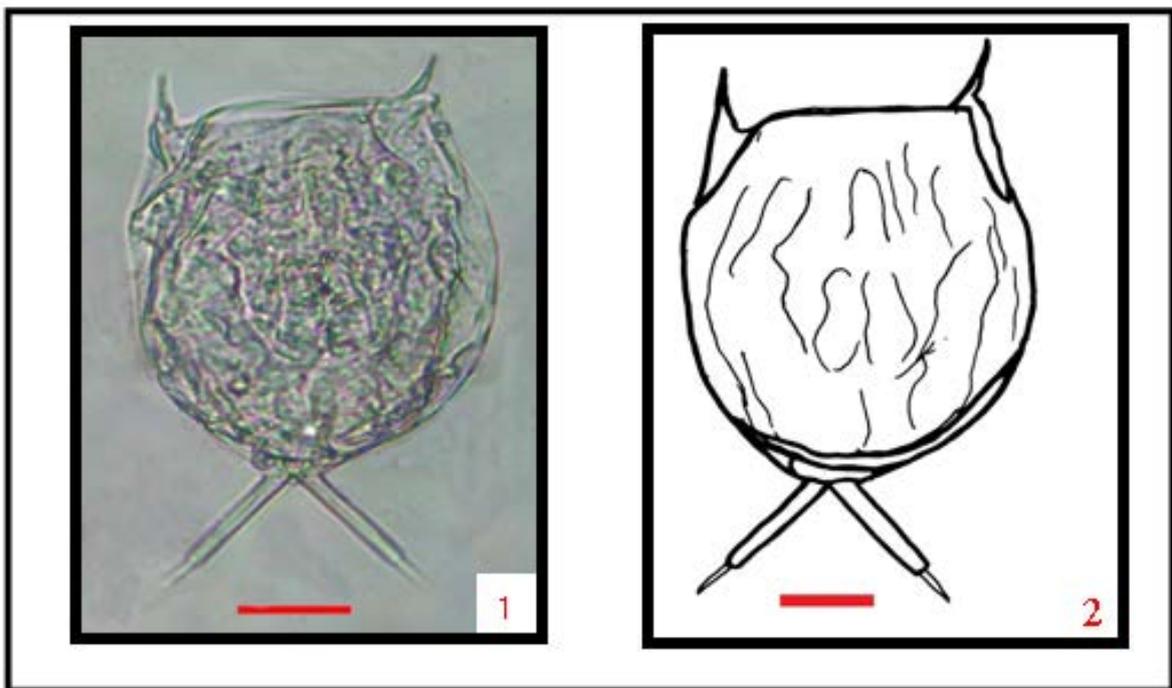


Figure 30: (1-2) *Lecane arcula*. Scale bar 50  $\mu$ m for all

mostly present. Lateral sulci shallow. Foot plate nearly as wide as long, with rounded triangular coxal plates. Prepedal fold narrow, elongate, posteriorly with median projection. Foot pseudosegment projecting, with lateral lobes. Toes parallel-sided, occasionally bent proximally. Claws completely separated, needle-like. Accessory claws present, these sometimes inconspicuous (Figure 30).

Dorsal plate length 57 – 63  $\mu\text{m}$  , Dorsal plate width 49 – 56  $\mu\text{m}$  , Ventral plate length 72 – 87  $\mu\text{m}$  , Ventral plate width 46 – 52  $\mu\text{m}$  , toe length 22 – 27  $\mu\text{m}$  , claw length 4 – 6  $\mu\text{m}$ , antero-lateral spine length 3 – 5  $\mu\text{m}$ .

#### 4 - *Lecane quadridentata* ( Ehrenberg, 1832)

##### Diagnosis :

Lorica stiff and smooth. Dorsal plate narrower than ventral plate, lateral margins reach anterior end of lorica. Head aperture margins dorsally variable with pair of conspicuous outward curved antero-median projections, ventrally variably concave. Antero-lateral corners angulate or projecting. Ventral plate longer than wide to elongate, widest in the distal third, transverse folds complete. Lateral margins smooth, curved. Lateral sulci deep. Foot plate relatively narrow, coxal plates rounded, simple. Prepedal fold broad, with rounded distal margin. Foot pseudosegment projecting. Toe single, nearly parallel-sided, bearing fused pseudoclaws and accessory claws (Figure 31).

Dorsal plate length 104 – 196  $\mu\text{m}$ , Dorsal plate width 79 – 130  $\mu\text{m}$ , Ventral plate length 110 – 194  $\mu\text{m}$ , Ventral plate width 81 – 133  $\mu\text{m}$ , toe length 45 – 93  $\mu\text{m}$ , claw length 14 – 26  $\mu\text{m}$ , antero-median projections length 18 – 51  $\mu\text{m}$ .

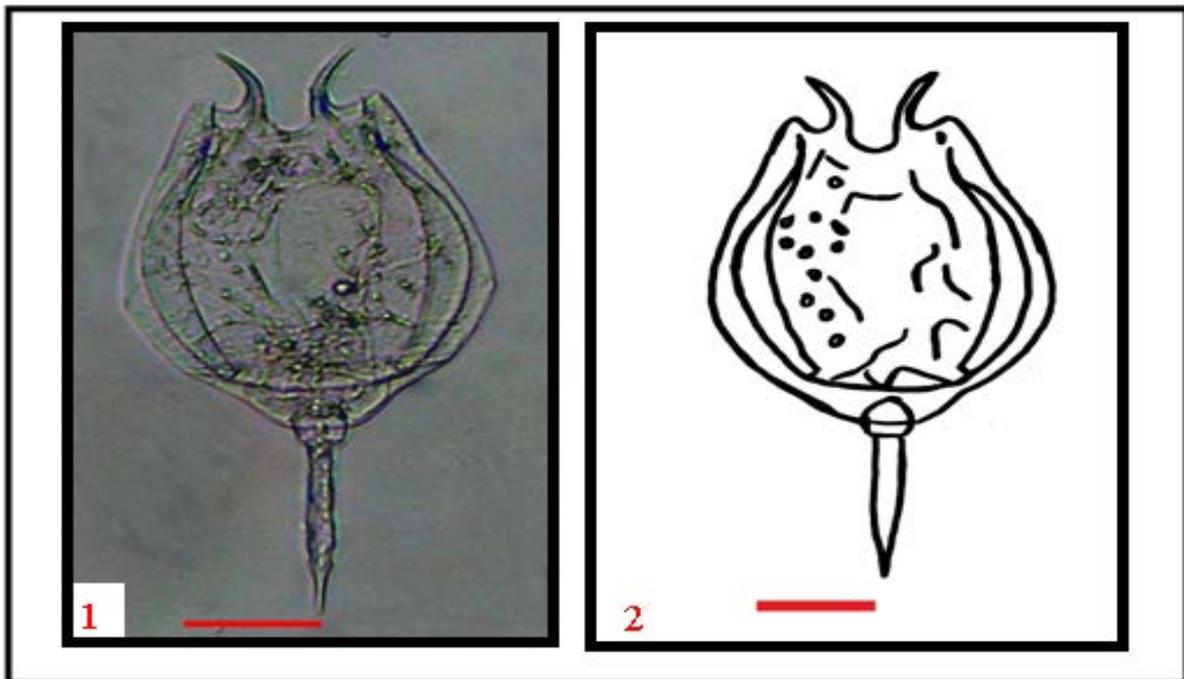


Figure 31: (1-2) *Lecane quadridentata*. Scale bar 50  $\mu\text{m}$

### 5 - *Lecane tenuiseta* (Harring, 1914)

#### Diagnosis :

Lorica occasionally soft, though distinct. Dorsal plate anteriorly narrower, medially as wide as ventral plate, smooth. Head aperture margins nearly coincident or parallel, almost straight, at times irregular. Antero-lateral corners angulate. Ventral plate longer than wide, with incomplete transverse and longitudinal folds, smooth or ornamented. Lateral margins smooth, nearly straight. Lateral sulci deep. Foot plate broad, rounded posteriorly, coxal plates rounded. Prepedal fold narrow, elongate, distally with median projection. Foot pseudosegment not or slightly projecting. Toes parallel-sided, bearing completely separated, long claws (Figure 32).

Dorsal plate length 51 – 58  $\mu\text{m}$ , Dorsal plate width 52 – 60  $\mu\text{m}$ , Ventral plate length 57 – 83  $\mu\text{m}$ , Ventral plate width 52 – 60  $\mu\text{m}$ , toe length 20-24  $\mu\text{m}$ , claw length 12 – 16  $\mu\text{m}$ .

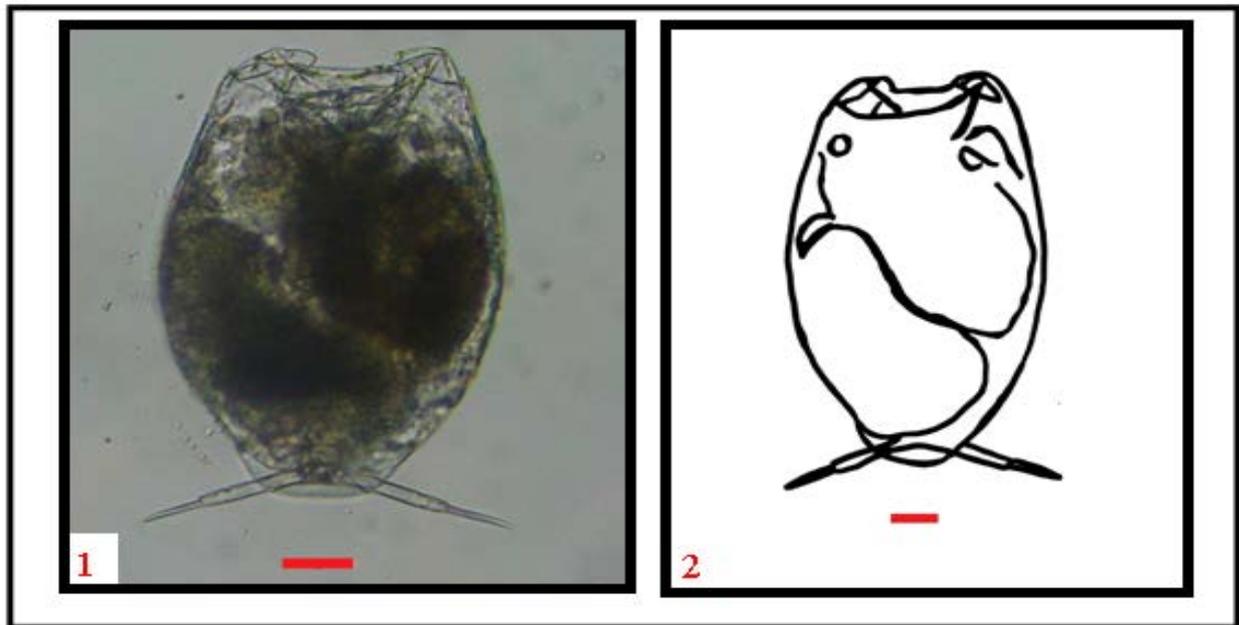


Figure 32: (1-2) *Lecane tenuiseta*. Scale bar 50  $\mu\text{m}$  for all

### 6 – *Lecane ungulata* (Gosse, 1887)

#### Diagnosis :

Loricata. Dorsal plate as wide as or narrower than ventral plate. Head aperture margins dorsally and ventrally concave, straight or dorsally slightly convex. Antero-lateral spines present. Dorsal plate smooth, occasionally with median dome anteriorly. Lateral edges don't reach the head aperture. Ventral plate longer than wide, occasionally ornamented. Incomplete transverse folds. Lateral margins slightly curved, smooth. Lateral sulci deep. Foot plate broad, truncate posteriorly, coxal plates rounded triangular. Prepedal fold broad, rounded distally. Foot pseudosegment trapezoidal, mostly non-projecting. Toes parallel-sided

commonly with local constrictions. Long, incompletely separated pseudoclaws and accessory claws present (Figure 33).

Dorsal plate length 140 – 260  $\mu\text{m}$ , Dorsal plate width 138 – 230  $\mu\text{m}$ , Ventral plate length 185 – 300  $\mu\text{m}$ , Ventral plate width 139 – 230  $\mu\text{m}$ , toe length 48 – 90  $\mu\text{m}$ , claw length 20 – 50  $\mu\text{m}$ .

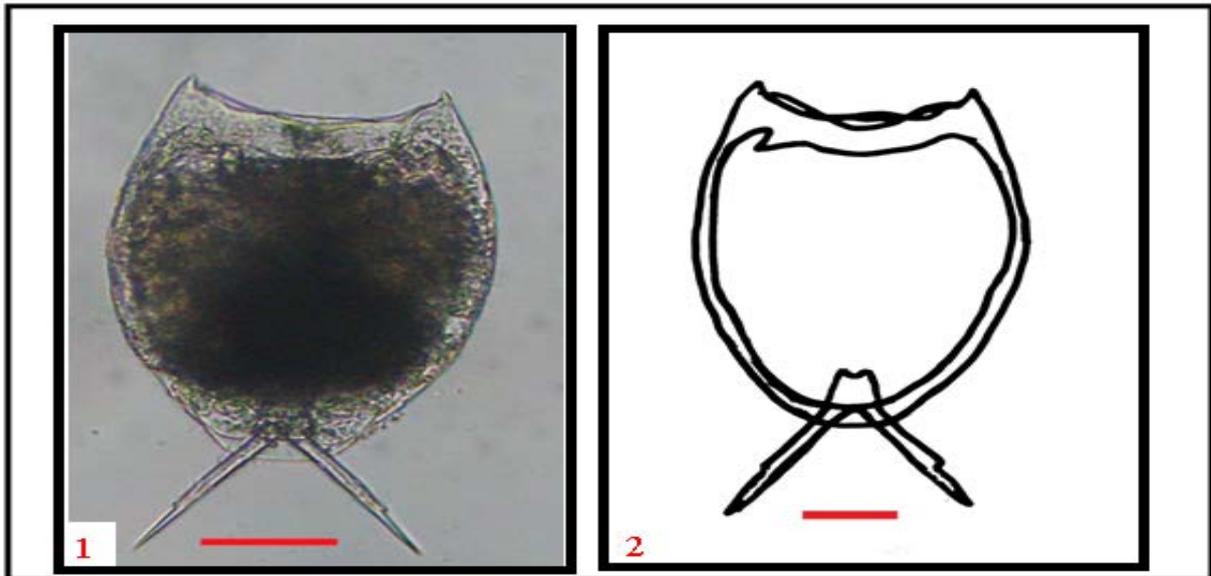


Figure 33: (1-2) *Lecane unguolata*. Scale bar 50  $\mu\text{m}$  for all

### 7 - *Scaridium longicaudum* (O.F. Muller, 1786)

#### Diagnosis :

Animal relatively large, second foot pseudosegment and toes short. Fulcrum with high midventral crest, basal plate well developed. Rami teeth large, aulacae rounded. Manubrium relatively broad and transversely divided proximally, ventral margin nearly straight. Ventroposterior projection large, rounded, exceptionally asymmetrical. No posterior lamella. Epipharynx with equal small teeth (Figure 34).

Total length 358 – 429  $\mu\text{m}$ , body 125 – 168  $\mu\text{m}$ , trun 91 – 134  $\mu\text{m}$ , second foot pseudosegment 34 – 44  $\mu\text{m}$ , third foot pseudosegment 72 –

90  $\mu\text{m}$ , toe 116 – 149  $\mu\text{m}$ , fulcrum 27 – 34  $\mu\text{m}$ , parthenogenetic egg 50  $\mu\text{m}$ .

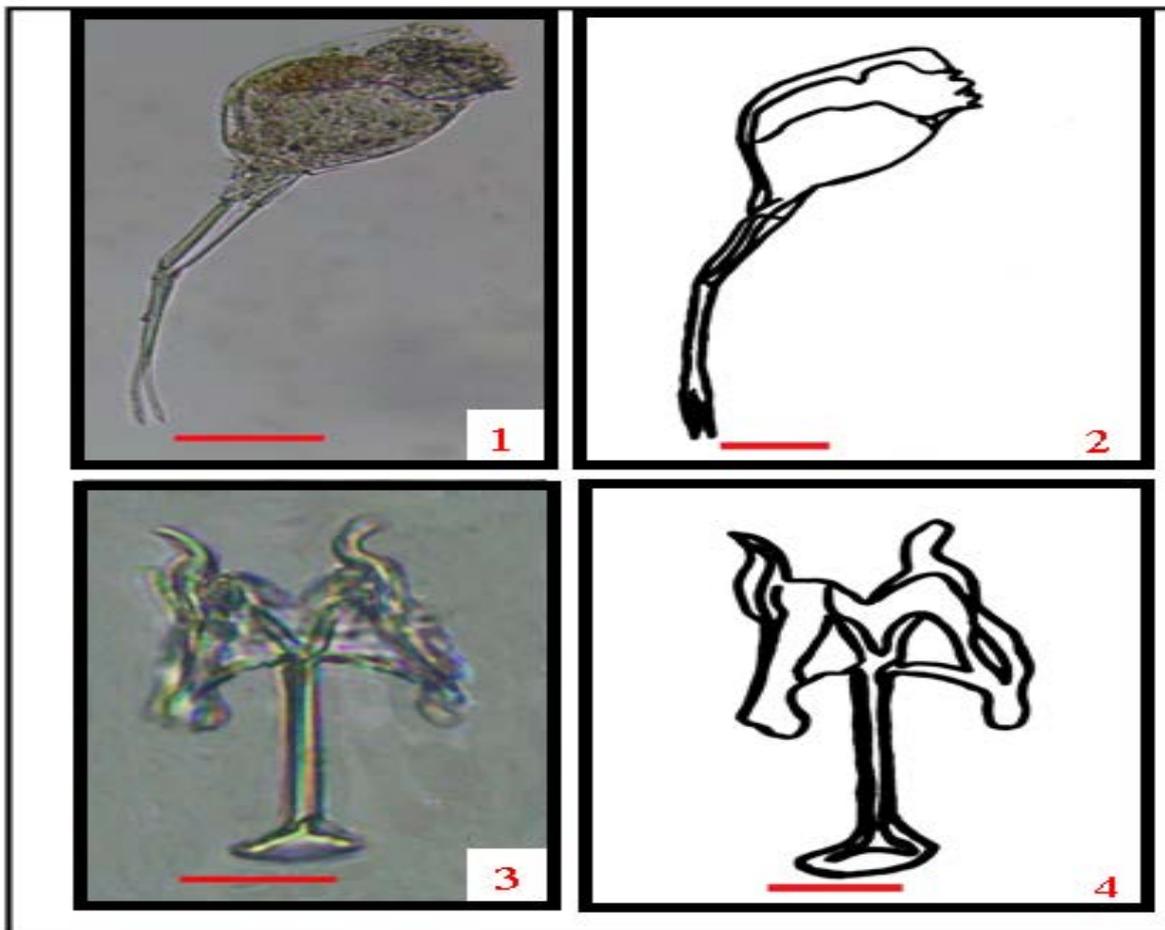


Figure 34: (1-2) *Scaridium longicaudum*, (3-4) Trophus of *Scaridium longicaudum* Scale bar 50  $\mu\text{m}$

### 3-8-2: New Record Species and Other Species of Cladocera

#### 1 - *Acantholeberis curvirostris* (O.F Muller, 1776)

##### Diagnosis :

General body outline (seen laterally) subquadrangular . Ventral margin of head even. On the ventro-posterior corner , 12 – 14 very long setae. The largest seta of the antenna with spines along its distal part. Labrum with an acute outgrowth (unpaired). Parthenogenetic eggs up to 12 ,

ephippium with 1-8 eggs. Length up to 2 mm. Vas deferens opens at base of claw. Length up to 0.7 mm (Figure 35).

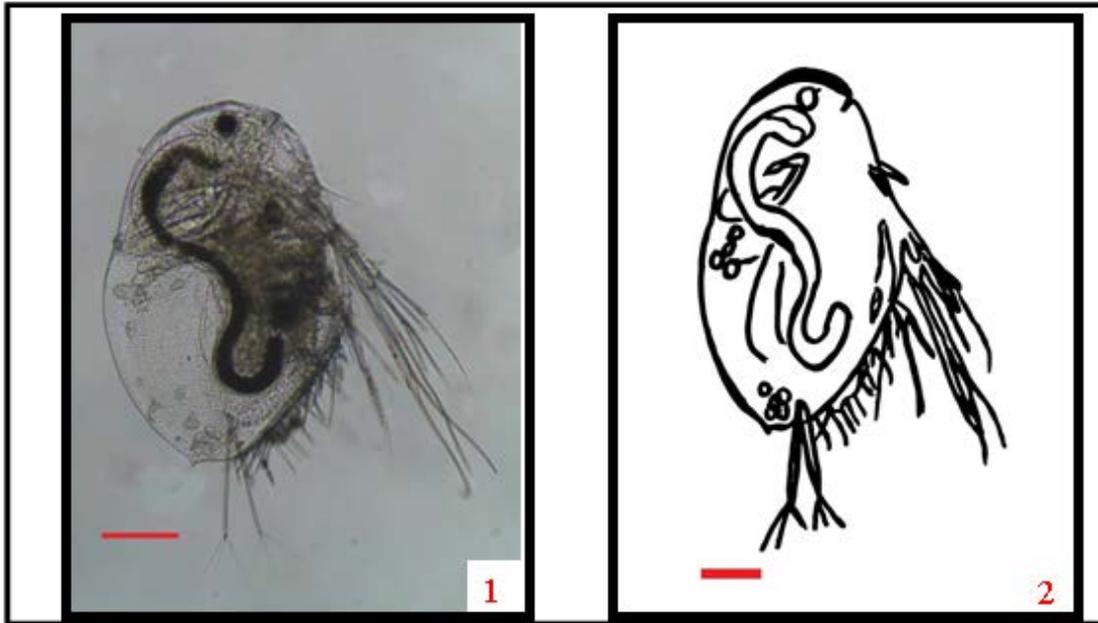


Figure 35: (1-2) *Acantholeberis curvirostris*. Scale bar 50  $\mu\text{m}$  for all

### 2 – *Acroperus angustatus* (Sars, 1863)

#### Diagnosis :

More compressed laterally with maximum height before or at the midline. the seta arising from the basal segment is long, approximately the same length as the second segment. Its eye is placed approximately in the centre of the head shield and the carapace has stripes going diagonally from the upper front corner down to the lower post corner. The post abdomen is long and slender and is normally protruding out of the carapace. The carapace is very transparent and almost complete colorless (Figure 36).

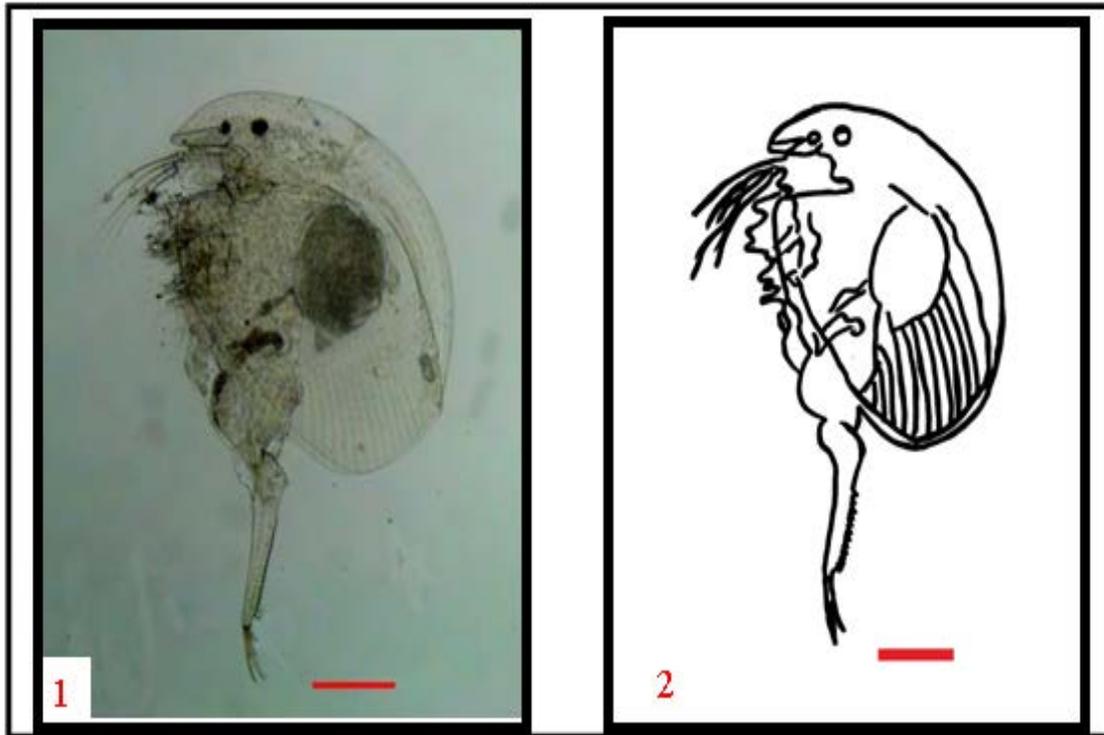


Figure 36: (1-2) *Acroperus angustatus*. Scale bar 50  $\mu\text{m}$  for all

### 3 – *Alona guttata* (Sars, 1862 )

#### Diagnosis :

Body oval, moderately high. Sculpture of valves as weakly developed longitudinal lines or tubercles. Three main head pores. Labral keel of moderate width, with blunt apex. Postabdomen short, moderately wide, slightly narrowing distally, with acute, protruding distal angle. Postanal denticles short. Postanal lateral setulae of postabdomen weakly developed, thin, the longest setule located at the middle of group. Inner distal lobe of thoracic limb I with three setae, 1 seta very small, rudimentary, setae 2–3 thin, armed with thin setulae (Figuer 37).

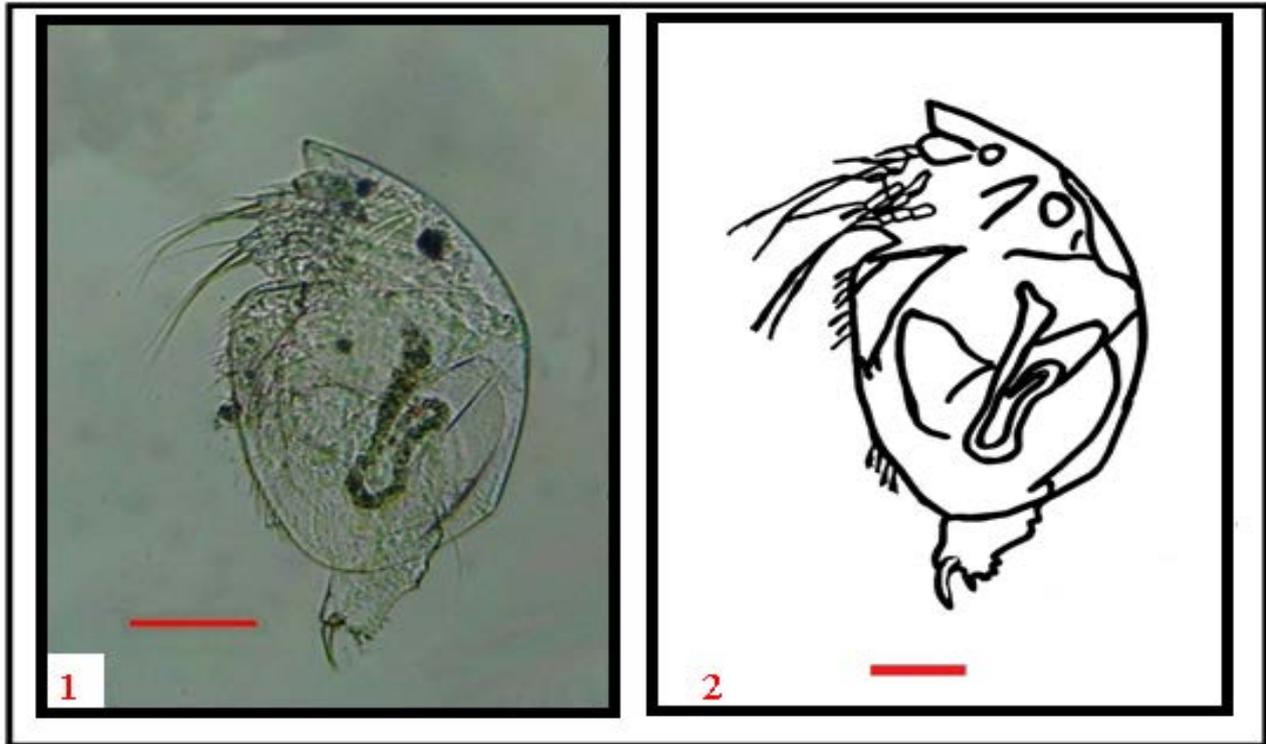


Figure 37: (1-2) *Alona guttata*. Scale bar 50  $\mu\text{m}$  for all

#### 4 - *Anthalona verrucosa* (Sars, 1901)

##### Diagnosis :

*Anthalona verrucosa* has a typical single denticle (or two) on the labral keel. The species is relatively small (0.34–0.36 mm on average). Valve setae with a posterior group are relatively long and widely spaced. Postabdomen with lateral fascicles not (or rarely) reaching beyond the marginal teeth; basal spinules on terminal claw half of basal spine or longer. Major pores very variable at some distance from the lateral pores. Body shape may differ with an expanded posteroventral portion. On antennae, terminal (swimming) setae are normal, unmodified and the group of spinules on first and second exopod segments are modified and strong. On limbs, *A. verrucosa* has 8–11 teeth on the sixth scraper of second limb (a good character) (Figure 38).

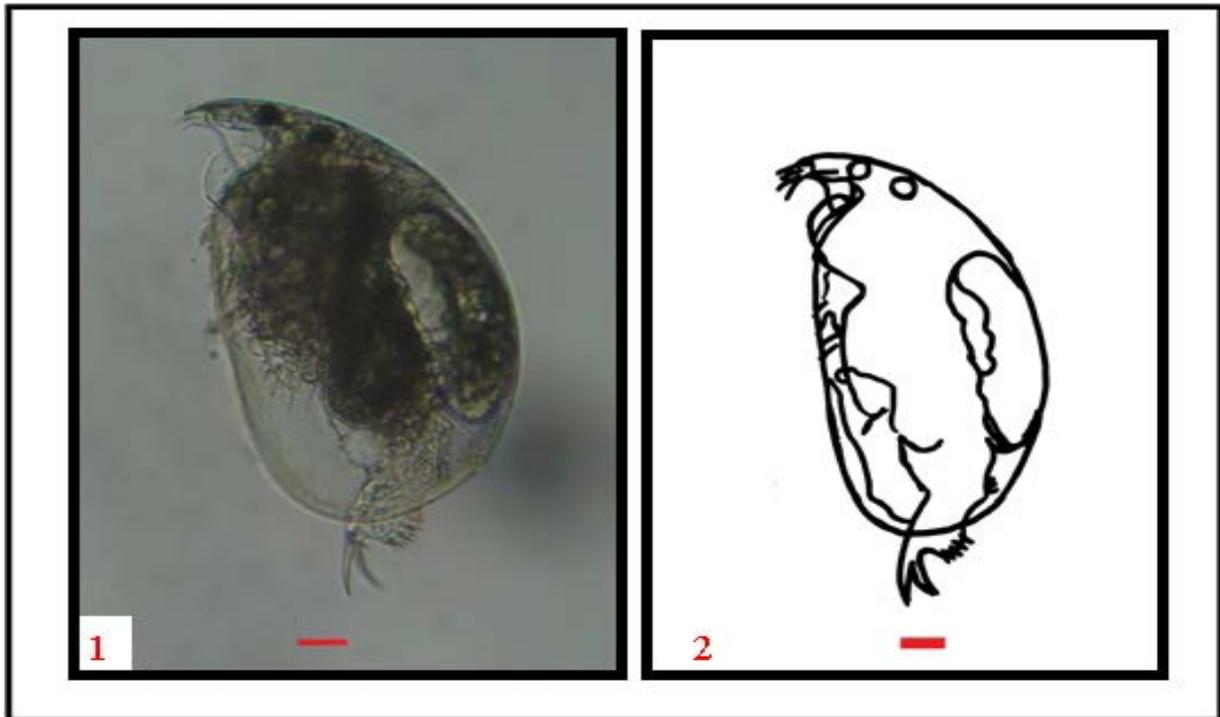


Figure 38: (1-2) *Anthalona verrucosa*. Scale bar 50  $\mu\text{m}$  for all

### 5 - *Alonella exigua* (Lilleborg, 1853)

#### Diagnosis :

Female: Postero-ventral angle of valve with denticles of variable acuity. Posterior margin straight. Valves with polygons (no striation in polygons). Ventral setae marginal on anterior and middle, and submarginal on posterior zone of valves. Embayment between valves (seen ventrally) greatly expanded at their middle part. Head shield with rounded anterior and posterior margins. Labral plate cuneiform. The antennule doesn't reach the tip of rostrum by less than the antennular length. The antenna with setae 0 -0-3/ 1-1-3. The postabdomen elongated, its preanal angle prominent. Anal teeth 9. Claw with 2 basal spines. The inner distal lobe of thoracic limb I with 3 setae of different length. Length up to 0.48 mm (Figure 39).

Male : The postabdomen with narrow distal part. Adult male have postabdomen with setae on narrow distal part , preanal angle prominent. Claw with 2 basal spines. Length up to 0.27 mm.

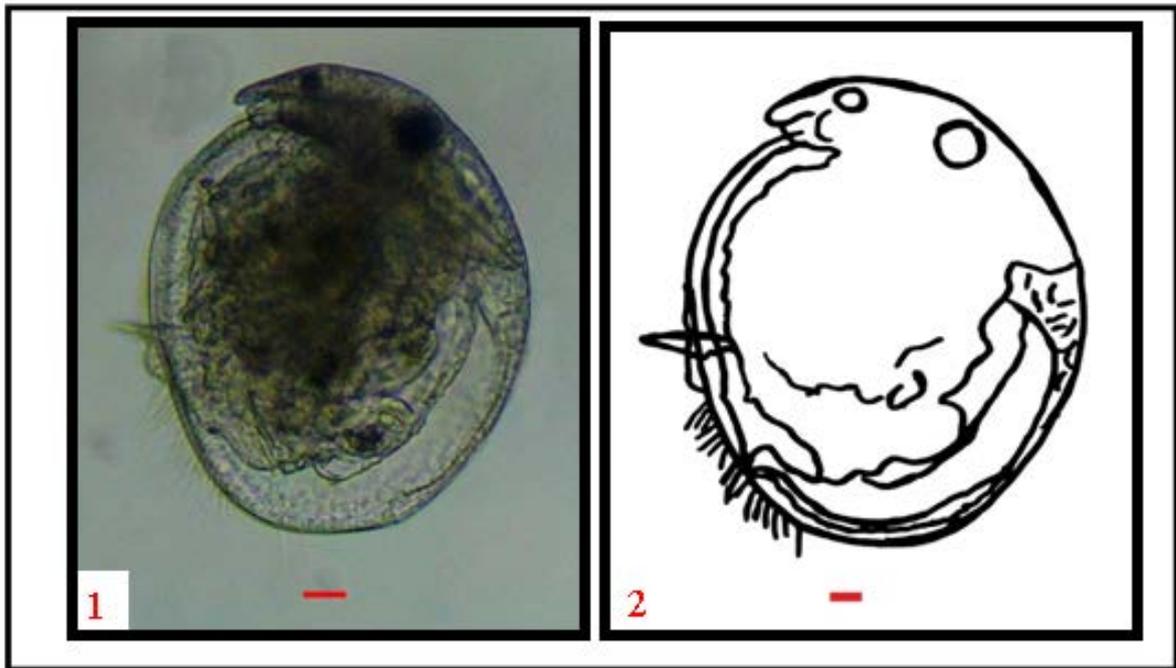


Figure 39: (1-2) *Alonella exigua*. Scale bar 50  $\mu$ m for all

### 6 - *Camptocercus uncinatus* (Smirnov,1971 )

#### Diagnosis :

Body curved. Rostrum blunt, ocellus situated slightly nearer to eye than to apex of rostrum. Antennules all reaching apex of rostrum. Posterior margin of the valves curved and convex, ventral margin with setae, valves with distinct longitudinal lines laterally; postero-ventral corner of valves without denticles. Postabdomen with approximately 16 pointed anal denticles decreasing in size proximally, most anal denticles multicuspid; lateral denticles indistinct and forming groups; claw with a distinct denticle in the middle of concave side and with a row of setae proximally, setae decreasing towards the base; basal spine of claw tapering. Leg with 3 setae on outer branch of endite, one of them is

shorter than other. The basal setule on both long setae is very large and hook-shaped (Figure 40).

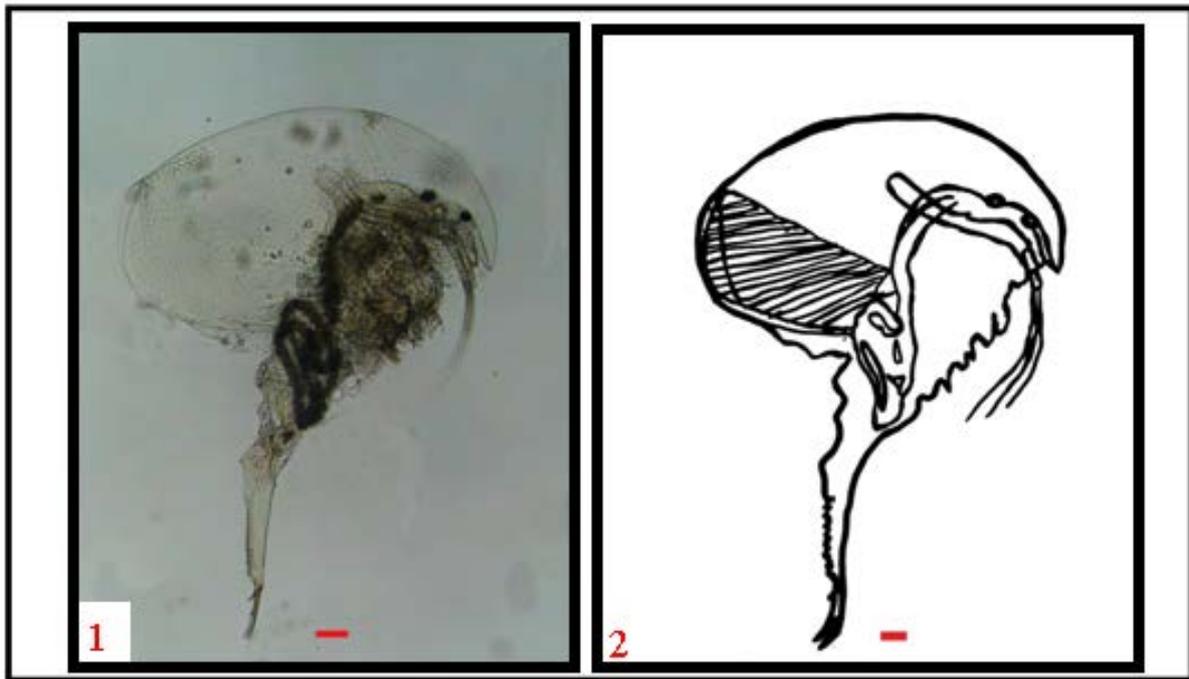


Figure 40: (1-2) *Camptocercus uncinatus*. Scale bar 50  $\mu\text{m}$  for all

### 7 – *Daphnia magna* (Straus, 1820 )

#### Diagnosis :

Has a short and stocky body shape with a short spine. In the middle of the dorsal rand there is a group of 20-25 long bristles. the postabdomen has two groups of about 10 coarse denticles, fastened to the hind edge where this constitutes an angle of about 45o. the abdominal claw has a varying number of coarse denticles close to the basis (Figure 41).

Female: Length 2.9–6.0 mm

Male: Length 2.2–3.5 mm

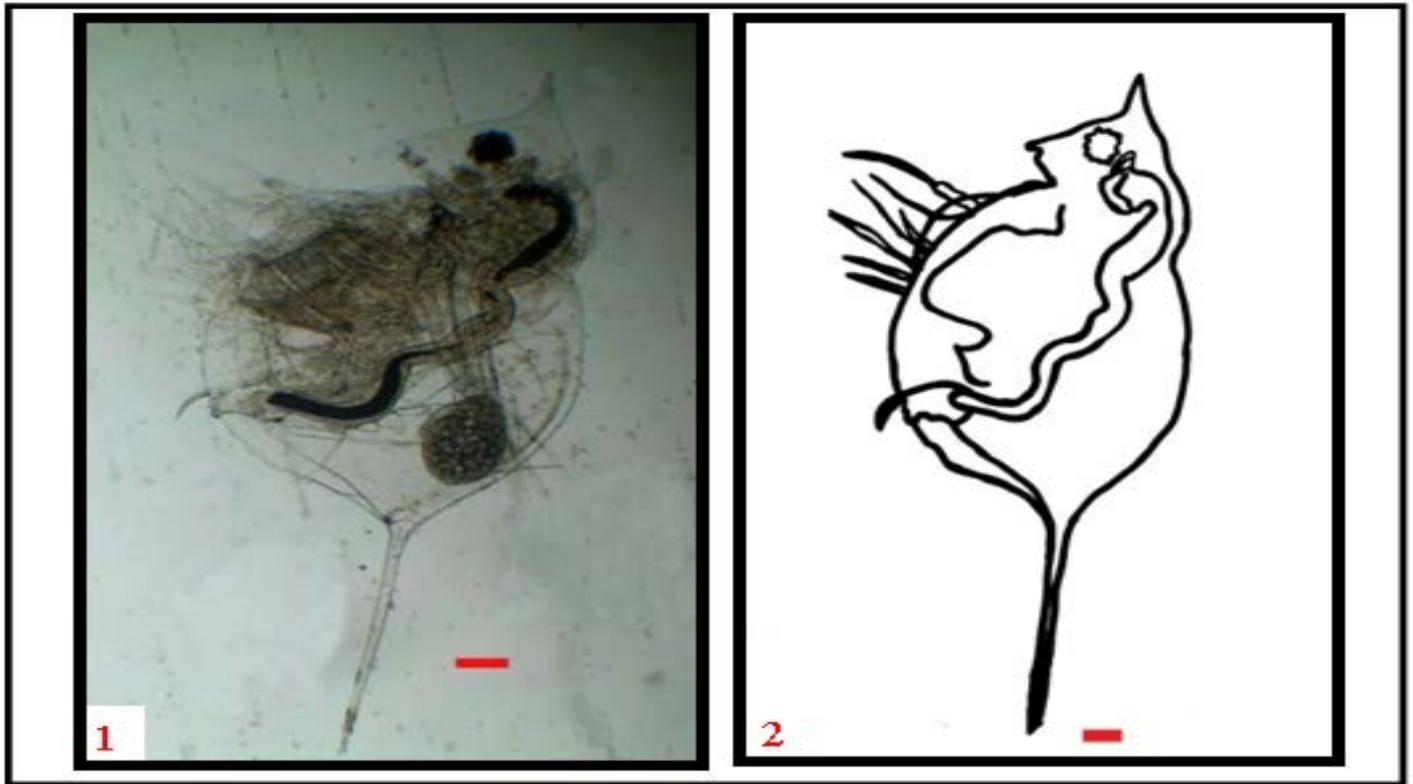


Figure 41: (1-2) *Daphnia magna*. Scale bar 50  $\mu\text{m}$  for all

### 8 - *Eubosmina longispina* (Leydig, 1860 )

#### Diagnosis :

The head shield have a pair of long, tusk-like antennules. Head pore is further from the margin of the head shield, located dorsal to the fornix reticulation fork. The pore can be obscured . The shell identified by the large spine or mucro on the posterior-ventral corner. The shell have small teeth on the ventral side of the mucro (Figure 42).

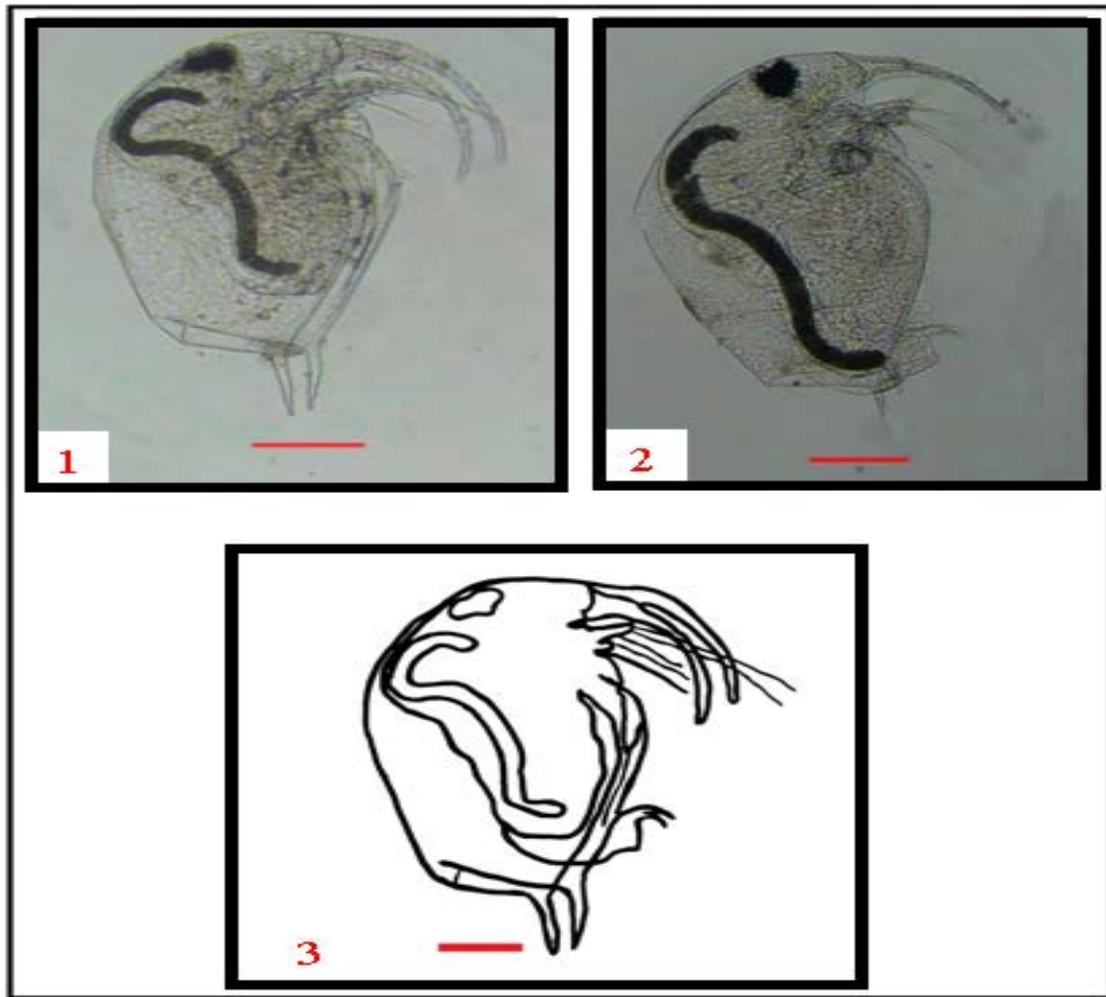


Figure 42: (1-3) *Eubosmina longispina*. Scale bar 50  $\mu\text{m}$

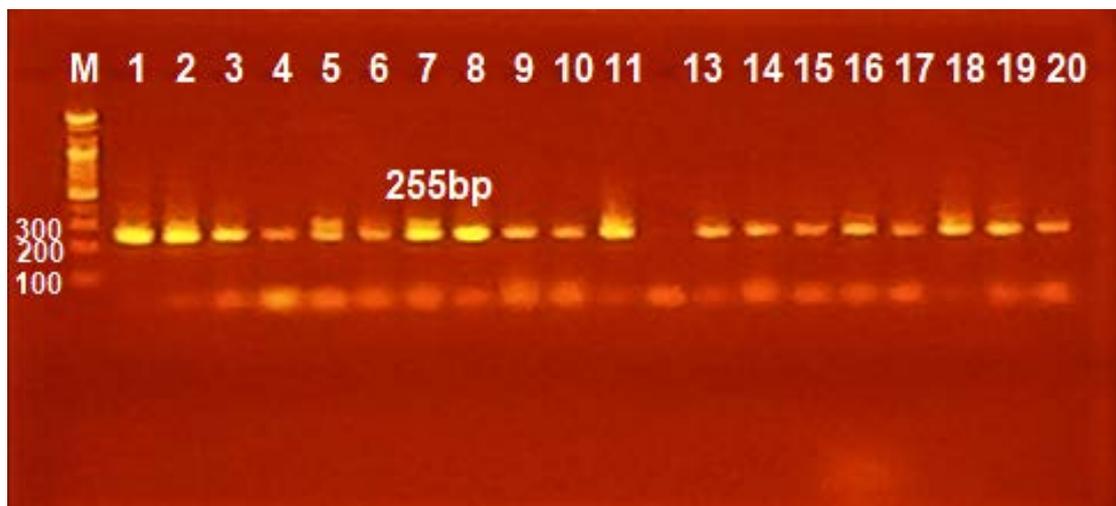
### 3 – 9: Molecular Identification of the Rotifera and Cladocera

#### 3 – 9-1: Extraction of Total DNA from the samples

By using specific Total DNA extraction kit (Cetyl trimethyl ammonium bromide (CTAB) Kit) the genomic DNA is extracted, the extracted DNA is checked for the presence of genomic DNA by nanodrop and the concentration is ranged between 5-10 ng/ $\mu\text{l}$  with purity ranged from 1.5 to 1.7, the low concentration of extracted DNA did not give a band during electrophoresis before PCR so it is directly applied for amplification process.

### 3 – 9-2 : Genotyping of Rotifera and Cladocera Gene

For rotifer and cladocera genotyping, the genomic DNA is amplified using specific primers and accomplished by the Thermo-cycler apparatus under the optimal condition as mentioned in (table 1). The results revealed that the presence a single band (255 bp) and (393 bp) of the target sequence of gene in agarose gel for rotifer and cladocera respectively (Figures 43-44).



**Figure (43):** Agarose gel electrophoresis of an amplified product patterns of rotifers samples on agarose 1.5% for 1.5 hrs with 60 volt



**Figure (44):** Agarose gel electrophoresis of an amplified product patterns of cladocera samples on agarose 1.5% for 1.5 hrs with 60 volt

**3–9-3: Sequences Alignment Fragment of 18s ribosomal RNA Gene (255 bp) of rotifera and (393 bp ) of Cladocera**

According to the findings of the alignment results of the 255 bp samples, (16,9,16,12) species are identified at station one during the seasons of spring, summer, autumn, and winter respectively. In addition, the molecular validation which representing the species that appeared in both genotypic and phenotypic identification includes (4,6,3,4) species identified during the seasons of spring, summer, autumn and winter respectively. Comparing the degree of similarity between species identified by genotypically detection and those identified through phenotypic detection, the lowest degree of similarity (21%) is observed during the autumn, while the maximum degree (44%) is observed during the winter as well as the biggest difference between species is (122%) during the winter (table 14). Given that there a difference between phenotypically diagnosed species and genotypically diagnosed species overall.

In the station two, (12, 26, 17, 7) species are identified in the spring, summer, autumn, and winter. Additionally, (4,6,3,7) species are found during the spring, summer, autumn, and winter seasons according to the molecular validation with lowest percentage of similarity (29%) in summer and highest percentage (78%) in winter, while the autumn showing the largest percentage of difference (170%) (table 15).

Table (14): Phenotypic and Molecular identification of rotifera in station one from Spring 2021 to Winter 2022

Station and Season	Phenotypic detection	Molecular Validation	Percentage of Molecular Precise	Molecular detection	Percentage of Molecular Precise	Total No of Taxa according Phenotypic and Genotypic Identification
Station 1- Spring	<i>Bdelloidea</i>	<i>Cephalodella gibba</i>	24 %	<i>Bryceella stylata</i>	94 %	Phenotypic identification : 17 Genotypic Identification : 16 Total No : 33
	<i>Cephalodella gibba</i>	<i>Polyarthra dolichoptera</i>		<i>Ptygura beauchampi</i>		
	<i>Cephalodella sp.</i>	<i>Scaridium longicaudum</i>		<i>Lecane furcata</i>		
	<i>Collotheca spp.</i>	<i>Trichotria tetractis</i>		<i>Pleurotrocha sigmoidea</i>		
	<i>Colurella sp.</i>			<i>Polyarthra major</i>		
	<i>Euchlanis dilatata</i>			<i>Polyarthra remata</i>		
	<i>Horaella brehmi</i>			<i>Collotheca tenuilobata</i>		
	<i>Lecane elsa</i>			<i>Lepadella triptera</i>		
	<i>Lecane hamata</i>			<i>Lecane ludwigii</i>		
	<i>Lepadella patella</i>			<i>Colurella obtusa</i>		
	<i>Platyias quadricornis</i>			<i>Collotheca ornata</i>		
	<i>Polyarthra dolichoptera</i>			<i>Filinia terminalis</i>		
	<i>Rotaria spp</i>			<i>Brachionus calyciflorus</i>		
	<i>Scaridium longicaudum</i>			<i>Octotrocha speciosa</i>		
	<i>Trichocerca rattus</i>			<i>Brachionus plicatilis</i>		
	<i>Trichotria pocillum</i>			<i>Lecane nana</i>		
	<i>Trichotria tetractis</i>					
No of Taxa	17	4		16		
Station 1- Summer	<i>Ascomorpha saltans</i>	<i>Cephalodella gibba</i>	35 %	<i>Bryceella stylata</i>	53 %	Phenotypic identification : 17 Genotypic Identification : 9 Total No : 26
	<i>Bdelloidea</i>	<i>Lecane bulla</i>		<i>Acyclus inquietus</i>		
	<i>Brachionus forficula</i>	<i>Lecane nana</i>		<i>Polyarthra major</i>		
	<i>Brachionus rubens</i>	<i>Testudinella patina</i>		<i>Polyarthra remata</i>		
	<i>Cephalodella gibba</i>	<i>Testudinella reflexa</i>		<i>Lecane tenuiseta</i>		
	<i>Euchlanis dilatata</i>	<i>Trichotria tetractis</i>		<i>Scaridium longicaudum</i>		
	<i>Lecane bulla</i>			<i>Brachionus angularis</i>		
	<i>Lecane closterocerca</i>			<i>Polyarthra dolichoptera</i>		
	<i>Lecane lunaris</i>			<i>Brachionus quadridentatus</i>		
	<i>Lecane nana</i>					
	<i>Lecane quadridentata</i>					
	<i>Lecane spp.</i>					
	<i>Lepadella patella</i>					
	<i>Macrotrachela angusta</i>					
	<i>Testudinella patina</i>					
	<i>Testudinella reflexa</i>					
	<i>Trichotria tetractis</i>					
No of Taxa	17	6		9		

Station 1- Autumn	<i>Ascomorpha ovalis</i>	<i>Trichotria tetractis</i>	21 %	<i>Bryceella stylata</i>	114 %	Phenotypic identification : 14  Genotypic Identification : 16  Total No : 30
	<i>Ascomorpha saltans</i>	<i>Testudinella patina</i>		<i>Encentrum fluviatile</i>		
	<i>Bdelloidea</i>	<i>Ascomorpha ovalis</i>		<i>Polyarthra major</i>		
	<i>Brachionus spp</i>			<i>Polyarthra remata</i>		
	<i>Brachionus falcatus</i>			<i>Lecane inermis</i>		
	<i>Colurella adriatica</i>			<i>Sinantherina semibullata</i>		
	<i>Encentrum porsildi</i>			<i>Lecane ludwigii</i>		
	<i>Keratella tecta</i>			<i>Scardium longicaudum</i>		
	<i>Lecane bulla</i>			<i>Cephalodella gibba</i>		
	<i>Lecane spp.</i>			<i>Brachionus calyciflorus</i>		
	<i>Lepadella spp</i>			<i>Floscularia armata</i>		
	<i>Mytilina ventralis</i>			<i>Lecane nana</i>		
	<i>Testudinella patina</i>			<i>Polyarthra dolichoptera</i>		
	<i>Trichotria tetractis</i>			<i>Brachionus quadridentatus</i>		
		<i>Brachionus angularis</i>				
		<i>Filinia terminalis</i>				
No of Taxa	14	3		16		
Station 1- Winter	<i>Asplanchna priodonta</i>	<i>Polyarthra major</i>	44 %	<i>Bryceella stylata</i>	122 %	Phenotypic identification : 9  Genotypic Identification : 12  Total No : 21
	<i>Bdelloidea</i>	<i>Cephalodella gibba</i>		<i>Brachionus zahniseri</i>		
	<i>Brachionus angularis</i>	<i>Synchaeta sp</i>		<i>Polyarthra remata</i>		
	<i>Cephalodella gibba</i>	<i>Brachionus angularis</i>		<i>Filinia terminalis</i>		
	<i>Keratella tecta</i>			<i>Brachionus calyciflorus</i>		
	<i>Macrochaetus sericus</i>			<i>Brachionus diversicornis</i>		
	<i>Polyarthra major</i>			<i>Trichotria tetractis</i>		
	<i>Synchaeta lakowitziana</i>			<i>Testudinella patina</i>		
	<i>Synchaeta spp.</i>			<i>Testudinella reflexa</i>		
				<i>Resticula gelida</i>		
				<i>Polyarthra dolichoptera</i>		
				<i>Brachionus quadridentatus</i>		
No of Taxa	9	4		12		

Table (15): Phenotypic and Molecular identification of rotifera in station two from Spring 2021 to Winter 2022

Station and Season	Phenotypic detection	Molecular Validation	Percentage of Molecular Precise	Molecular detection	Percentage of Molecular Precise	Total No of Taxa according Phenotypic and Genotypic Identification
Station 2 - Spring	<i>Bdelloidea</i>	<i>Cephalodella gibba</i>	40 %	<i>Bryceella stylata</i>	120 %	Phenotypic identification : 10  Genotypic Identification : 12  Total No : 22
	<i>Cephalodella gibba</i>	<i>Trichotria tetractis</i>		<i>Ptygura pilula</i>		
	<i>Cephalodella sp.</i>	<i>Synchaeta spp.</i>		<i>Lecane furcata</i>		
	<i>Euchlanis dilatata</i>	<i>Keratella cochlearis</i>		<i>Encentrum fluviatile</i>		
	<i>Keratella cochlearis</i>			<i>Polyarthra remata</i>		
	<i>Keratella spp.</i>			<i>Lepadella triptera</i>		
	<i>Synchaeta spp.</i>			<i>Encentrum marinum</i>		
	<i>Trichocerca rattus</i>			<i>Scaridium longicaudum</i>		
	<i>Trichocerca spp.</i>			<i>Filinia terminalis</i>		
	<i>Trichotria tetractis</i>			<i>Testudinella patina</i>		
				<i>Testudinella elliptica</i>		
				<i>Polyarthra dolichoptera</i>		
No of Taxa	10	4		12		
Station 2- Summer	<i>Bdelloidea</i>	<i>Lecane unguolata</i>	29 %	<i>Bryceella stylata</i>	124 %	Phenotypic identification : 21  Genotypic Identification : 26  Total No : 47
	<i>Brachionus forficula</i>	<i>Lecane tenuiseta</i>		<i>Polyarthra major</i>		
	<i>Encentrum porsildi</i>	<i>Lecane ludwigii</i>		<i>Polyarthra remata</i>		
	<i>Euchlanis dilatata</i>	<i>Trichotria tetractis</i>		<i>Lecane inermis</i>		
	<i>Euchlanis spp</i>	<i>Testudinella patina</i>		<i>Synchaeta oblonga</i>		
	<i>Filinia opoliensis</i>	<i>Lecane nana</i>		<i>Sinantherina socialis</i>		
	<i>Keratella cochlearis</i>			<i>Lacinaroides coloniensis</i>		
	<i>Keratella tropica</i>			<i>Stephanoceros fimbriatus</i>		
	<i>Lecane bulla</i>			<i>Notholca squamula</i>		
	<i>Lecane hamata</i>			<i>Notholca liepetterseni</i>		
	<i>Lecane ludwigii</i>			<i>Colurella obtusa</i>		
	<i>Lecane lunaris</i>			<i>Cephalodella gibba</i>		
	<i>Lecane nana</i>			<i>Brachionus angularis</i>		
	<i>Lecane quadridentata</i>			<i>Brachionus calyciflorus</i>		
	<i>Lecane spp.</i>			<i>Floscularia bifida</i>		
	<i>Lecane tenuiseta</i>			<i>Synchaeta pectinata</i>		
	<i>Lecane unguolata</i>			<i>Trichocerca tenuior</i>		
	<i>Macrotrachela angusta</i>			<i>Testudinella clypeata</i>		
	<i>Platyias quadricornis</i>			<i>Brachionus plicatilis</i>		
	<i>Testudinella patina</i>			<i>Testudinella reflexa</i>		
	<i>Trichotria tetractis</i>			<i>Ptygura stygis</i>		
				<i>Proales daphnicola</i>		
				<i>Notommata tripus</i>		
		<i>Mytilina bisulcata</i>				
		<i>Polyarthra dolichoptera</i>				
		<i>Brachionus quadridentatus</i>				
No of Taxa	21	6		26		
Station 2- Autumn	<i>Bdelloidea</i>	<i>Trichotria tetractis</i>	30 %	<i>Bryceella stylata</i>	170 %	Phenotypic identification : 10  Genotypic Identification : 17  Total No : 27
	<i>Keratella cochlearis</i>	<i>Testudinella patina</i>		<i>Brachionus zahniseri</i>		
	<i>Keratella spp.</i>	<i>Keratella cochlearis</i>		<i>Polyarthra major</i>		
	<i>Keratella tropica</i>			<i>Polyarthra remata</i>		
	<i>Lecane bulla</i>			<i>Lecane inermis</i>		
	<i>Lecane closterocerca</i>			<i>Lecane monostyla</i>		
	<i>Lecane thienemanni</i>			<i>Notholca foliacea</i>		
	<i>Trichocerca rattus</i>			<i>Lecane tenuiseta</i>		
	<i>Testudinella patina</i>			<i>Scaridium longicaudum</i>		
	<i>Trichotria tetractis</i>			<i>Collotheca ornata</i>		
				<i>Cephalodella gibba</i>		
				<i>Brachionus angularis</i>		
				<i>Brachionus calyciflorus</i>		
		<i>Lecane nana</i>				
		<i>Harringia eupoda</i>				
		<i>Polyarthra dolichoptera</i>				

No of Taxa	10	3		<i>Brachionus quadridentatus</i>	17	
Station 2- Winter	<i>Brachionus calyciflorus</i>	<i>Brachionus calyciflorus</i>	78 %	<i>Bryceella stylata</i>	78 %	Phenotypic identification : 9 Genotypic Identification : 7  Total No : 16
	<i>Cephalodella gibba</i>	<i>Cephalodella gibba</i>		<i>Brachionus zahneri</i>		
	<i>Keratella quadrata</i>	<i>Polyarthra major</i>		<i>Lecane inermis</i>		
	<i>Keratella tecta</i>	<i>Scardium longicaudum</i>		<i>Lecane tenuiseta</i>		
	<i>Polyarthra major</i>	<i>Synchaeta spp.</i>		<i>Encentrum semiplicatum</i>		
	<i>Scardium longicaudum</i>	<i>Testudinella patina</i>		<i>Lecane nana</i>		
	<i>Synchaeta spp.</i>	<i>Trichotria tetractis</i>		<i>Keratella cochlearis</i>		
	<i>Testudinella patina</i>					
	<i>Trichotria tetractis</i>					
No of Taxa	9	7		7		

In the station three, (11, 8, 10, and 21) species are identified in the spring, summer, autumn, and winter. The molecular validation also revealed that (3,5,1,4) species are discovered in the spring, summer, autumn, and winter seasons. The autumn season had the lowest percentage of similarity (17%) and the winter season had the highest percentage (44%) and is distinguished by the highest percentage of difference (233%), as well as the greatest number of species awareness (table 16).

In the station four, (11, 19, 16, 11) species are found in spring, summer, autumn, and winter. As well as , (1,6,4,9) species are found in the spring, summer, autumn, and winter seasons, according to the molecular validation. The percentage of similarity is lowest in the spring (17%) and highest in the winter (60%) while the percentage of difference is highest in the spring (183%) and the most species are found in the summer (table 17).

In the station five,(8,15, 11, 11) species are discovered in the spring, summer, autumn, and winter. Additionally, according to the molecular

validation, (7,6,3,2) species are discovered during the spring, summer, autumn, and winter seasons. While the proportion of differences is highest in the winter (183%), and the most species are detected in the summer, the percentage of similarity is lowest in the winter (33%) and highest in the spring (50%) (table 18).

**Table (16): Phenotypic and Molecular identification of rotifera in station three from Spring 2021 to Winter 2022**

Station and Season	Phenotypic detection	Molecular Validation	Percentage of Molecular Precise	Molecular detection	Percentage of Molecular Precise	Total No of Taxa according Phenotypic and Genotypic Identification
Station 3 - Spring	<i>Bdelloidea</i>	<i>Cephalodella gibba</i>	38 %	<i>Bryceella stylata</i>	138 %	Phenotypic identification : 8 Genotypic Identification : 11 Total No : 19
	<i>Cephalodella gibba</i>	<i>Polyarthra dolichoptera</i>		<i>Brachionus zahniseri</i>		
	<i>Euchlanis dilatata</i>	<i>Polyarthra remata</i>		<i>Polyarthra major</i>		
	<i>Lecane bulla</i>			<i>Lecane inermis</i>		
	<i>Macrochaetus spp</i>			<i>Lecane ludwigii</i>		
	<i>Polyarthra dolichoptera</i>			<i>Brachionus calyciflorus</i>		
	<i>Polyarthra remata</i>			<i>Pentatrocha gigantea</i>		
	<i>Trichocerca rattus</i>			<i>Lecane nana</i>		
				<i>Testudinella reflexa</i>		
				<i>Trichotria tetractis</i>		
		<i>Notommata cerberus</i>				
No of Taxa	8	3		11		
Station 3- Summer	<i>Bdelloidea</i>	<i>Brachionus calyciflorus</i>	25 %	<i>Bryceella stylata</i>	40 %	Phenotypic identification : 20 Genotypic Identification : 8 Total No : 28
	<i>Brachionus forficula</i>	<i>Cephalodella gibba</i>		<i>Brachionus zahniseri</i>		
	<i>Brachionus calyciflorus</i>	<i>Lecane unguolata</i>		<i>Polyarthra major</i>		
	<i>Brachionus falcatus</i>	<i>Testudinella patina</i>		<i>Lecane tenuiseta</i>		
	<i>Cephalodella gibba</i>	<i>Trichotria tetractis</i>		<i>Keratella cochlearis</i>		
	<i>Dicranophorus prionacis</i>			<i>Notholca acuminata</i>		
	<i>Enicentrum porsildi</i>			<i>Resticula anceps</i>		
	<i>Euchlanis dilatata</i>			<i>Dicranophorus luetkeni</i>		
	<i>Filinia opoliensis</i>					
	<i>Keratella tropica</i>					
	<i>Lecane arcuata</i>					
	<i>Lecane luna</i>					
	<i>Lecane lunaris</i>					
	<i>Lecane quadridentata</i>					
	<i>Lecane unguolata</i>					
	<i>Platyias quadricornis</i>					
	<i>Polyarthra minor</i>					
	<i>Trichocerca similis</i>					
<i>Testudinella patina</i>						
<i>Trichotria tetractis</i>						
No of Taxa	20	5		8		
Station 3- Autumn	<i>Bdelloidea</i>	<i>Polyarthra dolichoptera</i>	17 %	<i>Bryceella stylata</i>	167 %	Phenotypic identification : 6 Genotypic Identification : 10 Total No : 16
	<i>Collotheca spp.</i>			<i>Polyarthra euryptera</i>		
	<i>Filinia opoliensis</i>			<i>Polyarthra major</i>		
	<i>Lecane hamata</i>			<i>Collotheca tenuilobata</i>		
	<i>Lecane spp.</i>			<i>Collotheca campanulata</i>		
	<i>Polyarthra dolichoptera</i>			<i>Cephalodella gibba</i>		
				<i>Trichotria tetractis</i>		
				<i>Testudinella patina</i>		
		<i>Brachionus calyciflorus</i>				

No of Taxa	6	1		<i>Keratella cochlearis</i>	10	
Station 3- Winter	<i>Bdelloidea</i>	<i>Polyarthra major</i>	44 %	<i>Bryceella stylata</i>	233 %	Phenotypic identification : 9 Genotypic Identification : 21 Total No : 30
	<i>Colurella adriatica</i>	<i>Polyarthra remata</i>		<i>Brachionus zahniseri</i>		
	<i>Encentrum porsildi</i>	<i>Synchaeta sp.</i>		<i>Lacinularia flosculosa</i>		
	<i>Keratella tecta</i>	<i>Trichotria tetractis</i>		<i>Collotheca tenuilobata</i>		
	<i>Polyarthra major</i>			<i>Encentrum marinum</i>		
	<i>Polyarthra remata</i>			<i>Ptygura longicornis</i>		
	<i>Synchaeta spp.</i>			<i>Testudinella reflexa</i>		
	<i>Testudinella patina</i>			<i>Kellicottia bostoniensis</i>		
	<i>Trichotria tetractis</i>			<i>Encentrum semiplicatum</i>		
				<i>Collotheca pelagica</i>		
				<i>Collotheca mutabilis</i>		
				<i>Polyarthra vulgaris</i>		
				<i>Polyarthra dolichoptera</i>		
		<i>Proales similis</i>				
		<i>Notommata allantois</i>				
		<i>Ascomorpha ovalis</i>				
		<i>Filinia longiseta</i>				
		<i>Floscularia decora</i>				
		<i>Asplanchna brightwellii</i>				
		<i>Synchaeta tremula</i>				
		<i>Brachionus quadridentatus</i>				
No of Taxa	9	4		21		

Table (17): Phenotypic and Molecular identification of rotifera and in station four from Spring 2021 to Winter 2022

Station and Season	Phenotypic detection	Molecular Validation	Percentage of Molecular Precise	Molecular detection	Percentage of Molecular Precise	Total No of Taxa according Phenotypic and Genotypic Identification
Station 4 - Spring	<i>Bdelloidea</i>	<i>Cephalodella gibba</i>	17 %	<i>Bryceella stylata</i>	183 %	Phenotypic identification : 6 Genotypic Identification : 11 Total No : 17
	<i>Cephalodella gibba</i>			<i>Polyarthra major</i>		
	<i>Collotheca spp.</i>			<i>Polyarthra remata</i>		
	<i>Euchlanis dilatata</i>			<i>Lecane inermis</i>		
	<i>Lecane hamata</i>			<i>Synchaeta oblonga</i>		
	<i>Trichocerca rattus</i>			<i>Lecane ludwigii</i>		
				<i>Scaridium longicaudum</i>		
				<i>Lecane nana</i>		
				<i>Testudinella reflexa</i>		
				<i>Brachionus calyciflorus</i>		
		<i>Keratella cochlearis</i>				
No of Taxa	6	1		11		
Station 4- Summer	<i>Bdelloidea</i>	<i>Lecane unguolata</i>	40 %	<i>Bryceella stylata</i>	127 %	Phenotypic identification : 15 Genotypic Identification : 19 Total No : 34
	<i>Brachionus forficula</i>	<i>Cephalodella gibba</i>		<i>Polyarthra major</i>		
	<i>Cephalodella gibba</i>	<i>Lecane bulla</i>		<i>Sinanotherina socialis</i>		
	<i>Euchlanis dilatata</i>	<i>Trichotria tetractis</i>		<i>Lecane monostyla</i>		
	<i>Keratella tecta</i>	<i>Testudinella patina</i>		<i>Notholca squamula</i>		
	<i>Lecane bulla</i>	<i>Polyarthra dolichoptera</i>		<i>Lecane tenuiseta</i>		
	<i>Lecane elsa</i>			<i>Collotheca campanulata</i>		
	<i>Lecane spp.</i>			<i>Proales reinhardti</i>		
	<i>Lecane unguolata</i>			<i>Brachionus angularis</i>		
	<i>Macrotrachela angusta</i>			<i>Filinia terminalis</i>		
	<i>Platyias quadricornis</i>			<i>Floscularia bifida</i>		
	<i>Polyarthra dolichoptera</i>			<i>Synchaeta pectinata</i>		
	<i>Trichocerca rattus</i>			<i>Trichocerca tenuior</i>		
	<i>Testudinella patina</i>			<i>Testudinella clypeata</i>		

	<i>Trichotria tetractis</i>			<i>Lecane nana</i>		
				<i>Testudinella reflexa</i>		
				<i>Testudinella elliptica</i>		
				<i>Keratella cochlearis</i>		
				<i>Brachionus quadridentatus</i>		
No of Taxa	15	6		19		
Station 4- Autumn	<i>Bdelloidea</i>	<i>Filinia terminalis</i>	36 %	<i>Bryceella stylata</i>	145 %	Phenotypic identification : 11 Genotypic Identification : 16 Total No : 27
	<i>Filinia opoliensis</i>	<i>Trichotria tetractis</i>		<i>Brachionus zahniseri</i>		
	<i>Filinia terminalis</i>	<i>Testudinella patina</i>		<i>Encentrum fluviatile</i>		
	<i>Keratella cochlearis</i>	<i>Polyarthra dolichoptera</i>		<i>Polyarthra major</i>		
	<i>Keratella valga</i>			<i>Lecane inermis</i>		
	<i>Lecane bulla</i>			<i>Sinantharina semibullata</i>		
	<i>Platyias quadricornis</i>			<i>Lepadella triptera</i>		
	<i>Polyarthra dolichoptera</i>			<i>Lecane ludwigii</i>		
	<i>Trichocerca rattus</i>			<i>Colurella obtusa</i>		
	<i>Testudinella patina</i>			<i>Cephalodella gibba</i>		
	<i>Trichotria tetractis</i>			<i>Brachionus angularis</i>		
				<i>Brachionus calyciflorus</i>		
				<i>Synchaeta pectinata</i>		
		<i>Testudinella clypeata</i>				
		<i>Brachionus plicatilis</i>				
		<i>Lecane nana</i>				
No of Taxa	11	4		16		
Station 4- Winter	<i>Bdelloidea</i>	<i>Brachionus calyciflorus</i>	60 %	<i>Bryceella stylata</i>	73 %	Phenotypic identification : 15 Genotypic Identification : 11 Total No : 26
	<i>Brachionus calyciflorus</i>	<i>Cephalodella gibba</i>		<i>Polyarthra euryptera</i>		
	<i>Cephalodella gibba</i>	<i>Keratella cochlearis</i>		<i>Polyarthra remata</i>		
	<i>Colurella adriatica</i>	<i>Lecane bulla</i>		<i>Lecane inermis</i>		
	<i>Keratella cochlearis</i>	<i>Polyarthra dolichoptera</i>		<i>Lecane ludwigii</i>		
	<i>Lecane bulla</i>	<i>Polyarthra major</i>		<i>Filinia terminalis</i>		
	<i>Lecane closterocerca</i>	<i>Synchaeta spp.</i>		<i>Hexarthra intermedia</i>		
	<i>Lecane lunaris</i>	<i>Testudinella patina</i>		<i>Trichocerca tenuior</i>		
	<i>Lecane spp.</i>	<i>Trichotria tetractis</i>		<i>Brachionus plicatilis</i>		
	<i>Lepadella spp</i>			<i>Testudinella reflexa</i>		
	<i>Polyarthra dolichoptera</i>			<i>Brachionus quadridentatus</i>		
	<i>Polyarthra major</i>					
	<i>Synchaeta spp.</i>					
	<i>Testudinella patina</i>					
<i>Trichotria tetractis</i>						
No of Taxa	15	9		11		

Table (18): Phenotypic and Molecular identification of rotifera in station five from Spring 2021 to Winter 2022

Station and Season	Phenotypic detection	Molecular Validation	Percentage of Molecular Precise	Molecular detection	Percentage of Molecular Precise	Total No of Taxa according Phenotypic and Genotypic Identification
Station 5 - Spring	<i>Bdelloidea</i>	<i>Polyarthra major</i>	50 %	<i>Bryceella stylata</i>	57 %	Phenotypic identification : 14 Genotypic Identification : 8 Total No : 22
	<i>Brachionus calyciflorus</i>	<i>Lecane bulla</i>		<i>Polyarthra remata</i>		
	<i>Brachionus quadridentatus</i>	<i>Testudinella patina</i>		<i>Lecane inermis</i>		
	<i>Euchlanis dilatata</i>	<i>Brachionus quadridentatus</i>		<i>Sinantharina socialis</i>		
	<i>Horaella brehmi</i>	<i>Synchaeta sp.</i>		<i>Cephalodella gibba</i>		
	<i>Keratella cochlearis</i>	<i>Brachionus calyciflorus</i>		<i>Brachionus angularis</i>		
	<i>Keratella tecta</i>	<i>Keratella cochlearis</i>		<i>Trichotria tetractis</i>		

	<i>Lecane bulla</i>			<i>Polyarthra dolichoptera</i>		
	<i>Lecane closteroerca</i>					
	<i>Lecane lunaris</i>					
	<i>Polyarthra major</i>					
	<i>Synchaeta spp.</i>					
	<i>Trichocerca rattus</i>					
	<i>Testudinella patina</i>					
No of Taxa	14	7		8		
Station 5- Summer	<i>Bdelloidea</i>	<i>Polyarthra major</i>	35 %	<i>Bryceella stylata</i>	88 %	Phenotypic identification : 17 Genotypic Identification : 15 Total No : 32
	<i>Brachionus forficula</i>	<i>Brachionus angularis</i>		<i>Brachionus zahniseri</i>		
	<i>Brachionus rubens</i>	<i>Brachionus calyciflorus</i>		<i>Polyarthra remata</i>		
	<i>Brachionus angularis</i>	<i>Trichotria tetractis</i>		<i>Lecane ungulata</i>		
	<i>Brachionus budapestinensis</i>	<i>Testudinella patina</i>		<i>Notholca squamula</i>		
	<i>Brachionus calyciflorus</i>	<i>Lecane nana</i>		<i>Dicranophorus luetkeni</i>		
	<i>Dicranophorus prionacis</i>			<i>Colurella obtusa</i>		
	<i>Euchlanis dilatata</i>			<i>Brachionus diversicornis</i>		
	<i>Filinia opoliensis</i>			<i>Cephalodella gibba</i>		
	<i>Lecane bulla</i>			<i>Hexarthra intermedia</i>		
	<i>Lecane elsa</i>			<i>Floscularia armata</i>		
	<i>Lecane nana</i>			<i>Notommata glyphura</i>		
	<i>Platylas quadricornis</i>			<i>Mytilina brevispina</i>		
	<i>Polyarthra major</i>			<i>Polyarthra dolichoptera</i>		
	<i>Trichocerca rattus</i>			<i>Brachionus quadridentatus</i>		
	<i>Testudinella patina</i>					
	<i>Trichotria tetractis</i>					
No of Taxa	17	6		15		
Station 5- Autumn	<i>Bdelloidea</i>	<i>Lecane inermis</i>	38 %	<i>Bryceella stylata</i>	138 %	Phenotypic identification : 8 Genotypic Identification : 11 Total No : 19
	<i>Colurella adriatica</i>	<i>Testudinella patina</i>		<i>Lecane furcata</i>		
	<i>Dicranophorus prionacis</i>	<i>Synchaeta sp.</i>		<i>Polyarthra major</i>		
	<i>Encentrum porsildi</i>			<i>Polyarthra remata</i>		
	<i>Lecane inermis</i>			<i>Lecane ungulata</i>		
	<i>Synchaeta spp.</i>			<i>Lecane monostyla</i>		
	<i>Trichocerca rattus</i>			<i>Notholca liepetterseni</i>		
	<i>Testudinella patina</i>			<i>Cephalodella gibba</i>		
				<i>Brachionus plicatilis</i>		
				<i>Lecane nana</i>		
			<i>Polyarthra dolichoptera</i>			
No of Taxa	8	3		11		
Station 5- Winter	<i>Bdelloidea</i>	<i>Polyarthra major</i>	33 %	<i>Bryceella stylata</i>	183 %	Phenotypic identification : 6 Genotypic Identification : 11 Total No : 17
	<i>Lecane closteroerca</i>	<i>Testudinella patina</i>		<i>Polyarthra remata</i>		
	<i>Lecane elsa</i>			<i>Scaridium longicaudum</i>		
	<i>Polyarthra major</i>			<i>Cephalodella gibba</i>		
	<i>Polyarthra minor</i>			<i>Brachionus angularis</i>		
	<i>Testudinella patina</i>			<i>Brachionus calyciflorus</i>		
				<i>Trichotria tetractis</i>		
				<i>Mikrocodides chlaena</i>		
				<i>Notommata tripus</i>		
				<i>Lecane bulla</i>		
			<i>Polyarthra dolichoptera</i>			
No of Taxa	6	2		11		





*Polyarthra dolichoptera*

Query 14

GGATTGACAGATTGATAGCTCTTTCTCGATTCCGTGGGTGGTGGTGCATGG  
CCGTTCTTA 73

||||| ||||| ||| ||||||||||||||||

Sbjct 1147GGATTGACAGATTGAGAGCTCTTTCTTGATTCCGTGGGTGGTGGTGCATGG  
CCGTTCTTA 1206

Query 74

GTTGGTGGAGCGATTTGTCTGGTTAATTCCGATAACGAACGAGACTCTGG  
CATGCTAACT 133

||||| ||||| ||||| || ||||| |

Sbjct 1207GTTGGTGGAGCGATTTGTCTGGTTAATTCCGATAACGAACGAGACTCTAG  
CCTGCTAAAT 1266

Query 134 AGTTACGC-

GACCCCGAGCGGTCGGCGTCCCCAACTTCTTAGAGGGACAAGTGGCGT  
T 192

||| ||| || | ||| || ||| |||||||||||| ||| |

Sbjct 1267 AGT-ACGCCGATCTCTTGTCGG-CG-CGT-----

ACTTCTTAGAGGGACAAGTAGCGGT 1317

Query 193

CAGCCACCCGAGATTGAGCAATAACAGGTCTGTGATGCCCTTAG 236

||| ||| ||||||||||||||||||

Sbjct 1318

AAGCTACACGAAATTGAGCAATAACAGGTCTGTGATGCCCTTAG 1361

*Testudinella patina*

Query 14

GGATTGACAGATTGATAGCTCTTTCTCGATTCCGTGGGTGGTGGTGCATGG  
CCGTTCTTA 73

||||| ||||| ||| ||||||||||||||||

Sbjct 1180GGATTGACAGATTGAGAGCTCTTTCTTGATTCCGTGGGTGGTGGTGCATGG  
CCGTTCTTA 1239

Query 74

GTTGGTGGAGCGATTTGTCTGGTTAATTCCGATAACGAACGAGACTCTGG  
CATGCTAACT 133

||||| ||||| ||||| || ||||| |

Sbjct 1240GTTGGTGGAGCGATTTGTCTGGTTAATTCCGATAACGAACGAGACTCTAG  
CCTGCTAAAT 1299

Query 134 AGTTACGC-

GACCCCGAGCGGTCGGCGTCCCCAACTTCTTAGAGGGACAAGTGGCGT  
T 192

||| ||| || | ||| || ||| |||||||||||| ||| |

Sbjct 1300 AGT-ACGCCGATCTCTTGTCGG-CG-CGT-----

ACTTCTTAGAGGGACAAGTAGCGGT 1350

Query 193

CAGCCACCCGAGATTGAGCAATAACAGGTCTGTGATGCCCTTAG 236

||| ||| ||||||||||||||||||

Sbjct 1351

AAGCTACACGAAATTGAGCAATAACAGGTCTGTGATGCCCTTAG 1394

*Polyarthra remata*

Query 45

GATTCGGTGGGTGGTGGTGCATGGCCGTTCTTAGTTGGTGGAGCGATTTGT  
CTGGTTAAT 104

|||||

Sbjct 1242GATTCGGTGGGTGGTGGTGCATGGCCGTTCTTAGTTGGTGGAGCGATTTGT  
CTGGTTAAT 1301Query 105 TCCGATAACGAACGAGACTCTGGCATGCTAACTAGTTACGC-  
GACCCCGAGCGGTCGGC 163

|||||

Sbjct 1302 TCCGATAACGAACGAGACTCTAGCCTGCTAAATAGT-  
ACGCCGATCTCTTGTCCG-CG-C 1358

Query 164

GTCCCCAACTTCTTAGAGGGACAAGTGGCGTTCAGCCACCCGAGATTGA  
GCAATAACAG

223

|||

Sbjct 1359 GT-----ACTTCTTAGAGGGACAAGTGGCGGTAAGCCACACGAAATTGAGCAATAAC  
AG 1412

Query 224 GTCTGTGATGCCCTTAGA 241

|||||

Sbjct 1413 GTCTGTGATGCCCTTAGA 1430*Brachionus calyciflorus*

Query 13

GGTTGACAGAATTGATAGCTCTTTC TTGATTCCGTGGGTGGTGGTGCATG  
GCCGTTCTT 72

|||

Sbjct 1241 GGATTGACAG-ATTGAGAGCTCTTTC TTGATTCCGTTGGGTGGTGGTGCATGGCCGTTCTT  
1299

Query 73

AGTTGGTGGAGCGATTTGTCTGGTTAATTCCGATAACGAACGAGACTCTG  
GCCTGCTAAC 132

|||||

Sbjct 1300AGTTGGTGGAGCGATTTGTCTGGTTAATTCCGATAACGAACGAGACTCTA  
GCCTGCTAAA

1359

Query 133 TAGTTACGCCACCCCGAG-

CGGTCGGCGTCCCCCAACTTCTTAAAGGGACAAGTGGCGT 191

|||

Sbjct 1360 TAGT-ACGCCGATCTTTAGTCCG-CG-CGT-----

ACTTCTTAGAGGGACAAGTAGCGG 1410

Query 192

TCAGCCACCCGAGATTGAGCAATAACAGGTCTGTGATGCCCTTAGA 237

|||

Sbjct 1411

TAAGCTACACGAAATTGAGCAATAACAGGTCTGTGATGCCCTTAGA 1456



For the Cladocera , At station one, (16,2,2,11) are discovered in the spring, summer, autumn, and winter, respectively. Additionally, only one species identified during both the summer and the winter is included in the molecular validation. Comparing the degree of similarity between species discovered through genotypic detection and those discovered through phenotypic detection, the lowest degree of similarity (0%) is observed during summer and autumn, While the highest degree (50%) and the greatest difference between species (550%) are all discovered during the winter (table 19).

In Station 2, species from the spring, summer, autumn, and winter are recognized (15, 15, 3, 10). According to molecular validation, only 2 species are discovered during the spring, with the lowest proportion of similarity (0%) in summer, autumn and winter respectively and the maximum percentage (50%) is noticed in spring while largest percentage of difference (1500%) occurring in the summer (table 20).

In Station 3, (14,3,0,1) species are recognized during the spring, summer, autumn, and winter respectively. The molecular validation revealed no resemblance during the period of the investigation, with the spring showing the greatest difference (467%). (table 21).

Table (19): Phenotypic and Molecular identification of Cladocera in station one from Spring 2021 to Winter 2022

Station and Season	Phenotypic detection	Molecular Validation	Percentage of Molecular Precise	Molecular detection	Percentage of Molecular Precise	Total No of Taxa according Phenotypic and Genotypic Identification
Station 1- Spring	<i>Acroperus angustatus</i>	<i>Daphnia magna</i>	33 %	<i>Daphnia dubia</i>	533 %	Phenotypic identification : 3 Genotypic Identification : 16 Total No : 19
	<i>Daphnia magna</i>			<i>Daphnia laevis</i>		
	<i>Eubosmina longispina</i>			<i>Diaphanosoma brachyurum</i>		
				<i>Scapholeberis mucronata</i>		
				<i>Polyphemus pediculus</i>		
				<i>Leptodora kindtii</i>		
				<i>Cercopagis pengoi</i>		
				<i>Sida crystallina</i>		
				<i>Evadne nordmanni</i>		
				<i>Podon leuckartii</i>		
				<i>Bythotrephes longimanus</i>		
				<i>Macrothrix laticornis</i>		
				<i>Daphniopsis truncata</i>		
		<i>Daphnia longicephala</i>				
		<i>Daphnia ambigua</i>				
		<i>Daphnia occidentalis</i>				
No of Taxa	3	1		16		
Station 1- Summer	<i>Acantholeberis curvirostris</i>		0 %	<i>Daphnia pulicaria</i>	200 %	Phenotypic identification : 1 Genotypic Identification : 2 Total No : 3
				<i>Sida crystallina</i>		
No of Taxa	1	0		2		
Station 1- Autumn	<i>Eubosmina longispina</i>		0 %	<i>Daphnia pulicaria</i>	200 %	Phenotypic identification : 1 Genotypic Identification : 2 Total No : 3
				<i>Simocephalus vetulus</i>		
No of Taxa	1	0		2		
Station 1- Winter	<i>Daphnia magna</i>	<i>Daphnia magna</i>	50 %	<i>Daphnia dubia</i>	550 %	Phenotypic identification : 2 Genotypic Identification : 11 Total No : 13
	<i>Eubosmina longispina</i>			<i>Daphnia laevis</i>		
				<i>Diaphanosoma brachyurum</i>		
				<i>Scapholeberis mucronata</i>		
				<i>Polyphemus pediculus</i>		
				<i>Leptodora kindtii</i>		
				<i>Cercopagis pengoi</i>		
				<i>Sida crystallina</i>		
				<i>Bythotrephes longimanus</i>		
				<i>Macrothrix laticornis</i>		
				<i>Daphnia ambigua</i>		
No of Taxa	2	1		11		

Table (20): Phenotypic and Molecular identification of Cladocera in station two from Spring 2021 to Winter 2022

Station and Season	Phenotypic detection	Molecular Validation	Percentage of Molecular Precise	Molecular detection	Percentage of Molecular Precise	Total No of Taxa according Phenotypic and Genotypic Identification
Station 2 - Spring	<i>Acantholeberis curvirostris</i>	<i>Acantholeberis curvirostris</i>	50 %	<i>Daphnia dubia</i>	375 %	Phenotypic identification : 4 Genotypic Identification : 15 Total No : 19
	<i>Camptocercus spp</i>	<i>Daphnia magna</i>		<i>Daphnia laevis</i>		
	<i>Daphnia magna</i>			<i>Diaphanosoma brachyurum</i>		
	<i>Eubosmina longispina</i>			<i>Scapholeberis mucronata</i>		
				<i>Polyphemus pediculus</i>		
				<i>Leptodora kindtii</i>		
				<i>Cornigerius maeoticus</i>		
				<i>Sida crystallina</i>		
				<i>Evadne spinifera</i>		
				<i>Podon leuckartii</i>		
				<i>Bythotrephes longimanus</i>		
				<i>Streblocerus serricaudatus</i>		
				<i>Daphniopsis truncata</i>		
		<i>Daphnia longicephala</i>				
		<i>Daphnia ambigua</i>				
No of Taxa	4	2		15		
Station 2- Summer	<i>Acantholeberis curvirostris</i>		0 %	<i>Daphnia dubia</i>	1500 %	Phenotypic identification : 1 Genotypic Identification : 15 Total No : 16
				<i>Daphnia laevis</i>		
				<i>Daphnia magna</i>		
				<i>Scapholeberis mucronata</i>		
				<i>Polyphemus pediculus</i>		
				<i>Leptodora kindtii</i>		
				<i>Cornigerius maeoticus</i>		
				<i>Sida crystallina</i>		
				<i>Evadne spinifera</i>		
				<i>Evadne nordmanni</i>		
				<i>Ophryoxus gracilis</i>		
				<i>Daphniopsis truncata</i>		
				<i>Daphnia longicephala</i>		
		<i>Daphnia ambigua</i>				
		<i>Daphnia occidentalis</i>				
No of Taxa	1	0		15		
Station 2- Autumn	<i>Acantholeberis curvirostris</i>		0 %	<i>Daphnia dubia</i>	150 %	Phenotypic identification : 2 Genotypic Identification : 3 Total No : 5
	<i>Eubosmina longispina</i>			<i>Daphnia laevis</i>		
				<i>Diaphanosoma brachyurum</i>		
No of Taxa	2	0		3		
Station 2- Winter	<i>Acantholeberis curvirostris</i>		0 %	<i>Penilia avirostris</i>	500 %	Phenotypic identification : 2 Genotypic Identification : 10 Total No : 12
	<i>Eubosmina longispina</i>			<i>Eurycerus lamellatus</i>		
				<i>Alona affinis</i>		
				<i>Rynchotalona falcata</i>		
				<i>Drepanothrix dentata</i>		
				<i>Lathonura rectirostris</i>		
				<i>Macrothrix laticornis</i>		
				<i>Bosmina longirostris</i>		
				<i>Chydorus sphaericus</i>		

No of Taxa	2	0		<i>Eubosmina spp</i>		
				10		

**Table (21): Phenotypic and Molecular identification of Cladocera in station three from Spring 2021 to Winter 2022**

Station and Season	Phenotypic detection	Molecular Validation	Percentage of Molecular Precise	Molecular detection	Percentage of Molecular Precise	Total No of Taxa according Phenotypic and Genotypic Identification
Station 3 - Spring	<i>Acroperus angustatus</i>		0 %	<i>Daphnia dubia</i>	467 %	Phenotypic identification : 3 Genotypic Identification : 14 Total No : 17
	<i>Camptocercus spp</i>			<i>Diaphanosoma brachyurum</i>		
	<i>Eubosmina longispina</i>			<i>Daphnia magna</i>		
				<i>Scapholeberis mucronata</i>		
				<i>Polyphemus pediculus</i>		
				<i>Cornigerius maeoticus</i>		
				<i>Sida crystalline</i>		
				<i>Evadne spinifera</i>		
				<i>Podon leuckartii</i>		
				<i>Bythotrephes longimanus</i>		
				<i>Daphniopsis queenslandensis</i>		
				<i>Daphniopsis truncata</i>		
				<i>Daphnia longicephala</i>		
				<i>Daphnia ambigua</i>		
No of Taxa	3	0		14		
Station 3- Summer	<i>Acantholeberis curvirostris</i>		0 %	<i>Daphnia dubia</i>	150 %	Phenotypic identification : 2 Genotypic Identification : 3 Total No : 5
	<i>Eubosmina longispina</i>			<i>Daphnia laevis</i>		
				<i>Diaphanosoma brachyurum</i>		
No of Taxa	2	0		3		
Station 3- Autumn	<i>Acroperus angustatus</i>		0 %	No significant similarity found.	0 %	Phenotypic identification : 2 Genotypic Identification : 0 Total No : 2
	<i>Eubosmina longispina</i>					
No of Taxa	2	0		0		
Station 3- Winter	<i>Acantholeberis curvirostris</i>		0 %	<i>Daphnia pulicaria</i>	50 %	Phenotypic identification : 2 Genotypic Identification : 1 Total No : 3
	<i>Acroperus angustatus</i>					
No of Taxa	2	0		1		

In Station 4, (14, 5, 7, 7) species are discovered in the spring, summer, autumn and winter. A single species is discovered in the spring according to molecular validation. The spring exhibited low similarity (33%) although the proportion of difference is also largest in the spring (467%) and the most species are detected in the summer. There is no similarity in the summer, autumn, or winter (table 22).

In Station 5, the spring, summer, autumn and winter seasons led to the discovery of (1, 14, 14, 8) species. Molecular validation indicates that in the summer, a single species is found. Low similarity (25%) is seen in the summer, whereas the biggest difference (467%) is seen in the autumn (table 23 ).

**Table (22): Phenotypic and Molecular identification of Cladocera in station four from Spring 2021 to Winter 2022**

Station and Season	Phenotypic detection	Molecular Validation	Percentage of Molecular Precise	Molecular detection	Percentage of Molecular Precise	Total No of Taxa according Phenotypic and Genotypic Identification
Station 4 - Spring	<i>Acroperus angustatus</i>	<i>Daphnia magna</i>	33 %	<i>Daphnia laevis</i>	467 %	Phenotypic identification : 3 Genotypic Identification : 14 Total No : 17
	<i>Daphnia magna</i>			<i>Diaphanosoma brachyurum</i>		
	<i>Eubosmina longispina</i>			<i>Scapholeberis mucronata</i>		
				<i>Polyphemus pediculus</i>		
				<i>Leptodora kindtii</i>		
				<i>Cercopagis pengoi</i>		
				<i>Evadne nordmanni</i>		
				<i>Podon leuckartii</i>		
				<i>Ophryoxus gracilis</i>		
				<i>Daphniopsis truncata</i>		
				<i>Daphnia longicephala</i>		
				<i>Daphnia ambigua</i>		
				<i>Daphnia occidentalis</i>		
				<i>Daphnia pulicaria</i>		
No of Taxa	3	1		14		
Station 4- Summer	<i>Alona guttata</i>		0 %	<i>Daphnia dubia</i>	250 %	Phenotypic identification : 2 Genotypic Identification : 5 Total No : 7
	<i>Eubosmina longispina</i>			<i>Daphnia laevis</i>		
				<i>Diaphanosoma brachyurum</i>		
				<i>Sida crystallina</i>		
				<i>Daphnia pulicaria</i>		
No of Taxa	2	0		5		

Station 4- Autumn	<i>Alonella exigua</i>		0 %	<i>Daphnia pulicaria</i>	233 %	Phenotypic identification : 3  Genotypic Identification : 7  Total No : 10
	<i>Camptocercus uncinatus</i>			<i>Ceriodaphnia pulchella</i>		
	<i>Eubosmina longispina</i>			<i>Daphnia pulex</i>		
				<i>Ceriodaphnia rotunda</i>		
				<i>Simocephalus serrulatus</i>		
				<i>Daphniopsis ephemeralis</i>		
				<i>Daphnia dentifera</i>		
No of Taxa	3	0		7		
Station 4- Winter	<i>Daphnia magna</i>		0 %	<i>Daphnia dubia</i>	350 %	Phenotypic identification : 2  Genotypic Identification : 7  Total No : 9
	<i>Eubosmina longispina</i>			<i>Daphnia laevis</i>		
				<i>Diaphanosoma brachyurum</i>		
				<i>Sida crystallina</i>		
				<i>Streblocerus serricaudatus</i>		
				<i>Ophryoxus gracilis</i>		
				<i>Daphnia pulicaria</i>		
No of Taxa	2	0		7		

Table (23): Phenotypic and Molecular identification of Cladocera in station five from Spring 2021 to Winter 2022

Station and Season	Phenotypic detection	Molecular Validation	Percentage of Molecular Precise	Molecular detection	Percentage of Molecular Precise	Total No of Taxa according Phenotypic and Genotypic Identification
Station 5 - Spring	<i>Acantholeberis curvirostris</i>		0 %	<i>Daphnia pulicaria</i>	25 %	Phenotypic identification : 4 Genotypic Identification : 1 Total No : 5
	<i>Acroperus angustatus</i>					
	<i>Daphnia magna</i>					
	<i>Eubosmina longispina</i>					
No of Taxa	4	0		1		
Station 5- Summer	<i>Acantholeberis curvirostris</i>	<i>Daphnia magna</i>	25 %	<i>Daphnia dubia</i>	350 %	Phenotypic identification : 4  Genotypic Identification : 14  Total No : 18
	<i>Acroperus angustatus</i>			<i>Daphnia laevis</i>		
	<i>Camptocercus spp</i>			<i>Scapholeberis mucronata</i>		
	<i>Daphnia magna</i>			<i>Leptodora kindtii</i>		
				<i>Cornigerius maeoticus</i>		
				<i>Sida crystallina</i>		
				<i>Podon leuckartii</i>		
				<i>Polyphemus pediculus</i>		
				<i>Bythotrephes longimanus</i>		
				<i>Streblocerus serricaudatus</i>		
				<i>Ophryoxus gracilis</i>		
				<i>Daphniopsis queenslandensis</i>		
				<i>Daphnia longicephala</i>		
		<i>Daphnia ambigua</i>				
No of Taxa	4	1		14		

Station 5- Autumn	<i>Alona spp</i>		0 %	<i>Daphnia dubia</i>	467 %	Phenotypic identification : 3 Genotypic Identification : 14  Total No : 17
	<i>Anthalona verrucosa</i>			<i>Daphnia laevis</i>		
	<i>Eubosmina longispina</i>			<i>Diaphanosoma brachyurum</i>		
				<i>Daphnia magna</i>		
				<i>Scapholeberis mucronata</i>		
				<i>Polyphemus pediculus</i>		
				<i>Cornigerius maeoticus</i>		
				<i>Sida crystallina</i>		
				<i>Evadne spinifera</i>		
				<i>Podon leuckartii</i>		
				<i>Bythotrephes longimanus</i>		
				<i>Daphniopsis truncata</i>		
				<i>Daphnia ambigua</i>		
				<i>Daphnia occidentalis</i>		
No of Taxa	3	0		14		
Station 5- Winter	<i>Acroperus angustatus</i>		0 %	<i>Acantholeberis curvirostris</i>	267 %	Phenotypic identification : 3 Genotypic Identification : 8  Total No : 11
	<i>Alona spp</i>			<i>Bosmina longirostris</i>		
	<i>Eubosmina longispina</i>			<i>Eurycerus lamellatus</i>		
				<i>Penilia avirostris</i>		
				<i>Alona affinis</i>		
				<i>Drepanothrix dentata</i>		
				<i>Chydorus sphaericus</i>		
				<i>Lathonura rectirostris</i>		
No of Taxa	3	0		8		

According to the molecular identification and sequencing alignment, Al-Hilla River has (40) species during the study period and the diagnostic of these species revealed that *Daphnia dubia* , *Daphnia laevis*, *Daphnia ambigua*, *Daphnia pulicaria*, *Diaphanosoma brachyurum*, *Polyphemus pediculus*, *Scapholeberis mucronata* and *Sida crystallina* are the most prevalent in most stations. Additionally, the current study revealed the first-time diagnoses of species (*Daphnia dubia*, *Daphnia laevis*, *Daphnia ambigua*, *Daphnia pulicaria*, *Daphnia longicephala* , *Polyphemus pediculus* and *Acantholeberis curvirostris* ) in Iraq within the Hilla River.

Finally, In the current study, five rotifer species and seven cladocera species are discovered for the first time in Iraq in Al-Hilla River and their sequences are entered into NCBI (table 24).

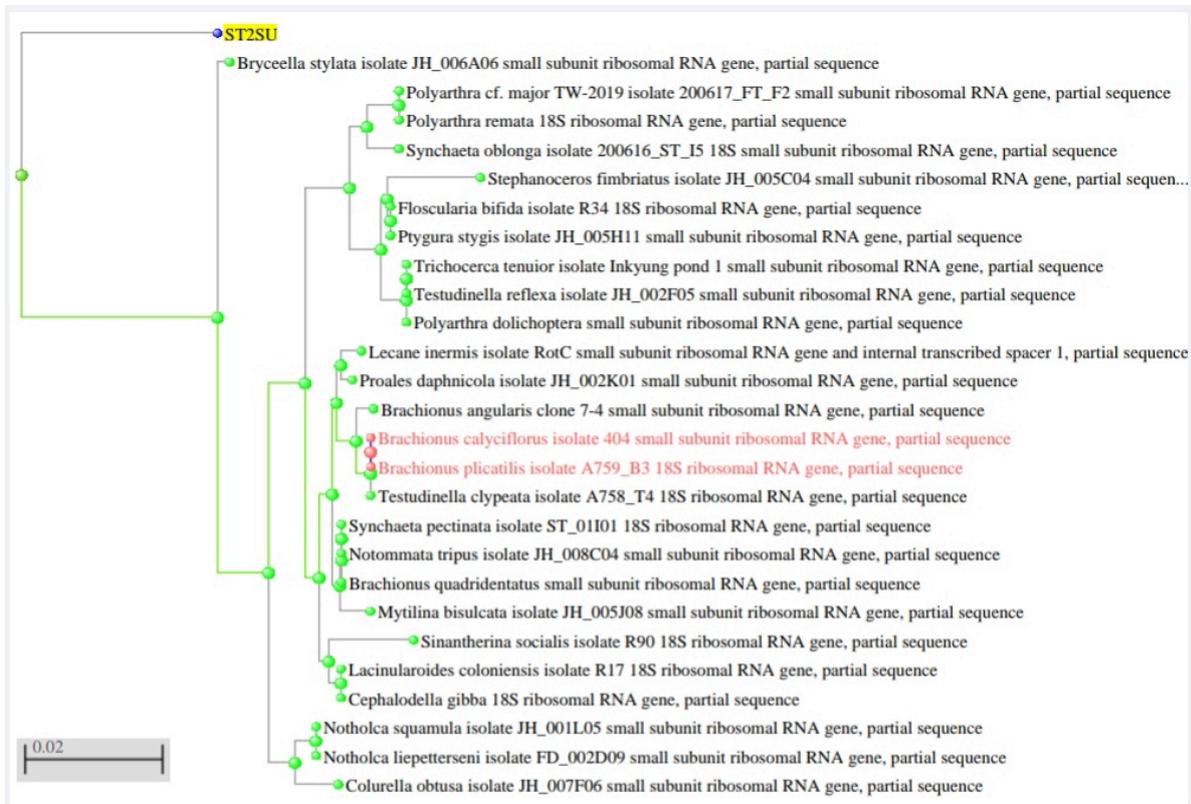
Table (24): New record species of the current study with Accession number in NCBI

New record species	Accession number in NCBI of the studied sample of 18s gene	Website Link
<i>Bryceella stylata</i>	LC739677.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739677.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739677.1/</a>
<i>Ascomorpha ovalis</i>	LC739681.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739681.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739681.1/</a>
<i>Lecane tenuista</i>	LC739679.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739679.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739679.1/</a>
<i>Lecane ungulata</i>	LC739680.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739680.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739680.1/</a>
<i>Scaridium longicaudum</i>	LC739678	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739678/">https://www.ncbi.nlm.nih.gov/nuccore/LC739678/</a>
<i>Daphnia dubia</i>	LC739683.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739683.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739683.1/</a>
<i>Daphnia laevis</i>	LC739684.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739684.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739684.1/</a>
<i>Daphnia ambigua</i>	LC739686.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739686.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739686.1/</a>
<i>Daphnia pulicaria</i>	LC739687.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739687.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739687.1/</a>
<i>Daphnia longicephala</i>	LC739685.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739685.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739685.1/</a>
<i>Polyphemus pediculus</i>	LC739688.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739688.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739688.1/</a>
<i>Acantholeberis curvirostris</i>	LC739682.1	<a href="https://www.ncbi.nlm.nih.gov/nuccore/LC739682.1/">https://www.ncbi.nlm.nih.gov/nuccore/LC739682.1/</a>

### 3 – 9-4 : Phylogenetic Tree of Zooplankton

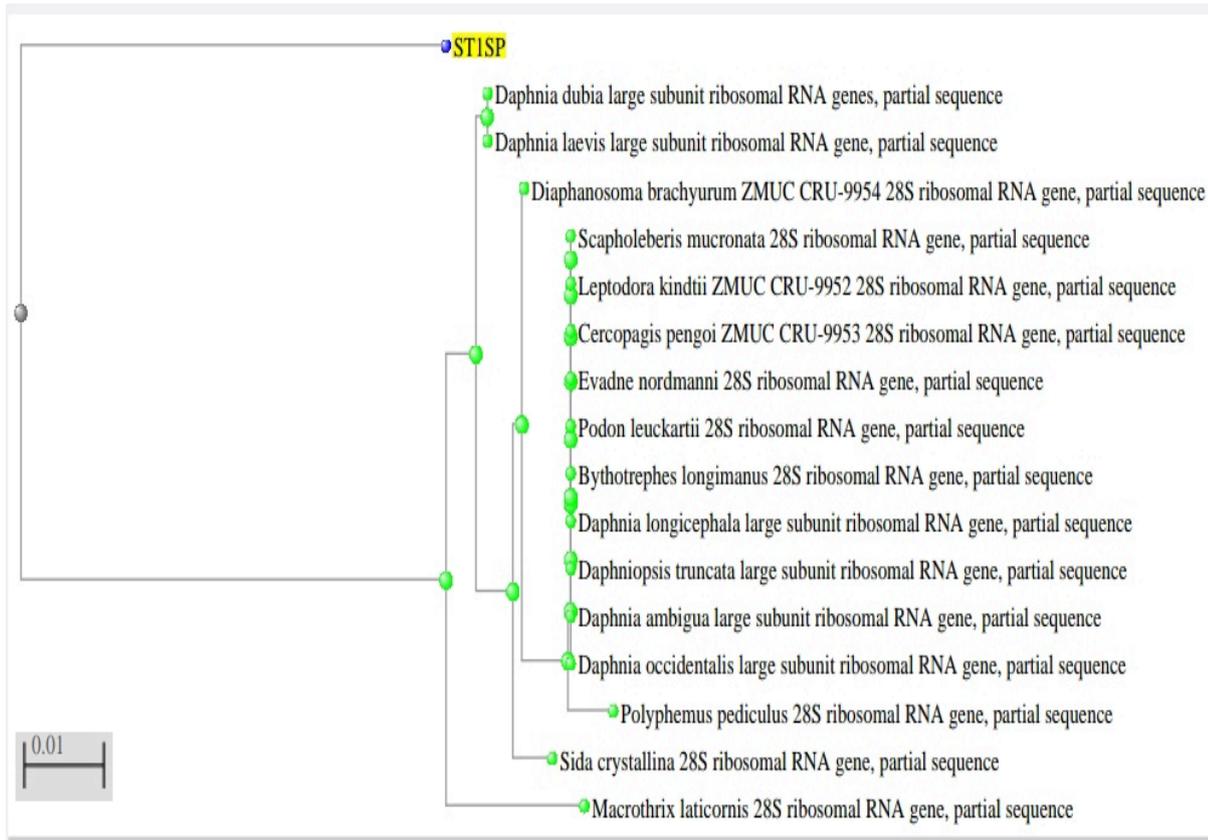
A thorough phylogenetic tree is created in the current study using nucleic acid observed in the amplified 255 bp of the 18s ribosomal RNA gene amplicons (rotifers) and the amplified 393 bp of the 18s ribosomal RNA gene amplicons (cladocera) to provide a phylogenetic understanding of the actual distances between these investigated samples and the most relative reference genotypes of rotifers and cladocera. Because it contains the majority of species along with other closely related Nuclotide sequences of rotifers, this phylogenetic tree made reference to the summer at station two (Figure 45). The species mentioned in the phylogenetic tree belong to 12 families, where the Brachionidae and Synchaetidae included most of these species. The

highest similarity (82%) was between *T.tenuior* , *T.reflexa* and *P.dolichopectera*.



**Figure (45): Phylogenetic tree among studied rotifers species.**

The phylogenetic tree made reference to the spring at station one, since it includes the most of species as well as other closely related Nuclotide sequences of cladocera (Figure 46). There are 8 families represented by the species listed in the phylogenetic tree, with the Daphniidae having the majority of these species. The highest similarity (92%) is between *Scapholeberis mucronata*, *Leptodora kindtii*, *Cercopagis pengoi*, *Evadne nordmanni*, *Podon leuckartii*, *Bythotrephes longimanus*, *Daphnia longicephala*, *Daphniopsis truncata*, *Daphnia ambigua* and *Daphnia occidentalis*.



**Figure (46): Phylogenetic tree among studied cladocera species.**

The genetic convergent between the stations during the study period with respect to rotifers is represented by the phylogenetic tree (Figure 47). The results showed that the highest convergent (85%) is between station one in summer and station five during the autumn.

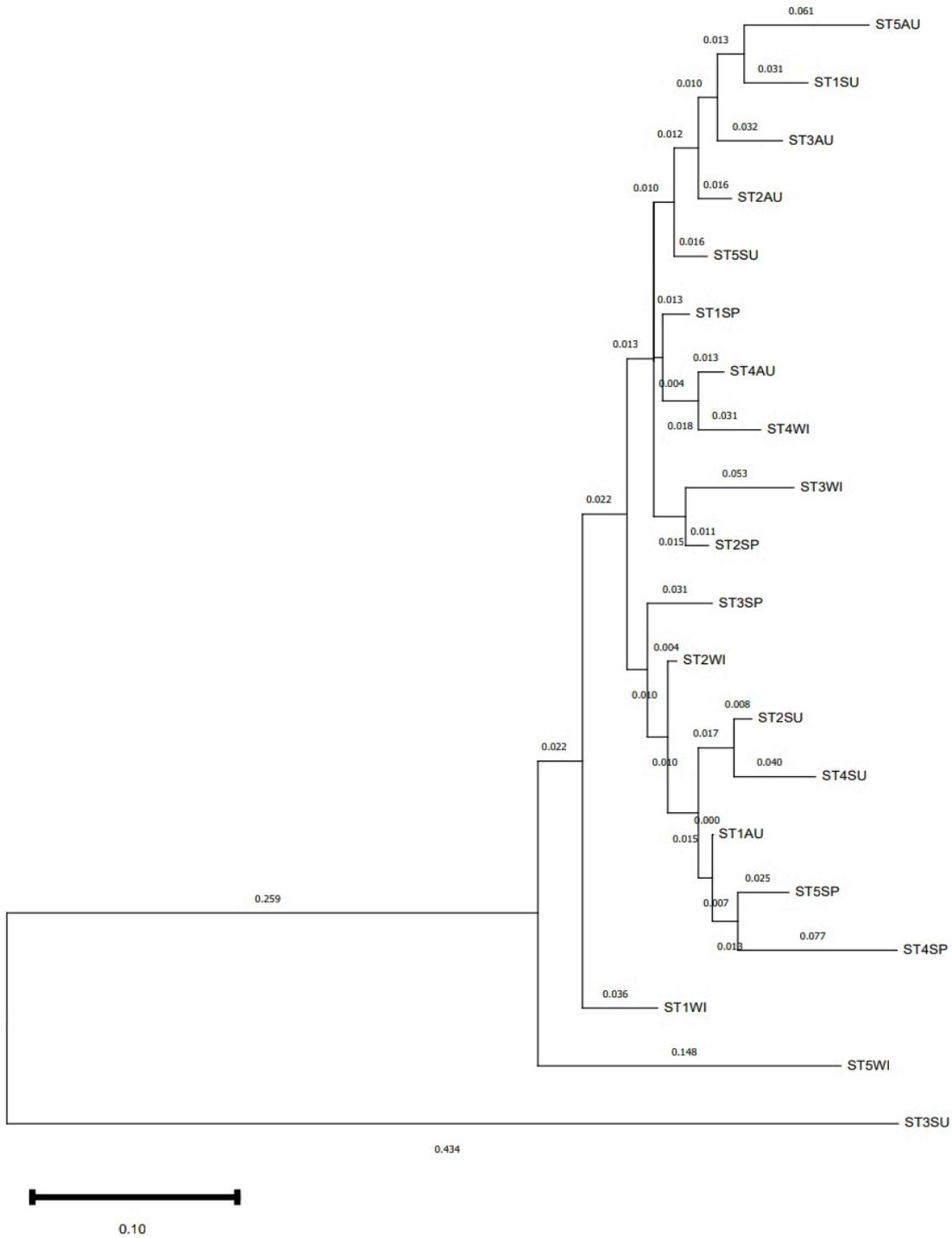


Figure (47): Phylogenetic tree of rotifers for the studied stations among different seasons.

For the Cladocera, The genetic convergent between the stations during the study period is represented by the phylogenetic tree (Figure 48). The results showed that the highest convergent (75 %) was between station one in summer and station five during the winter.

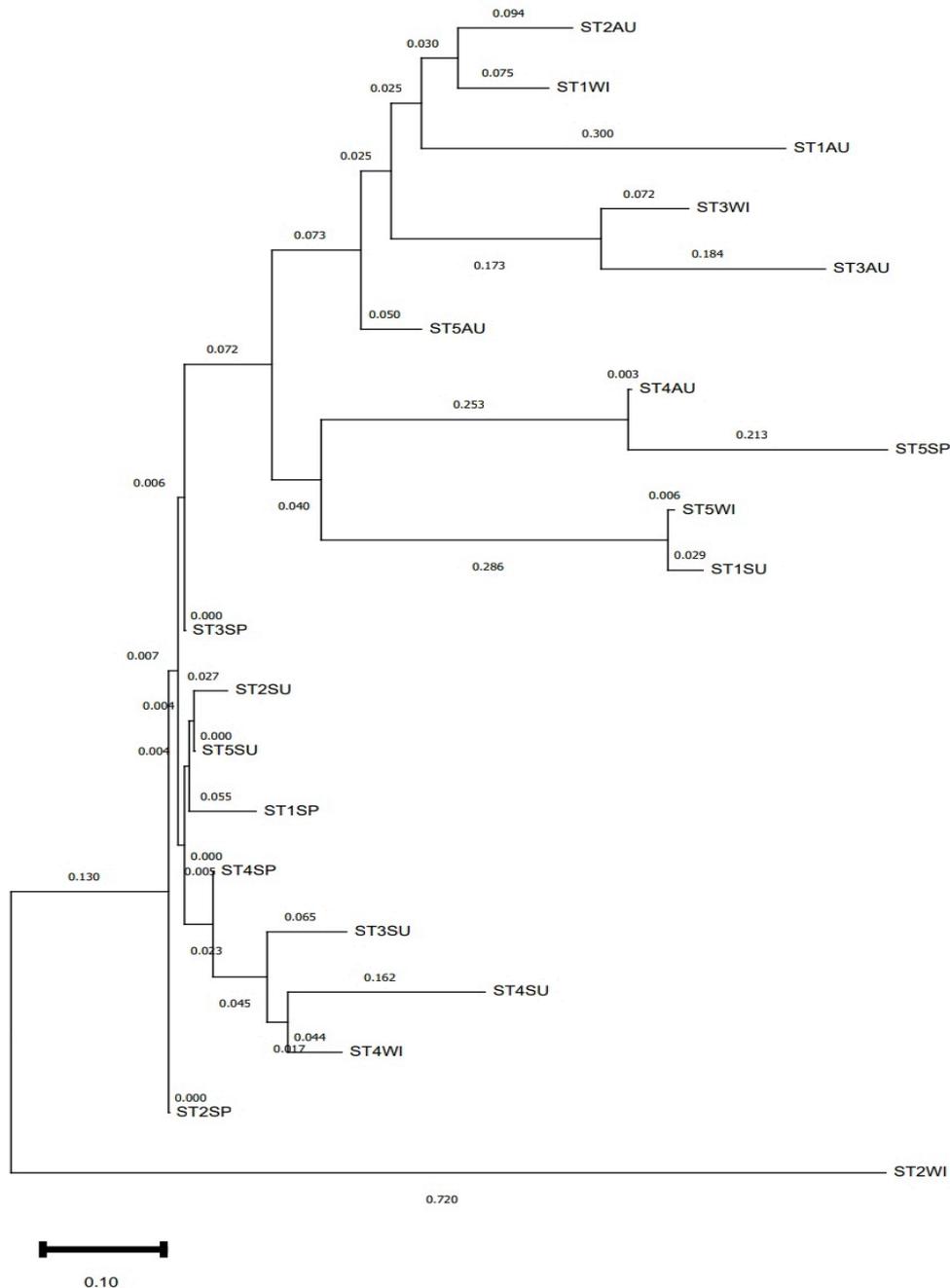


Figure (48): Phylogenetic tree of cladocera for the studied stations among different seasons.

# *Chapter Four*

## *The Discussion*

## 4 – 1 :Physical and Chemical Parameters

In an environment when the conditions are normal, the quality of water should be appropriate for the high rate of diversity, on the contrary, when an imbalance has occurred in that environment, this may lead to loss of diversity such as the fewness of species that feed on other types causes a lack of diversity and increases the opportunity for the growth of species of high densities which are originally present in small quantities in natural conditions (Jonge, 1995). Banse, (2002) concluded that a principal goal of ecology is to understand and be able to predict the abundance of organisms and the rate of changes that may occurred in them. A central challenge in ecology is to understand which factors make possible the coexistence of similar species, which affects community organization and biodiversity (Ciros-Pérez *et al.*, 2004).

The spatial and temporal distribution of zooplankton varies according to the prevailing environmental conditions in the region (Kinnesh, 1986). Patil and Auti, (2005) indicated that the environmental factors like temperature, pH, total solids, salinity and dissolved oxygen play an important role in regulating zooplankton diversity and seasonal population densities.

### 4 – 1-1 :Air and Water Temperature

Temperature is one of the most important external factors which has a profound influence and direct or indirect effect on biota of an ecosystem (Manickam *et al.*, 2014). All metabolic and physiological activities and life processes such as feeding, reproduction, movements and distribution of aquatic organism are greatly influenced by water temperature (Manickam, *et al.*, 2014). Gaikwad *et al.*, (2008) reported that water temperature in the range of 13.5°C - 32°C is appropriate for the growth of

the planktonic organisms. An increase or decrease in temperature could kill zooplankton that are sensitive to temperature changes in water (Andrulewicz *et al.*, 2008 ; Tunowski, 2009). Thus, a major factor affecting the distribution of zooplankton (Chen, 2020) and a determinant of oxygen level in the water is temperature. At a very high temperature, zooplankton tend to avoid the surface of water to avoid hypoxia (Roman *et al.*, 2019).

In the present study, temperature varied between the four seasons and for all locations. Changes in air temperature reflected the general attitude of the geographical location. As well as, water temperature followed similar trend to air temperature and they are closely related (Tiwari *et al.*, 2004 ; Manickam *et al.*, 2014). The findings of this study are consistent with the findings of the majority of Iraqi environmental studies. During the study period, the air and water temperatures showed obvious seasonal changes, owing to the nature of Iraq's climate, which is generally warm in the summer and cool in the winter.

#### **4 – 1-2 : The Potential of Hydrogen (pH)**

Aquatic organisms are affected by pH because most of their metabolic activities are depend on pH level (Wang *et al.*, 2002). A fall in pH level decreases abundance/diversity of most aquatic organisms (Andrulewicz *et al.*, 2008). On the other hand, high pH and moderate temperature promotes primary productivity which favors growth and survival of zooplankton (Mustapha, 2009).

Our analysis indicates that the higher pH value is recorded during spring 2021 and lowest in Autumn 2022 (tables 3-4). High pH may be result of decrease in water level and high photosynthetic activity of phytoplankton and other aquatic plants causing high level of oxygen

(Shiddamallayya and Pratima, 2008). Iraqi water that characterized moderate acidity to slightly alkalinity, Al-Shammary, (2014) studied Al-Hilla River and founded that water of the river is slightly alkalinity, this agree with our result and similar for many Iraqi studies. These results may be due to the type and compositions of clay elements in the areas where the river is flowing as well as the result of adding carbonates and bicarbonates to the water (Al-Amari, 2011; Prabhakar *et al.*, 2012; Al-Kasser, 2012 ; Abd Al-Hasan, 2018). According to Tanner *et al.*,(2005), the pH range between 6.0 and 8.5 indicates the medium productive nature of a reservoir; more than 8.5 highly productive and less than 6.0 low productive nature of a reservoir.

The current study observed a narrow pH range which is linked to the buffering capacity of carbonic acid and bicarbonate which are resistant to pH changes (Wetzel, 2001). Additionally, numerous investigations have noticed the small range of pH (Hassan, 2004; Salman, 2006; Al-Fanhrawi, 2010 ; Abd Al-Rezzaq, 2014).

Variations in pH are observed during different months, increase in pH indicated that density and diversity of rotifera changed with the changes in the pH either negatively or positively (Thomaz *et al.*, 2007; Sulehria *et al.*, 2009 b; Borges and Pedrozo, 2009; Dai *et al.*, 2014 ; Ejaz *et al.*, 2016).

#### **4 – 1-3 : Electrical conductivity, Total dissolved solids and Salinity**

Electrical conductivity (EC) is a good indicator of water quality (Dhanasekaran *et al.*, 2017). The salts in ionic form that dissolved in water and nutrient status are responsible for its EC. Natural water

possesses low EC but contamination increases its level (Kadam, 1990). As well as it can be used as an indirect measure of salinity (Angeler *et al.*, 2010).

It is known, the increased conductivity is recorded in the dry season probably as a result of high temperature, less solubility and high degradation of organic substances. This agree with many studies around the world (Muhammad and Ali, 2013; Tavsanoglu *et al.*, 2015; Dhanasekaran *et al.*, 2017 ; Dastgeer *et al.*, 2020). While the current study disagree with them because the highest EC values are recorded in winter and lowest values are in spring (tables 3-4).

Total dissolved solid (TDS) in water supplies originate from natural sources, sewage, urban and agricultural run-off and industrial wastewater (Muhammad and Ali, 2013). TDS is basically used to define the total ions in solution in the water (Golmarvi *et al.*, 2017). Determination of dissolved solids in the aquatic environment is very crucial. It can be toxic by increasing salinity, which causes harm to living communities, reduces diversity and causes chronic or severe effects in life cycles (Prasad, 2010).

In the present study, the highest TDS value is recorded in winter and lowest value was in spring (tables 3-4), this agree with (Alwan, 2013 ; Abd Al-Hasan, 2018) who showed that the wintertime increases in dissolved solids may be caused by increased agricultural land runoff that contains clay and fertilizers, as well as the demise and decay of some phytoplankton and diatoms.

According to many research on Al- Hilla River, the waters range from freshwater to oligohaline waters (Al-Duhaimi, 2006 ; Al-Fanhrawi, 2010). Salinity levels grew as we traveled south due to the excretion of

waste water and the river's influence on agricultural operations (Al-Nimrawi, 2005 ; Al-Fanhravi, 2010). As well as the higher salinity noticed in summer and the lower salinity noticed during winter by (Abd Al-Rezzaq, 2014). The present study disagree with him because the the higher salinity recorded in winter and the lower salinity is during spring (tables 3-4).

Regarding to EC, TDS and salinity, The present findings dosent agree with most studies because The Water Resources Department/Babylon is notified by the Meteorological Authority that an increase in water release during the summer was necessary to empty reservoirs and dams in order to prepare for the anticipated quantity of rain during the winter. Due to the lack of rain as predicted by the weather, the amount of discharge through the Hilla River reduced during the winter to 100-110 m<sup>3</sup>/s which caused the water's salinity to rise. As a result, the amount of discharge via the Hilla River was 140-150 m<sup>3</sup>/s during the summer.

#### **4 – 1-4 : Dissolved Oxygen**

Dissolved oxygen is a critical factor for the survival of aquatic life (Ejaz *et al.*, 2016). The DO level in natural water depends upon the atmospheric air pressure, photosynthetic activity, temperature, organic matter, salinity and turbulence. The solubility of oxygen increases with decrease in temperature (Singh *et al.*, 1990).

In the current study, DO values showed seasonal variations. The minimum values are measured in summer while the maximum are in winter (tables 3-4), this may be due to decline in the ratios of consumption and decomposition associated with increase in gas solubility. This is in a accordance with the conclusion of (Stevens, 2000), this may have changed because of increased temperature which reduced

the solubility of gases in the water. In addition, the decline in DO during the summer may have been brought on by increased activity of organisms that break down organic matter which increased dissolved oxygen consumption (Shakirat and Akinpelu, 2013). The negative relationship between DO and air and water temperatures supports this. Our results agree with several studies such as (Al-Jizany, 2005; Al-Mahmood *et al.*, 2008; Sulehria and Malik, 2012; Abd Al-Rezzaq, 2014; Ajeel *et al.*, 2015; Hussain *et al.*, 2016; Dhanasekaran *et al.*, 2017; Abd Al-Hasan, 2018 ; Dastgeer *et al.*, 2020).

#### **4 -2: Distribution of Zooplankton**

The distribution and density of zooplankton such Rotifera and Cladocera not only depend on their physiological requirements, but also depend on their ability to spread without being affected by external factors, and thus their ability to invade existing communities (Shurin, 2000). It is evident from the current results that all the physico-chemical characteristics of water under study affected the diversity and distribution of Rotifera and Cladocera either positively or negatively throughout the period of study. Similar reports have been made by (Chittapun *et al.*, 2007).

In the current study, class Monogononta of Rotifera was only identified in spite of the sample is stored with formalin, identified depending on the external shape and trophi, while it is difficult to identify class Bdelloidea because of its shrinkage. The result of our study demonstrated that the population density of Rotifera and Cladocera at Al-Hilla river increased during the warm months and decreased in the cold months, where it is the highest during summer and the lowest during autumn (tables 5-9) and this come agree with many studies such as Arora

and Mehra, (2003) while analyzing the seasonal dynamics of Rotifera in relation to physico-chemical conditions of water body reached to a similar conclusion, i.e. increased densities of zooplankton in summers and reduced densities in winters. In summer, the absence of inflow of water brings stability to the water body and the availability of food is more due to production of organic matter and decomposition, hence, contributing for the high species density in that season (Kiran *et al.*, 2007; Chattoopadhyay ; Barik, 2009). Similar results are also obtained by Ajeel *et al.*, (2001) in Shatt Al-Arab and some temporary ponds in Basrah; Ajeel, (2004) in Garmat Ali River and Al-Qarooni, (2005) in Al-Fuhud marshes. This could possibly be due to higher phytoplankton population concentration in the former season (Ajeel *et al.*, 2015). As well as, Mangalo and Akbar, (1986) got similar results and attributed these variations in the population densities to many factors such as the effluent disposal from the sewage treatment plant, temperature, DO, pH and EC. Erdugan and Guher, (2005) demonstrated that species diversity of Rotifera increased gradually from spring to summer months. On the other hand, when the weather began to be colder, that is the end of autumn, a decrease occurred, this diversity patterns greatly depend on the water temperature and the food supply provided within the water. In spring, when the heat of the water started to increase, an increase was observed in the phytoplankton population which is food for Rotifera. Rotifera species composition varied with seasons, Rotifera can be classified according to temperature as perennial, summer and winter species. Likely, Ahangar *et al.*, (2012) studied seasonal variation in zooplankton community structure of Anchar lake, Kashmir and showed that both of Rotifera and Cladocera have maximum peak of density during the summer season and the minimum during the autumn season, zooplankton growth is registered during moderate temperature conditions which may

be due to regeneration and availability of minerals, being an outcome of decomposition of organic matter in sediments and the algal food during this period.

In the present study, both Rotifera and Cladocera diversity tended to be greater in the dry season. Similarly, Rabee, (2015) and Ajeel *et al.*, (2019) found the peak in cladoceran species population during summer months showed the preference of this group towards the increasing temperature along with an increase in alkalinity and pH. During summer, increasing temperature enhances the rate of decomposition due to which the water becomes nutrient rich similarly due to concentration followed by evaporation in summer season, the nutrient concentration increases and abundant food present in the form of phytoplankton and microorganism to zooplankton while low density during the spring season attributed to heavy flood and fresh water inflow (Krishnamoorthy *et al.*, 2007). Manickam *et al.*, (2014) and Ajeel and Abbas, (2019) found the maximum population of cladocera was reported in summer could be attributed to favourable temperature and availability of favourable food such as bacteria, nanoplankton and suspended detritus. While in monsoon, the factors like water temperature, dissolved oxygen, turbidity and transparency play an important role in controlling the diversity and density of cladocera.

Among zooplankton samples collected in the current study, Rotifera are the dominant group. Similar findings are also observed by most local studies such as (Ahmed *et al.*, 2005 ; Salman *et al.*, 2008). Moreover, Póltorak *et al.*, (2001) claimed that Rotifera constituted more than 90% of the total zooplankton when they study the horizontal distribution of zooplankton in lakes Tharthar, Habbaniya and Razzazah. One factor that could possibly aid in Rotifera dominance is the composition of the

predator community at the studied stations (Burnes, 2006). Also, Rabee, (2015) recorded that the Zooplankton community is dominated by Rotifera during the assessment the Abundance and Diversity of Zooplankton Communities in Al-Habbaniya Lake, Iraq. Such dominance could be assigned to their short generation time and high reproductive rate (Van Dijk and Van Zanten, 1995). The great contribution of Rotifera to the species richness and abundance of the zooplankton community may be due to the opportunistic features of this group (Neves *et al.*, 2003) including their wide range of foods (from filamentous algae to bacteria) and high turnover rate which makes it possible for them to colonize even unstable environments. Rotifera and small copepods are known to be more tolerant to adverse environmental conditions than the Cladocera. The decrease in the density of Cladocera in some months could have also been due to their lower ability to take advantage of the resource (Meshram, 2005). While Ismail and Zaidin, (2015) have explained why the Cladocera were observed in lower species richness and abundance compared to Rotifera when they made a comparative study of zooplankton diversity and abundance from different types of water body and showed this is due to the effects of size-selective predation by fish (Pankow, 1991) and the changes in chemical characteristics of the water condition (Medeiros and Arthington, 2008). As well as the abundance of nauplii is always higher than the adult stages (Zakaria *et al.*, 2007). This is probably due to the larger size of adult forms which increase the predation intensity compared to juvenile forms (Sampaio *et al.*, 2002).

The densities of Rotifera and Cladocera during the current study period are shown in (tables 5-9). For rotifera, the Bdelloidea, *E.delatata*, *T.rattus*, *T.tetractis*, *L.closterocerca*, *T.patina*, *P.major*, *P. dolichoptera* and *L.bulla* were high density species of rotifera during period study.

This result agrees with result of (Abd Al- Rezzaq, 2014 ; Abd Al-Hasan, 2018) in their study on Al-Hilla river. As well as Mangalo *et al.*, (1998) pointed to *polyarthra dolichoptera* is dominant on the rest of the species. For Cladocera, *E.longispina*, *A.angustatus* and *A.curvirostris* are high density species during the period study and these species are recorded for the first time in Iraq. Our results showed differences in densities of Cladocera in different regions and different seasons this might be due to the differences in environmental conditions. This comes accordance with several studies such as Ajeel, (1998) and Salman *et al.*, (2008) during their study on southern Iraqi marshes.

#### **4–3: The correlation between the environmental factors, Rotifera and Cladocera**

The current findings suggest that seasons have strongly affected Rotifera and Cladocera density, species abundance, richness and evenness. These variations are correlated with physico-chemical parameters either positively or negatively. Availability of resources, competition, disturbances and environmental changes influence local community structure and diversity (Cottenie and Meester, 2004).

In the present study, The statistical analysis's findings showed a significant positive correlation between air and water temperature. This findings are agree with the findings of the majority of Iraqi environmental studies such as (Abd Al-Rezzaq, 2014 ; Abd Al-Hasan, 2018) indicated in their study on Al-Hilla river that there is a significant positive correlation between temperatures and this is similar to the study of Rabee, (2010) on the Tharthar channel and the Euphrates River and Hammadi, (2010) on Shatt Al-Arab. While there is nonsignificant correlation between the temperature and the density of the Rotifera and Cladocera species.

The present findings demonstrated that the highest zooplankton population was observed in summer and lowest in winter and this is agree with many studies such as (Ajeel *et al.*, 2001; Al-Qarooni, 2005; Tripathi *et al.*, 2006; Baloch *et al.*, 2008; Sulehria *et al.*, 2009 a,b ; Sulehria and Malik, 2012).

The statistical analysis of current study indicated that pH had a negative significant correlation with EC, TDS and salinity and this come agree with (Hossain and Hassan, 2020). While the EC, TDS and salinity had a positive significant correlation. These correlations agree with (Abd Al-Rezzaq, 2014; Ejaz, 2016 ; Abd Al-Hasan, 2018).

Regarding to the density of Rotifera and Cladocera species, the statistical analysis of the present study indicated that most species recorded a positive significant correlation with pH and this agree with (Sharma *et al.*, 2013 , Abd Al-Hasan, 2018). Due to pH values approaching throughout all seasons, pH had a smaller impact on the density of rotifera than other environmental parameters. Also, it had a positive significant correlation with DO and this confirmed by many studies such as (Baruah *et al.*, 1993; Banik *et al.*, 1994; Rabee, 2010 ; Abd Al-Hasan, 2018). While it reported a negative significant correlation with EC, TDS and salinity and these outcomes matched what (Al-Qarooni, 2005; Ajeel *et al.*, 2006; Sulehria *et al.*, 2009 b ; Abbas *et al.*, 2014) had predicted.

Regarding to the Biodiversity Indices, the statistical analysis of the current study indicated that the species richness indices (Menhinick index and Margalef index) recorded a positive significant correlation with air and water temperature and this agreement with (Friberg *et al.*, 2001; Castro *et al.*, 2005; Erdugan and Guher, 2005; Hessen *et al.*, 2007; Kaya

*et al.*, 2010; Ajeel *et al.*, 2019; Al-Bahathy ; Nashaat, 2021). Additionally, Nashaat *et al.*, (2016) found that the diversity of Rotifera depends mainly on the water temperature and available food and that the diversity is gradually increasing in the Spring to the Summer at rising of water temperature and increasing in the density of phytoplankton which is the source of food for Rotifera and decrease diversity at the end of Autumn when the water becomes colder. Also, it showed a positive correlation with EC and TDS, this findings agreement with (Golmarvi *et al.*, 2017) but disagreement with many studies such as (Schabetsberger *et al.*, 2009). Hammadi *et al.*, (2016) emphasize that as salinity increases, the abundance and species richness of Rotifera and microcrustaceans generally decrease, as the threshold for the majority of Microzooplankton has been placed at salinities of less than 1–2 ppt; Tavsanoğlu *et al.*, (2015) noticed the decline in zooplankton species diversity and richness with increasing salinity is likely due to their limited osmoregulation capacity (Aladin, 1991; Frey, 1993; Jeppesen *et al.*, 1994; Boix *et al.*, 2008 ; Brucet *et al.*, 2009). Likely, it showed a positive correlation with Evenness indices (Shannon index and Evenness index) which is similar to other observations like Maia-Barbosa *et al.*, (2008) who mentioned that the highest diversity index of the crustacean community is found among submerged vegetation in summer. Additionally, Our results indicated the positive correlation between species richness and the total density of the zooplankton and the greater diversity is recorded in summer season whereas low values in winter. This may be attributed to the addition of new species and resources during monsoon (Hussain *et al.*, 2016) this finding agreement with Adesalu and Nwankwo, (2008) who noted the highest number of rotifera population in the summer and the lowest in winter that may be credited to food abundance, suitability of the physico-chemical parameters of water and higher reproduction rate. Higher values

of species richness are an indication of the presence of larger food chains in the summer season as found by (Sulehria *et al.*, 2009b) who observed the greater rotifera diversity and density in the summer as compared to the winter as a result of seasonal fluctuations in rotifer populations may be attributed to reduced turbidity and high dissolved oxygen during summer and spring (Sharma *et al.*, 2013). But it recorded a negative significant correlation with DO, pH and Dominance indices (Simpson index and Berger-Parker index).

As well as, Evenness indices (Shannon index and Evenness index) demonstrated a positive significant correlation with air and water temperature and this agreement with (Majeed *et al.*, 2022) and this could be related to the favorable temperatures and phytoplankton abundance (Ajeel *et al.*, 2019). These results are consistent with (Abdulwahab and Rabee, 2015) who showed the evenness values of Rotifera in Tigris River ranged from 0.41 to 0.93.

As for Dominance indices (Simpson index and Berger-Parker Index), it showed a positive significant correlation with DO and salinity but it display a negative significant correlation with total number of taxa, total density, air and water temperature and pH.

#### **4 – 4: Biodiversity Indices**

Diversity index is commonly used as a biocriteria for the interpretation of the environmental status, as well as to measure the average degree of uncertainty within the community. The species diversity tends to be low in a stressed and polluted ecosystem (Bass and Harrel, 1981).

The high species diversity is probably also due to the season in which these habitats trap the resting eggs carried by water from higher

elevations, especially during increased flows (Arora and Mehra, 2003). Biodiversity assessments of any water body depend on the ability to identify the complement of species present, although the degree of sampling required is often uncertain (Muirhead *et al.*, 2006).

#### **4 -4-1 : Shannon - Wiener diversity index**

The Shannon-Wiener index of diversity is the most biodiversity indicators commonly used, ranging from 0-5 and when the value of this indicator is higher than 3 means that the composition of habitat is stable, and less than 1, it indicates a defect in the ecosystem caused by pollution (Turkmen and Kazanci, 2010).

The present study indicated that the second station recorded the highest value (2.83) in the summer and the fifth station recorded the lowest value (1.721) in the fall. This could be associated to increase in temperature, transparency and Chlorophyll-*a*, these factors are important for the availability of phytoplankton as a food for zooplankton (Sharmila-Sree and Shameem, 2017). Whereas the values of this index decreased in winter probably due to the higher amount of turbidity and suspended matter which effects the diversity of rotifera as mentioned by (Abdulwahab and Rabee, 2015). Or this may be due to favorable environmental conditions at these two stations because whenever the environmental conditions more stable and consistent increase diversity in the river (Trout-Haney, 2006). This findings comes agree with many studies such as Rabee, (2015) who examined the Shannon-Wiener index of Cladocera; Nashaat *et al.*, (2016) studys on Kufa river; Al-Bahathy and Nashaat, (2021) who assessment the Impact of Hindiya Dam on Rotifera Community of Euphrates River on the Northern of Babil Governorate,

Iraq and Majeed *et al.*, (2022) who evaluate the effects of Tharthar Arm on the composition and diversity of Rotifera in Tigris River.

#### **4 -4-2 : Margalef's richness and Menhinick Index index**

The Menhinick index will estimate the richness of species more accurately compared to the Margalef richness index. The Menhinick richness index assumes a relationship between the number of species and the number of individuals (Tinio and Sebual, 2021). Menhinick values vary with samples containing different numbers of individuals. Also, the Menhinick index would be useful in measuring the species richness of the community. Besides, according to (Williams *et al.*, 2005), the Menhinick index is easy to compute, but it is sensitive to sample size.

In the present study, the high values of Margalef's index (3.351) is in summer and the low value (1.278) is in autumn. As well as high values of Menhinick index (0.776) is in summer and the low value (0.343) is in spring. High values of Margalef's index and Menhinick Index indicates high diversity and thus the nutrient rich status of the water body. This is a characteristic feature of stable physico-chemical conditions. This is agreement with (Imoobe and Ohiozebau, 2009; Ashutosh mishra *et al.*, 2010 ; Rao, 2017).

#### **4 -4-3 : Index of Dominance (Simpson Index)**

The Simpson index in the present study shows high value (0.250) in station two during summer and low value (0.071) in station five during autumn. This findings indicated that the communities are mature and stable as the dominance is shared by large number of species. Low diversity which is usually showed close to zero values is a signal that the communities are under stress conditions (Dash, 2003).

#### **4 -4-4 : Evenness index**

The current study indicated that the third station reported the highest value (0.913) and the fifth station recorded the lowest value (0.717) both in the autumn perhaps it is because the third station is more stable and balanced and this is consistent with (Armengol and Miracle, 1999) during their study on Zooplankton communities in doline lakes and pools.

#### **4 – 4-5: Berger-Parker Index**

The Berger-Parker Index in the present study shows high value (0.4) in station five during autumn and low value (0.13) in station one during summer. The current findings came agree with (Priya *et al.*, 2019) through the study on zooplankton population of river Thamiraparani, Kanyakumari District, Tamilnadu and found that maximum value of Berger-Parker index (0.61) was during Post monsoon season.

#### **4 – 4-6: Jaccard's Index (%)**

The Jaccard's similarity index is used to compare the faunal similarity coefficients for the Rotifera and Cladocera communities at the five stations. These coefficients indicated that stations 2 and 3 were very much similar to each other (59%) this could be attributed to values of physical chemical characteristics of Euphrates River at these sites as well as the species that appeared in the sites. The least similarity was obtained between station 1 and 5 the (49%). Similarity of water bodies is indicative if the value of the Ss% coefficient is greater than 50% (Póltorak *et al.*, 2001). Similar observations was obtained by (Abbas *et al.*, 2014; Mirza and Nashaat, 2019 and Al-Bahathy and Nashaat, 2021).

## 4 – 5 : Morphological Identification of the Zooplankton

In the current study, 69 taxa of rotifera comprising (54 species and 12 genus) following to (17) families and 14 taxa of cladocera comprising (8 species and 2 genus) following to (4) families are identified. Three species of Rotifera and five species of Cladocera are recorded for the first time in Iraq, as well as four species of rotifera are recorded in Al-Hilla River for the first time. It was difficult to identify class Bdelloidea because the sample is kept in formalin that causes its shrinkage, therefore only the class Monogononta is recognized. The differences and changes in the number of taxonomic units of the current study compared with the previous studies may be return to several reasons, including the dominant nature of environmental conditions in the area, as well as, the nature distribution of phytoplankton and the size of the aperture size of plankton net, which may be control the quantity and quality of zooplankton collected (Ajeel *et al.*, 2008).

Many different studies identified different number of taxa of rotifera such as Ibrahim, (2005) recorded 70 species in the Dagharah Rivers; Abd Al-Rezzaq, (2014) recorded 128 taxa through his study on Hilla river; Nashaat *et al.*, (2016) found 92 taxonomic units belonging to 32 genera; Abd Al-Hasan, (2018) identified 65 taxa in Al-Hilla river. The reasons for increasing the species number of rotifera in freshwater ecosystem may be related to the lacking of interaction of rotifera with different trophic levels. Some species, such as *Keratella cochlearis* possibly live in a high-nutrition environments, as well as with a low level of nutrition (Ferraz *et al.*, 2009), in addition to their small size and parthenogenesis reproduction and the short periods of their growth (Özbay and Altındag, 2009). Also, their ability to adapt to living in low oxygen content (Jappesen *et al.*, 2002). While Jose and Sanalkumar, (2012) explained the

increasing number of rotifera attributed to the lack of food specialization, parthenogenesis reproduction and high fertility of it. The current study showed that the Brachionidae and Lecanidae numerically dominant in environments at higher temperatures and increased content of dissolved oxygen and this agreement with (Okogwu, 2010). The results also showed that some species may be frequented to appear in abundance in all study stations, including *B. angularis*, *B. calyciflorus*, *C. adriatica*, *E. delatata*, *K. cochlearis*, *K. valga* and *L. luna*.

For Cladocera, Ajeel *et al.*, (2006) identified 18 species of Cladocera belonging to 12 genera are identified, 5 species are new records to the Iraqi marshes; Ajeel and Abbas, (2012) recorded 20 species of Cladocera belonging to 14 genera are recorded in the Shatt Al-Arab River and Ajeel and Abbas, (2019) recorded 16 species of Cladocera belonging to 12 genera in the study of Diversity, Abundance and Distribution of Cladocera at the end of the Tigris River North of Basrah– Iraq.

#### **4–6 : Molecular Identification of the Rotifera and Cladocera**

The majority of genetic studies conducted worldwide have confirmed numerous errors in the diagnosis of rotifers and cladocera because the ability to discriminate between species on the basis of morphological characteristics is limited by the high level of phenotypic variation (García-Morales and Elías-Gutiérrez, 2013). Different morphological variants have often been described as different species, subspecies or forms (Segers and De Smet, 2008)). Moreover, modern genetic techniques allow accurate species identification based on minute amounts of DNA and can be used to confirm morphological identifications (Duffy *et al.*, 2000).

The current study on rotifera and cladocera at the molecular level is the first study in Iraq and it aim to solve some taxonomic problem and to encourage researchers to use these methods to do so. A PCR device is used to amplify fragment of DNA (18s ribosomal RNA Gene) and using two pairs of primers.

#### **4 – 6-1: Sequences Alignment Fragment of 18s rRNA (255 bp) of Rotifera and (393 bp ) of Cladocera**

The nitrogen bases of the 18s gene's sequence are examined for study samples that are gathered from various stations within the province of Babylon. Sequencing findings for the studied species compared with the published sequences of the species in the gene bank. As well as the new record species are deposited on the NCBI. The presence of genetic diversity among species of rotifera and cladocera confirmed that there is no complete match in the DNA sequence of this species which is agreed with some studies that confirm the existence of genetic differences due to the different geographical locations from which samples are collected (Zuykova *et al.*, 2017).

The degree of similarity in the sequence of the 18s gene is compared between the studied species with a sample studied the same gene which is previously registered at NCBI (Van Damme *et al.*, 2007) as they represent the highest similarity between them and the studied samples in the sequences of the nitrogen bases, high genetic diversity is observed between the studied samples. These results confirm variations in different environments and their impact on the environmental genomics and the interaction between genes and the environment (Stillman and Hurt, 2015). The results of sequencing analysis of 18s gene of species from Iraq when compared with that studied in world, as it represent the highest similarity ranged from ( 85 % - 93% ) according to NCBI data.

For a wide range of reasons, the current study concentrated on molecular identification of rotifera and cladocera. For rotifera, often have complex life cycles, high dispersal capacities and rapid local adaptations which may facilitate interspecific gene flow and intraspecific divergence (GoÂmez *et al.*, 2002 ; Cristescu *et al.*, 2012). Previous studies has discussed the high divergence and cryptic species in zooplankton (Gilbert and Walsh, 2005; Suatoni *et al.*, 2006 ; Elias-Gutierrez *et al.*, 2008). For cladocera, This genetic diversity is of great scientific benefit as the cladocera undergoes two breeding cycles during its lifetime, sexual reproduction and asexual reproduction. Therefore, this reproductive pattern leads to genetic changes at the gene level, giving full information about the evolutionary history of this family (Guo *et al.*, 2018).

Another possible reason for the high divergence of zooplankton in the NCBI database is the misidentification of zooplankton especially for rotifera where taxonomy remains unclear (Wallace, 2002) with few taxonomist experts (Segers, 2008). Overall, incompleteness, inaccuracy and high divergence of zooplankton reference databases is a challenge for studying zooplankton metabarcoding.

Two explanations for the geographic patterns observed in zooplankton species. Firstly, the high dispersal capacity of resting eggs allows high levels of gene flow across large areas resulting in widely separated areas with similar haplotypes and morphotypes. In this case, zooplankton displays little or no geographic structure (Xiang *et al.*, 2010). Long-distance dispersal and colonization of distant habitats, even transcontinental have been recorded in several zooplankton taxa as noticed by (De Gelas and De Meester, 2005; Mills *et al.*, 2007 ; Campillo *et al.*, 2011). Secondly, a rapid adaptation of resident populations to local environmental conditions after a historical coloni-

zation event reduces effective gene flow among populations and cause high levels of genetic differentiation and population subdivision (De Meester *et al.*, 2002). High dispersal and colonization capacity are evident in *B. quadridentatus* populations investigated in the study by (García-Morales and Domínguez-Domínguez, 2019). Based upon the results of analysis of the 18S gene at least three putative species exist within *B. quadridentatus*, however as many as seven species may be present based upon the results of analysis of the COI gene and morphological analysis.

Few studies have focused on the genetic diversity of zooplankton for many reasons and among these studies, Joachim *et al.*, (2005) indicated *Daphnia laevis* equally consists of more than four sibling species (Taylor *et al.*, 1996 ; Adamowicz *et al.*, 2004) while the African *D. laevis* is morphologically and genetically distinctive from American siblings; García-Morales and Domínguez-Domínguez, 2020 identified 13 cryptic species of *L. bulla* and displayed a significant genetic divergence. These highly divergent cryptic species also display a high dispersal and colonization capacity for long distances, as well as a high capacity to adapt to different environmental conditions.

#### **4 – 6-2 : Phylogenetic Tree of Zooplankton**

In the present study, When comparing our samples and the standard sample references, there is a genetic convergence between rotifera species is equal to 82 % and cladocera species 92%. Also, There is a similarity between some study stations. One of the most important findings of this study is that, through the evolutionary tree, there are variations in the genetic level of the studied species and have high similarity percentage of the nucleotide sequences according to data obtained from the NCBI. Most

of the studies conducted for these species indicate that there are variations in the genome within the same species according to different environments. And the ability of these species to do adaptation to changed environments and the results of our study on diversity within species agreed with the study conducted in the United States of America by (Stillman and Hurt, 2015).

The sequences of NCBI come from all over the world. These sequences show high levels of intraspecific divergence of most zooplankton species, suggesting a geographical difference. Furthermore, indigenous species sequences also show a high level of divergence compared with the sequences from NCBI. This explains why some species cannot be assigned to the species level by NCBI. Our results agreement with some studies such as ( Albadly and Jawad, 2019; Yang *et al.*, 2017).

## **Conclusions and Recommendations**

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### **Conclusions :-**

- 1 - The current findings indicated that the high values of zooplankton density and biodiversity indices were in spring and summer
- 2 – In the current study, Biodiversity indices can be adopted as excellent indicators of Al-Hilla river ecosystem and its health.
- 3 - According to the genetic identification, the current study included identification more species of Rotifera and Cladocera compared to the phenotypic identification.

### **Recommendations :-**

- 1 – Studying the biodiversity of zooplankton and invertebrates in all Iraqi waters.
- 2 - Reliance on genetic and molecular methods to identification the species of zooplankton in addition to the phenotypic identification

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# *Appendixes*

**Appendix (1): The Correlation between some environmental parameters , species density and number of taxa among station one**

	<i>A. saltans</i>	<i>A. priodonta</i>	<i>Bdelloidea</i>	<i>B. forficula</i>	<i>B. rubens</i>	<i>Brachionus spp</i>	<i>B. angularis</i>	<i>B. falcatus</i>	<i>C. gibba</i>	<i>Cephalodella sp</i>	<i>Collotheca sp</i>	<i>C. adriatica</i>	<i>Colurella sp</i>	<i>E. porsildi</i>	<i>E. dilatata</i>	<i>H. brehmi</i>	<i>K. tecta</i>
<i>A. ovalis</i>	.577	-.333	-.032	-.333	-.333	1.000**	-.333	1.000**	-.919	-.333	-.333	1.000**	-.333	1.000**	-.478	-.333	.577
<i>A. saltans</i>		-.577	.078	.577	.577	.577	-.577	.577	-.423	-.577	-.577	.577	-.577	.577	-.397	-.577	0.000
<i>A. priodonta</i>			.765	-.333	-.333	-.333	1.000**	-.333	.548	-.333	-.333	-.333	-.333	-.333	-.478	-.333	.577
<i>Bdelloidea</i>				.122	.122	-.032	.765	-.032	.414	-.855	-.855	-.032	-.855	-.032	-.861	-.855	.635
<i>B. forficula</i>					1.000**	-.333	-.333	-.333	.430	-.333	-.333	-.333	-.333	-.333	.020	-.333	-.577
<i>B. rubens</i>						-.333	-.333	-.333	.430	-.333	-.333	-.333	-.333	-.333	.020	-.333	-.577
<i>Brachionus spp</i>							-.333	1.000**	-.919	-.333	-.333	1.000**	-.333	1.000**	-.478	-.333	.577
<i>B. angularis</i>								-.333	.548	-.333	-.333	-.333	-.333	-.333	-.478	-.333	.577
<i>B. falcatus</i>									-.919	-.333	-.333	1.000**	-.333	1.000**	-.478	-.333	.577
<i>C. gibba</i>										-.059	-.059	-.919	-.059	-.919	.098	-.059	-.322
<i>Cephalodella sp</i>											1.000**	-.333	1.000**	-.333	.936	1.000**	-.577
<i>Collotheca sp</i>												-.333	1.000**	-.333	.936	1.000**	-.577
<i>C. adriatica</i>													-.333	1.000**	-.478	-.333	.577
<i>Colurella sp</i>														-.333	.936	1.000**	-.577
<i>E. porsildi</i>															-.478	-.333	.577
<i>E. dilatata</i>																.936	-.828
<i>H. brehmi</i>																	-.577
<i>K. tecta</i>																	

Continue to Appendix (1)

	<i>L. bulla</i>	<i>L. closterocerca</i>	<i>L. elsa</i>	<i>L. hamata</i>	<i>L. lunaris</i>	<i>L. nana</i>	<i>L. quadridentata</i>	<i>Lecane sp</i>	<i>L. patella</i>	<i>Lepadella spp</i>	<i>M. sericus</i>	<i>M. angusta</i>	<i>M. ventralis</i>	<i>Naupilus</i>	<i>P. quadricornis</i>	<i>P. dolichoptera</i>	<i>P. major</i>	<i>Rotaria sp</i>	<i>Rotifers</i>	<i>S. longicaudum</i>
<i>A. ovalis</i>	.577	-.333	-.333	-.333	-.333	-.333	-.333	.987*	-.577	1.000**	-.333	-.333	1.000**	.025	-.333	-.333	-.333	-.333	-.333	-.333
<i>A. saltans</i>	1.000**	.577	-.577	-.577	.577	.577	.577	.700	0.000	.577	-.577	.577	.577	.422	-.577	-.577	-.577	-.577	-.577	-.577
<i>A. priodonta</i>	-.577	-.333	-.333	-.333	-.333	-.333	-.333	-.404	-.577	-.333	1.000**	-.333	-.333	-.951*	-.333	-.333	1.000*	-.333	-.333	-.333
<i>Bdelloidea</i>	.078	.122	-.855	-.855	.122	.122	.122	-.013	-.635	-.032	.765	.122	-.032	-.801	-.855	-.855	.765	-.855	-.855	-.855
<i>B. forficula</i>	.577	1.000**	-.333	-.333	1.000**	1.000**	1.000**	-.179	.577	-.333	-.333	1.000**	-.333	.463	-.333	-.333	-.333	-.333	-.333	-.333
<i>B. rubens</i>	.577	1.000**	-.333	-.333	1.000**	1.000**	1.000**	-.179	.577	-.333	-.333	1.000**	-.333	.463	-.333	-.333	-.333	-.333	-.333	-.333
<i>Brachionus spp</i>	.577	-.333	-.333	-.333	-.333	-.333	-.333	.987*	-.577	1.000**	-.333	-.333	1.000**	.025	-.333	-.333	-.333	-.333	-.333	-.333
<i>B. angularis</i>	-.577	-.333	-.333	-.333	-.333	-.333	-.333	-.404	-.577	-.333	1.000**	-.333	-.333	-.951*	-.333	-.333	1.000*	-.333	-.333	-.333
<i>B. falcatus</i>	.577	-.333	-.333	-.333	-.333	-.333	-.333	.987*	-.577	1.000**	-.333	-.333	1.000**	.025	-.333	-.333	-.333	-.333	-.333	-.333
<i>C. gibba</i>	-.423	.430	-.059	-.059	.430	.430	.430	-.886	.322	-.919	.548	.430	-.919	-.278	-.059	-.059	.548	-.059	-.059	-.059
<i>Cephalodella sp</i>	-.577	-.333	1.000**	1.000**	-.333	-.333	-.333	-.404	.577	-.333	-.333	-.333	-.333	.463	1.000**	1.000**	-.333	1.000**	1.000**	1.000**
<i>Collotheca sp</i>	-.577	-.333	1.000**	1.000**	-.333	-.333	-.333	-.404	.577	-.333	-.333	-.333	-.333	.463	1.000**	1.000**	-.333	1.000**	1.000**	1.000**
<i>C. adriatica</i>	.577	-.333	-.333	-.333	-.333	-.333	-.333	.987*	-.577	1.000**	-.333	-.333	1.000**	.025	-.333	-.333	-.333	-.333	-.333	-.333
<i>Colurella sp</i>	-.577	-.333	1.000**	1.000**	-.333	-.333	-.333	-.404	.577	-.333	-.333	-.333	-.333	.463	1.000**	1.000**	-.333	1.000**	1.000**	1.000**
<i>E. porsildi</i>	.577	-.333	-.333	-.333	-.333	-.333	-.333	.987*	-.577	1.000**	-.333	-.333	1.000**	.025	-.333	-.333	-.333	-.333	-.333	-.333
<i>E. dilatata</i>	-.397	.020	.936	.936	.020	.020	.020	-.495	.828	-.478	-.478	.020	-.478	.664	.936	.936	-.478	.936	.936	.936
<i>H. brehmi</i>	-.577	-.333	1.000**	1.000**	-.333	-.333	-.333	-.404	.577	-.333	-.333	-.333	-.333	.463	1.000**	1.000**	-.333	1.000**	1.000**	1.000**
<i>K. tecta</i>	0.000	-.577	-.577	-.577	-.577	-.577	-.577	.505	-	.577	.577	-.577	.577	-.802	-.577	-.577	.577	-.577	-.577	-.577
<i>L. bulla</i>		.577	-.577	-.577	.577	.577	.577	.700	1.000**	.577	-.577	.577	.577	.422	-.577	-.577	-.577	-.577	-.577	-.577
<i>L. closterocerca</i>			-.333	-.333	1.000**	1.000**	1.000**	-.179	.577	-.333	-.333	1.000**	-.333	.463	-.333	-.333	-.333	-.333	-.333	-.333
<i>L. elsa</i>				1.000**	-.333	-.333	-.333	-.404	.577	-.333	-.333	-.333	-.333	.463	1.000**	1.000**	-.333	1.000**	1.000**	1.000**
<i>L. hamata</i>					-.333	-.333	-.333	-.404	.577	-.333	-.333	-.333	-.333	.463	1.000**	1.000**	-.333	1.000**	1.000**	1.000**
<i>L. lunaris</i>						1.000**	1.000**	-.179	.577	-.333	-.333	1.000**	-.333	.463	-.333	-.333	-.333	-.333	-.333	-.333
<i>L. nana</i>							1.000**	-.179	.577	-.333	-.333	1.000**	-.333	.463	-.333	-.333	-.333	-.333	-.333	-.333
<i>L. quadridentata</i>								-.179	.577	-.333	-.333	1.000**	-.333	.463	-.333	-.333	-.333	-.333	-.333	-.333
<i>Lecane sp</i>									-.505	.987*	-.404	-.179	.987*	.104	-.404	-.404	-.404	-.404	-.404	-.404





<i>S.longicaudum</i>	<b>-0.333</b>	<b>-0.333</b>	<b>1.000*</b>	-	<b>-0.333</b>	<b>1.000*</b>	<b>-0.166</b>	<b>-0.333</b>	<b>.995**</b>	<b>-0.333</b>	<b>-0.333</b>	<b>.577</b>	<b>-0.333</b>	<b>-0.005</b>	<b>-0.333</b>	<b>.133</b>	<b>.137</b>	<b>.589</b>	<b>-0.914</b>	<b>-0.918</b>	-	-	<b>.463</b>
			<b>.551</b>																		<b>.950</b>	<b>.258</b>	
<i>S.lakowitziana</i>		<b>1.000*</b>	<b>-0.333</b>	-	<b>-0.333</b>	<b>-0.333</b>	<b>-0.847</b>	<b>-0.333</b>	<b>-0.381</b>	<b>-0.333</b>	<b>-0.333</b>	<b>.577</b>	<b>1.000*</b>	<b>.822</b>	<b>-0.333</b>	<b>-0.842</b>	<b>-0.901</b>	<b>.339</b>	<b>.607</b>	<b>.553</b>	<b>.317</b>	<b>.978</b>	<b>-</b>
			<b>.551</b>																			<b>.841</b>	
<i>Synchaeta sp</i>			<b>-0.333</b>	-	<b>-0.333</b>	<b>-0.333</b>	<b>-0.847</b>	<b>-0.333</b>	<b>-0.381</b>	<b>-0.333</b>	<b>-0.333</b>	<b>.577</b>	<b>1.000*</b>	<b>.822</b>	<b>-0.333</b>	<b>-0.842</b>	<b>-0.901</b>	<b>.339</b>	<b>.607</b>	<b>.553</b>	<b>.317</b>	<b>.978</b>	<b>-</b>
			<b>.551</b>																			<b>.841</b>	
<i>T.rattus</i>			-	<b>-0.333</b>	<b>1.000*</b>	<b>-0.166</b>	<b>-0.333</b>	<b>.995**</b>	<b>-0.333</b>	<b>-0.333</b>	<b>.577</b>	<b>-0.333</b>	<b>-0.005</b>	<b>-0.333</b>	<b>.133</b>	<b>.137</b>	<b>.589</b>	<b>-0.914</b>	<b>-0.918</b>	-	-	<b>.463</b>	
			<b>.551</b>																		<b>.950</b>	<b>.258</b>	
<i>T.patina</i>				<b>.307</b>	<b>-0.551</b>	<b>.908</b>	<b>.307</b>	<b>-0.534</b>	<b>.795</b>	<b>.307</b>	<b>-0.954*</b>	<b>-0.551</b>	<b>-0.528</b>	<b>.307</b>	<b>.431</b>	<b>.513</b>	-	<b>.328</b>	<b>.391</b>	<b>.617</b>	-	<b>.162</b>	
																<b>.939</b>					<b>.536</b>		
<i>T.reflexa</i>					<b>-0.333</b>	<b>.313</b>	<b>1.000**</b>	<b>-0.233</b>	<b>-0.333</b>	<b>1.000*</b>	<b>-0.577</b>	<b>-0.333</b>	<b>-0.811</b>	<b>1.000*</b>	<b>.776</b>	<b>.707</b>	<b>.005</b>	<b>-0.048</b>	<b>-0.061</b>	<b>.060</b>	-	<b>.601</b>	
																					<b>.521</b>	<b>.601</b>	
<i>T.pocillum</i>						<b>-0.166</b>	<b>-0.333</b>	<b>.995**</b>	<b>-0.333</b>	<b>-0.333</b>	<b>.577</b>	<b>-0.333</b>	<b>-0.005</b>	<b>-0.333</b>	<b>.133</b>	<b>.137</b>	<b>.589</b>	<b>-0.914</b>	<b>-0.918</b>	-	-	<b>.463</b>	
																					<b>.950</b>	<b>.258</b>	
<i>T.tetractis</i>						<b>.313</b>	<b>-0.137</b>	<b>.700</b>	<b>.313</b>	<b>-0.878</b>	<b>-0.847</b>	<b>-0.715</b>	<b>.313</b>	<b>.665</b>	<b>.752</b>	-	<b>-0.096</b>	<b>-0.027</b>	<b>.232</b>	-	<b>.501</b>		
																<b>.778</b>					<b>.816</b>	<b>.501</b>	
<i>A.curvirostris</i>								<b>-0.233</b>	<b>-0.333</b>	<b>1.000*</b>	<b>-0.577</b>	<b>-0.333</b>	<b>-0.811</b>	<b>1.000*</b>	<b>.776</b>	<b>.707</b>	<b>.005</b>	<b>-0.048</b>	<b>-0.061</b>	<b>.060</b>	-	<b>.601</b>	
																					<b>.521</b>	<b>.601</b>	
<i>A.angustatus</i>										<b>-0.381</b>	<b>-0.233</b>	<b>.532</b>	<b>-0.381</b>	<b>-0.095</b>	<b>-0.233</b>	<b>.223</b>	<b>.219</b>	<b>.608</b>	<b>-0.948</b>	<b>-0.954*</b>	-	<b>.544</b>	
																					<b>.973</b>	<b>.324</b>	
<i>Bosminidae</i>											<b>-0.333</b>	<b>-0.577</b>	<b>-0.333</b>	<b>-0.005</b>	<b>-0.333</b>	<b>-0.068</b>	<b>.058</b>	-	<b>.355</b>	<b>.426</b>	<b>.572</b>	-	
																		<b>.933</b>				<b>.199</b>	<b>.223</b>
<i>Chydoridae</i>												<b>-0.577</b>	<b>-0.333</b>	<b>-0.811</b>	<b>1.000*</b>	<b>.776</b>	<b>.707</b>	<b>.005</b>	<b>-0.048</b>	<b>-0.061</b>	<b>.060</b>	-	
																						<b>.521</b>	<b>.601</b>
<i>D.magna</i>												<b>.577</b>	<b>.707</b>	<b>-0.577</b>	<b>-0.614</b>	<b>-0.662</b>	<b>.804</b>	<b>-0.266</b>	<b>-0.317</b>	-	<b>.623</b>	-	
																					<b>.548</b>	<b>.327</b>	
<i>Daphniidae</i>																					<b>.317</b>	<b>.978</b>	-
																						<b>.841</b>	
<i>E.longispina</i>														<b>-0.811</b>	-	<b>-0.986*</b>	<b>.207</b>	<b>.406</b>	<b>.379</b>	<b>.160</b>	<b>.921</b>	-	
															<b>.991**</b>							<b>.885</b>	
<i>Cladocera</i>															<b>.776</b>	<b>.707</b>	<b>.005</b>	<b>-0.048</b>	<b>-0.061</b>	<b>.060</b>	-	<b>.601</b>	
																						<b>.521</b>	<b>.601</b>
Air Temperature																	<b>.991**</b>	-	<b>-0.521</b>	<b>-0.498</b>	-	<b>.939</b>	
																		<b>.105</b>			<b>.289</b>	<b>.936</b>	
Water Temperature																			<b>-0.515</b>	<b>-0.481</b>	-	<b>.925</b>	
																					<b>.255</b>	<b>.972</b>	
pH																				<b>-0.499</b>	<b>-0.564</b>	-	<b>.149</b>
																					<b>.740</b>	<b>.271</b>	
E.C																					<b>.997**</b>	<b>.588</b>	-
																						<b>.782</b>	
TDS																						<b>.965</b>	<b>.543</b>
																						<b>.766</b>	
Salinity																						<b>.307</b>	<b>.598</b>
																						<b>.908</b>	
D.O																							

Appendix (2): The Correlation between some environmental parameters , species density and number of taxa among station two

	<i>B. forficula</i>	<i>B. calyciflorus</i>	<i>C. gibba</i>	<i>Cephalodella</i> sp	<i>E. porsildi</i>	<i>E. dilatata</i>	<i>Euchlanis</i> spp	<i>F. opoliensis</i>	<i>K. cochlearis</i>	<i>K. quadrata</i>	<i>Keratella</i> sp	<i>K. tecta</i>	<i>K. tropica</i>
<i>Bdelloidea</i>	.227	-.992**	-.487	.430	.227	.569	.227	.227	.915	-.992**	.663	-.992**	.487
<i>B. forficula</i>		-.333	-.577	-.333	1.000**	.577	1.000**	1.000**	.435	-.333	-.577	-.333	.577
<i>B. calyciflorus</i>			.577	-.333	-.333	-.577	-.333	-.333	-.915	1.000**	-.577	1.000**	-.577
<i>C. gibba</i>				.577	-.577	0.000	-.577	-.577	-.317	.577	0.000	.577	-1.000**
<i>Cephalodella</i> sp					-.333	.577	-.333	-.333	.549	-.333	.577	-.333	-.577
<i>E. porsildi</i>						.577	1.000**	1.000**	.435	-.333	-.577	-.333	.577
<i>E. dilatata</i>							.577	.577	.852	-.577	0.000	-.577	0.000
<i>Euchlanis</i> spp								1.000**	.435	-.333	-.577	-.333	.577
<i>F. opoliensis</i>									.435	-.333	-.577	-.333	.577
<i>K. cochlearis</i>										-.915	.416	-.915	.317
<i>K. quadrata</i>											-.577	1.000**	-.577
<i>Keratella</i> sp												-.577	0.000
<i>K. tecta</i>													-.577

Continue to Appendix (2)

	<i>L. bulla</i>	<i>L. closterocerca</i>	<i>L. hamata</i>	<i>L. ludwigii</i>	<i>L. lunaris</i>	<i>L. nana</i>	<i>L. quadridentata</i>	<i>Lecane</i> sp	<i>L. tenuiseta</i>	<i>L. thienemanni</i>	<i>L. unguolata</i>	<i>M. angusta</i>	<i>Naupilus</i>	<i>P. quadricornis</i>	<i>P. major</i>	Rotifers	<i>S. longicaudum</i>
<i>Bdelloidea</i>	.291	.335	.227	.227	.227	.227	.227	.227	.227	.335	.227	.227	.551	.227	-.992**	.227	-.992**
<i>B. forficula</i>	.988*	-.333	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	-.333	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333
<i>B. calyciflorus</i>	-.401	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.494	-.333	1.000**	-.333	1.000**
<i>C. gibba</i>	-.695	-.577	-.577	-.577	-.577	-.577	-.577	-.577	-.577	-.577	-.577	-.577	.367	-.577	.577	-.577	.577
<i>Cephalodella</i> sp	-.401	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	.917	-.333	-.333	-.333	-.333
<i>E. porsildi</i>	.988*	-.333	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	-.333	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333
<i>E. dilatata</i>	.508	-.577	.577	.577	.577	.577	.577	.577	.577	-.577	.577	.577	.855	.577	-.577	.577	-.577
<i>Euchlanis</i> spp	.988*	-.333	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	-.333	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333

<i>F.opoliensis</i>	.988*	-.333	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	-.333	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333	
<i>K.cochlearis</i>	.442	-.069	.435	.435	.435	.435	.435	.435	.435	.435	-.069	.435	.435	.765	.435	-.915	.435	-.915	
<i>K.quadrata</i>	-.401	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.494	-.333	1.000**	-.333	1.000**	
<i>Keratella sp</i>	-.508	.577	-.577	-.577	-.577	-.577	-.577	-.577	-.577	-.577	.577	-.577	-.577	.367	-.577	-.577	-.577	-.577	
<i>K.tecta</i>	-.401	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.494	-.333	1.000**	-.333	1.000**	
<i>K.tropica</i>	.695	.577	.577	.577	.577	.577	.577	.577	.577	.577	.577	.577	.577	-.367	.577	-.577	.577	-.577	
<i>L.bulla</i>		-.185	.988*	.988*	.988*	.988*	.988*	.988*	.988*	.988*	-.185	.988*	.988*	-.007	.988*	-.401	.988*	-.401	
<i>L.closterocerca</i>			-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	1.000**	-.333	-.333	-.494	-.333	-.333	-.333	-.333	
<i>L.hamata</i>				1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	-.333	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333	
<i>L.ludwigii</i>					1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	-.333	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333	
<i>L.lunaris</i>						1.000**	1.000**	1.000**	1.000**	1.000**	-.333	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333	
<i>L.nana</i>							1.000**	1.000**	1.000**	1.000**	-.333	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333	
<i>L.quadridentata</i>								1.000**	1.000**	1.000**	-.333	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333	
<i>Lecane sp</i>									1.000**	1.000**	-.333	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333	
<i>L.tenuiseta</i>										-.333	1.000**	1.000**	1.000**	.071	1.000**	-.333	1.000**	-.333	
<i>L.thienemanni</i>												-.333	-.333	-.494	-.333	-.333	-.333	-.333	
<i>L.ungulata</i>													1.000**	.071	1.000**	-.333	1.000**	-.333	
<i>M.angusta</i>														.071	1.000**	-.333	1.000**	-.333	
<i>Naupilus</i>															.071	-.494	.071	-.494	
<i>P.quadricornis</i>																-.333	1.000**	-.333	
<i>P.major</i>																		-.333	1.000**
<i>Rotifers</i>																			-.333

	<i>Synchaeta spp</i>	<i>T.rattus</i>	<i>Trichocerca spp</i>	<i>T.patina</i>	<i>T.tetractis</i>	<i>A.curvirostris</i>	<i>Campocercus sp</i>	<i>D.magna</i>	<i>Daphniidae</i>	<i>E.longispina</i>	<i>Cladocera</i>	Air Temperature	Water Temperature	pH	E.C	TDS	Salinity	D.O	Total No of Taxa
<i>Bdelloidea</i>	-.831	.639	.430	-.124	.168	-.799	.430	.430	.430	.246	.186	.781	.846	.349	-.571	-.565	-.435	-.968*	.510
<i>B.forficula</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>B.calyciflorus</i>	.887	-.538	-.333	0.000	-.174	.862	-.333	-.333	-.333	-.125	-.144	-.840	-.899	-.269	.506	.504	.359	.985*	-.566
<i>C.gibba</i>	.889	.256	.577	-.707	.302	.705	.577	.577	.577	.659	-.249	-.618	-.671	.605	-.380	-.368	-.528	.558	-.306
<i>Cephalodella sp</i>	.140	.833	1.000**	-.816	.522	-.048	1.000**	1.000**	1.000**	.885	-.144	.126	.125	.968*	-.945	-.929	-.968*	-.341	.213
<i>E.porsildi</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849

<i>E. dilatata</i>	-.323	.256	.577	0.000	.905	-.705	.577	.577	.577	.134	-.685	.785	.724	.709	-.810	-.837	-.750	-.695	.920
<i>Euchlanis spp</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>F.opoliensis</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>K.cochlearis</i>	-.692	.524	.549	-.070	.550	-.859	.549	.549	.549	.213	-.220	.886	.897	.563	-.756	-.765	-.638	-	.779
<i>K.quadrata</i>	.887	-.538	-.333	0.000	-.174	.862	-.333	-.333	-.333	-.125	-.144	-.840	-.899	-.269	.506	.504	.359	.958*	-.566
<i>Keratella sp</i>	-.323	.932	.577	-.707	-.302	-.083	.577	.577	.577	.740	.685	.052	.163	.363	-.446	-.405	-.400	-.454	-.245
<i>K.tecta</i>	.887	-.538	-.333	0.000	-.174	.862	-.333	-.333	-.333	-.125	-.144	-.840	-.899	-.269	.506	.504	.359	.985*	-.566
<i>K.tropica</i>	-.889	-.256	-.577	.707	-.302	-.705	-.577	-.577	-.577	-.659	.249	.618	.671	-.605	.380	.368	.528	-.558	.306
<i>L.bulla</i>	-.618	-.522	-.401	.851	.403	-.806	-.401	-.401	-.401	-.766	-.523	.802	.751	-.245	.079	.036	.189	-.511	.805
<i>L.closterocerca</i>	-.513	.243	-.333	0.000	-.870	-.048	-.333	-.333	-.333	-.030	.935	-.066	.064	-.549	.430	.462	.507	-.183	-.496
<i>L.hamata</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>L.ludwigii</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>L.lunaris</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>L.nana</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>L.quadridentata</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>Lecane sp</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>L.tenuiseta</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>L.thienemanni</i>	-.513	.243	-.333	0.000	-.870	-.048	-.333	-.333	-.333	-.030	.935	-.066	.064	-.549	.430	.462	.507	-.183	-.496
<i>L.ungulata</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>M.angusta</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	-.849
<i>Naupilus</i>	-.069	.654	.917	-.518	.774	-.375	.917	.917	.917	.627	-.426	.463	.433	.961*	-	-	-	-.556	.585
														.996*	.999*	.981*			
<i>P.quadricornis</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>P.major</i>	.887	-.538	-.333	0.000	-.174	.862	-.333	-.333	-.333	-.125	-.144	-.840	-.899	-.269	.506	.504	.359	.985*	-.566
<i>Rotifers</i>	-.513	-.538	-.333	.816	.522	-.766	-.333	-.333	-.333	-.730	-.647	.780	.711	-.150	.009	-.037	.103	-.461	.849
<i>S.longicaudum</i>	.887	-.538	-.333	0.000	-.174	.862	-.333	-.333	-.333	-.125	-.144	-.840	-.899	-.269	.506	.504	.359	.985*	-.566
<i>Synchaeta sp</i>		-.157	.140	-.400	.073	.881	.140	.140	.140	.303	-.221	-.820	-.883	.191	.068	.074	-.098	.868	-.490
<i>T.rattus</i>			.833	-.840	.027	-.077	.833	.833	.833	.893	.400	.091	.166	.674	-.720	-.685	-.699	-.458	-.072
<i>Trichocerca sp</i>				-.816	.522	-.048	1.000**	1.000**	1.000**	.885	-.144	.126	.125	.968*	-.945	-.929	-	-.341	.213
																	.968*		
<i>T.patina</i>					0.000	-.440	-.816	-.816	-.816	-.989*	-.308	.400	.359	-.684	.584	.546	.656	-.074	.390
<i>T.tetractis</i>						-.400	.522	.522	.522	.097	-.901	.508	.403	.714	-.713	-.746	-.717	-.323	.814
<i>A.curvirostris</i>							-.048	-.048	-.048	.308	.250	-.993**	-.996**	-.103	.340	.364	.190	.920	-.844
<i>Camptocercus sp</i>								1.000**	1.000**	.885	-.144	.126	.125	.968*	-.945	-.929	-	-.341	.213
																	.968*		

<i>D.magna</i>	1.000**	.885	-.144	.126	.125	.968*	-.945	-.929	-	-.341	.213
<i>Daphniidae</i>		.885	-.144	.126	.125	.968*	-.945	-.929	.968*	-.341	.213
<i>E.longispina</i>			.251	-.264	-.224	.762	-.689	-.654	-.744	-.061	-.260
<i>Cladocera</i>				-.348	-.217	-.387	.344	.388	.371	.026	-.722
Air Temperature					.991**	.201	-.424	-.450	-.283	-.913	.898
Water Temperature						.166	-.404	-.425	-.254	-	.831
pH							.966*	.963*	.995*	-.319	.378
E.C								.999*	.986*	.553	-.520
TDS									.983*	.560	-.558
Salinity										.407	-.430
D.O											-.698

Appendix (3): The Correlation between some environmental parameters , species density and number of taxa among station three

	<i>B.forficula</i>	<i>B.calyciflorus</i>	<i>B.falcatus</i>	<i>C.gibba</i>	<i>Collotheca sp</i>	<i>C.adriatica</i>	<i>D.prionacis</i>	<i>E.porsildi</i>	<i>E.dilatata</i>	<i>F.opoliensis</i>	<i>K.tecta</i>	<i>K.tropica</i>
<i>Bdelloidea</i>	.990**	.990**	.990**	.536	-.462	-.264	.990**	.629	-.210	-.316	-.264	.990**
<i>B.forficula</i>		1.000**	1.000**	.479	-.333	-.333	1.000**	.577	-.280	-.180	-.333	1.000**
<i>B.calyciflorus</i>			1.000**	.479	-.333	-.333	1.000**	.577	-.280	-.180	-.333	1.000**
<i>B.falcatus</i>				.479	-.333	-.333	1.000**	.577	-.280	-.180	-.333	1.000**
<i>C.gibba</i>					-.574	-.574	.479	-.081	.708	-.518	-.574	.479
<i>Collotheca sp</i>						-.333	-.333	-.577	-.359	.987*	-.333	-.333
<i>C.adriatica</i>							-.333	.577	-.359	-.404	1.000**	-.333
<i>D.prionacis</i>								.577	-.280	-.180	-.333	1.000**
<i>E.porsildi</i>									-.554	-.506	.577	.577
<i>E.dilatata</i>										-.422	-.359	-.280
<i>F.opoliensis</i>											-.404	-.180
<i>K.tecta</i>												-.333

Continue to Appendix (3)

	<i>L.arcula</i>	<i>L.bulla</i>	<i>L.hamata</i>	<i>L.luna</i>	<i>L.lunaris</i>	<i>L.quadricornata</i>	<i>Lecane sp</i>	<i>L.ungulata</i>	<i>Macrochaetetes spp</i>	<i>Naupilus</i>	<i>P.quadricornis</i>	<i>P.dolichoptera</i>	<i>P.major</i>	<i>P.minor</i>	<i>P.remata</i>	<i>Rotifers</i>
<i>Bdelloidea</i>	.990**	-.264	-.462	.990**	.990**	.990**	-.462	.990**	-.264	.095	.990**	-.533	-.264	.990**	-.457	-.264
<i>B.forficula</i>	1.000**	-.333	-.333	1.000**	1.000**	1.000**	-.333	1.000**	-.333	.020	1.000**	-.533	-.333	1.000**	-.577	-.333
<i>B.calyciflorus</i>	1.000**	-.333	-.333	1.000**	1.000**	1.000**	-.333	1.000**	-.333	.020	1.000**	-.533	-.333	1.000**	-.577	-.333
<i>B.falcatus</i>	1.000**	-.333	-.333	1.000**	1.000**	1.000**	-.333	1.000**	-.333	.020	1.000**	-.533	-.333	1.000**	-.577	-.333
<i>C.gibba</i>	.479	.668	-.574	.479	.479	.479	-.574	.479	.668	.886	.479	.368	-.574	.479	.081	.668
<i>Collotheca sp</i>	-.333	-.333	1.000**	-.333	-.333	-.333	1.000**	-.333	-.333	-.514	-.333	.218	-.333	-.333	-.577	-.333
<i>C.adriatica</i>	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.441	-.333	-.533	1.000**	-.333	.577	-.333

<i>D.prionacis</i>	1.000**	-.333	-.333	1.000**	1.000**	1.000**	-.333	1.000**	-.333	.020	1.000**	-.533	-.333	1.000**	-.577	-.333
<i>E.porsildi</i>	.577	-.577	-.577	.577	.577	.577	-.577	.577	-.577	-.365	.577	-.923	.577	.577	0.000	-.577
<i>E.dilatata</i>	-.280	.998**	-.359	-.280	-.280	-.280	-.359	-.280	.998**	.953*	-.280	.831	-.359	-.280	.554	.998**
<i>F.opoliensis</i>	-.180	-.404	.987*	-.180	-.180	-.180	.987*	-.180	-.404	-.533	-.180	.138	-.404	-.180	-.699	-.404
<i>K.tecta</i>	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.333	-.441	-.333	-.533	1.000**	-.333	.577	-.333
<i>K.tropica</i>	1.000**	-.333	-.333	1.000**	1.000**	1.000**	-.333	1.000**	-.333	.020	1.000**	-.533	-.333	1.000**	-.577	-.333
<i>L.arcula</i>		-.333	-.333	1.000**	1.000**	1.000**	-.333	1.000**	-.333	.020	1.000**	-.533	-.333	1.000**	-.577	-.333
<i>L.bulla</i>			-.333	-.333	-.333	-.333	-.333	-.333	1.000**	.935	-.333	.848	-.333	-.333	.577	1.000**
<i>L.hamata</i>				-.333	-.333	-.333	1.000**	-.333	-.333	-.514	-.333	.218	-.333	-.333	-.577	-.333
<i>L.luna</i>					1.000**	1.000**	-.333	1.000**	-.333	.020	1.000**	-.533	-.333	1.000**	-.577	-.333
<i>L.lunaris</i>						1.000**	-.333	1.000**	-.333	.020	1.000**	-.533	-.333	1.000**	-.577	-.333
<i>L.quadridentata</i>							-.333	1.000**	-.333	.020	1.000**	-.533	-.333	1.000**	-.577	-.333
<i>Lecane sp</i>								-.333	-.333	-.514	-.333	.218	-.333	-.333	-.577	-.333
<i>L.ungulata</i>									-.333	.020	1.000**	-.533	-.333	1.000**	-.577	-.333
<i>Macrochaetus sp</i>										.935	-.333	.848	-.333	-.333	.577	1.000**
<i>Naupilus</i>											.020	.679	-.441	.020	.428	.935
<i>P.quadricornis</i>												-.533	-.333	1.000**	-.577	-.333
<i>P.dolichoptera</i>													-.533	-.533	.273	.848
<i>P.major</i>														-.333	.577	-.333
<i>P.minor</i>															-.577	-.333
<i>P.remata</i>																.577

	<i>Synchaeta spp</i>	<i>T.rattus</i>	<i>T.similis</i>	<i>T.patina</i>	<i>T.tetractis</i>	<i>A.curvirostris</i>	<i>A.angustatus</i>	<i>Camptocercus sp</i>	<i>E.longispina</i>	<i>Cladocera</i>	Air Temperature	Water Temperature	pH	E.C	TDS	Salinity	D.O	Total No of Taxa
<i>Bdelloidea</i>	-.264	-.264	.990**	.629	.857	.629	-.541	-.264	-.213	-.302	.758	.629	.515	-.024	-.069	-.287	-.461	.860
<i>B.forficula</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829
<i>B.calyciflorus</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829
<i>B.falcatus</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829
<i>C.gibba</i>	-.574	.668	.479	-.081	.148	-.081	.395	.668	.418	.642	.726	.677	.896	-.831	-.873	-.943	-.645	.888
<i>Collotheca sp</i>	-.333	-.333	-.333	-.577	-.541	-.577	.110	-.333	.451	-.271	-.072	.085	-.168	.190	.318	.329	-.206	-.540
<i>C.adriatica</i>	1.000**	-.333	-.333	.577	.257	.577	-.451	-.333	-.877	-.364	-.828	-.916	-.873	.702	.623	.723	.978*	-.540
<i>D.prionacis</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829

<i>E.porsildi</i>	.577	-.577	.577	1.000**	.937	1.000**	-.873	-.577	-.893	-.630	-.029	-.202	-.305	.612	.520	.407	.395	.250	
<i>E.dilatata</i>	-.359	.998**	-.280	-.554	-.502	-.554	.882	.998**	.582	.994**	.154	.191	.561	-.913	-.937	-.828	-.285	.305	
<i>F.opoliensis</i>	-.404	-.404	-.180	-.506	-.426	-.506	.022	-.404	.445	-.343	.058	.204	-.088	.199	.328	.301	-.302	-.224	
<i>K.tecta</i>	1.000**	-.333	-.333	.577	.257	.577	-.451	-.333	-.877	-.364	-.828	-.916	-.873	.702	.623	.723	.978*	-.540	
<i>K.tropica</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829	
<i>L.arcula</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829	
<i>L.bulla</i>	-.333	1.000**	-.333	-.577	-.541	-.577	.898	1.000**	.580	.998**	.105	.148	.521	-.897	-.919	-.799	-.250	.251	
<i>L.hamata</i>	-.333	-.333	-.333	-.577	-.541	-.577	.110	-.333	.451	-.271	-.072	.085	-.168	.190	.318	.329	-.206	-.540	
<i>L.luna</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829	
<i>L.lunaris</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829	
<i>L.quadridentata</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829	
<i>Lecane sp</i>	-.333	-.333	-.333	-.577	-.541	-.577	.110	-.333	.451	-.271	-.072	.085	-.168	.190	.318	.329	-.206	-.540	
<i>L.ungulata</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829	
<i>Macrochaetus sp</i>	-.333	1.000**	-.333	-.577	-.541	-.577	.898	1.000**	.580	.998**	.105	.148	.521	-.897	-.919	-.799	-.250	.251	
<i>Naupilus</i>	-.441	.935	.020	-.365	-.243	-.365	.728	.935	.521	.919	.388	.384	.727	-.934	-.973*	-.930	-.427	.576	
<i>P.quadricornis</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829	
<i>P.dolichoptera</i>	-.533	.848	-.533	-.923	-.865	-.923	.992**	.848	.855	.881	.068	.201	.444	-.822	-.772	-.642	-.375	-.044	
<i>P.major</i>	1.000**	-.333	-.333	.577	.257	.577	-.451	-.333	-.877	-.364	-.828	-.916	-.873	.702	.623	.723	.978*	-.540	
<i>P.minor</i>	-.333	-.333	1.000**	.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829	
<i>P.remata</i>	.577	.577	-.577	0.000	-.246	0.000	.388	.577	-.257	.549	-.626	-.665	-.305	-.168	-.256	-.066	.630	-.250	
<i>Rotifers</i>	-.333	1.000**	-.333	-.577	-.541	-.577	.898	1.000**	.580	.998**	.105	.148	.521	-.897	-.919	-.799	-.250	.251	
<i>Synchaeta sp</i>		-.333	-.333	.577	.257	.577	-.451	-.333	-.877	-.364	-.828	-.916	-.873	.702	.623	.723	.978*	-.540	
<i>T.rattus</i>			-.333	-.577	-.541	-.577	.898	1.000**	.580	.998**	.105	.148	.521	-.897	-.919	-.799	-.250	.251	
<i>T.similis</i>				.577	.825	.577	-.558	-.333	-.155	-.364	.795	.683	.521	.005	-.023	-.253	-.522	.829	
<i>T.patina</i>					.937	1.000**	-.873	-.577	-.893	-.630	-.029	-.202	-.305	.612	.520	.407	.395	.250	
<i>T.tetractis</i>						.937	-.842	-.541	-.684	-.590	.319	.152	.011	.425	.350	.174	.050	.526	
<i>A.curvirostris</i>							-.873	-.577	-.893	-.630	-.029	-.202	-.305	.612	.520	.407	.395	.250	
<i>A.angustatus</i>								.898	.789	.925	.012	.130	.414	-.828	-.793	-.650	-.297	-.040	
<i>Camptocercus sp</i>									.580	.998**	.105	.148	.521	-.897	-.919	-.799	-.250	.251	
<i>E.longispina</i>										.624	.474	.616	.684	-.789	-.699	-.683	-.761	.186	
<i>Cladocera</i>											.103	.157	.520	-.902	-.916	-.793	-.270	.219	
Air Temperature													.982*	.903	-.515	-.490	-.681	-.927	.878
Water Temperature														.913	-.567	-.520	-.693	-.980*	.788
pH															-.832	-.816	-.926	-.916	.844

<b>E.C</b>	<b>.991**</b>	<b>.964*</b>	<b>.652</b>	<b>-.527</b>
<b>TDS</b>		<b>.971*</b>	<b>.589</b>	<b>-.568</b>
<b>Salinity</b>			<b>.728</b>	<b>-.734</b>
<b>D.O</b>				<b>-.684</b>

Appendix (4): The Correlation between some environmental parameters , species density and number of taxa among station four

	<i>B.forficula</i>	<i>B.calyciflorus</i>	<i>C.gibba</i>	<i>Collothea sp</i>	<i>C.adriatica</i>	<i>E.dilatata</i>	<i>F.opoliensis</i>	<i>F.terminalis</i>	<i>K.cochlearis</i>	<i>K.tecta</i>	<i>K.valga</i>
<i>Bdelloidea</i>	.158	.413	.410	.413	.413	.516	-.984*	-.984*	-.953*	.158	-.984*
<i>B.forficula</i>		-.333	.893	-.333	-.333	.394	-.333	-.333	-.410	1.000**	-.333
<i>B.calyciflorus</i>			.123	-.333	1.000**	-.565	-.333	-.333	-.166	-.333	-.333
<i>C.gibba</i>				-.462	.123	.192	-.554	-.554	-.557	.893	-.554
<i>Collothea sp</i>					-.333	.735	-.333	-.333	-.410	-.333	-.333
<i>C.adriatica</i>						-.565	-.333	-.333	-.166	-.333	-.333
<i>E.dilatata</i>							-.565	-.565	-.694	.394	-.565
<i>F.opoliensis</i>								1.000**	.985*	-.333	1.000**
<i>F.terminalis</i>									.985*	-.333	1.000**
<i>K.cochlearis</i>										-.410	.985*
<i>K.tecta</i>											-.333

	<i>L.bulla</i>	<i>L.closteroeca</i>	<i>L.elsa</i>	<i>L.hamata</i>	<i>L.lunaris</i>	<i>Lecane sp</i>	<i>L.ungulata</i>	<i>Lepadella spp</i>	<i>M.angusta</i>	<i>Naupilus</i>	<i>P.quadricornis</i>	<i>P.dolichopectera</i>	<i>P.major</i>	<i>Rotifers</i>
<i>Bdelloidea</i>	-.034	.413	.158	.413	.413	.494	.158	.413	.158	.617	-.715	.038	.413	.266
<i>B.forficula</i>	.883	-.333	1.000**	-.333	-.333	.577	1.000**	-.333	1.000**	-.429	.577	.511	-.333	-.404
<i>B.calyciflorus</i>	-.022	1.000**	-.333	-.333	1.000**	.577	-.333	1.000**	-.333	-.002	-.577	.511	1.000**	.987*
<i>C.gibba</i>	.897	.123	.893	-.462	.123	.880	.893	.123	.893	-.402	.293	.752	.123	.036
<i>Collothea sp</i>	-.732	-.333	-.333	1.000**	-.333	-.577	-.333	-.333	-.333	.942	-.577	-.892	-.333	-.404
<i>C.adriatica</i>	-.022	1.000**	-.333	-.333	1.000**	.577	-.333	1.000**	-.333	-.002	-.577	.511	1.000**	.987*
<i>E.dilatata</i>	-.079	-.565	.394	.735	-.565	-.148	.394	-.565	.394	.610	-.148	-.502	-.565	-.684
<i>F.opoliensis</i>	-.129	-.333	-.333	-.333	-.333	-.577	-.333	-.333	-.333	-.511	.577	-.129	-.333	-.180
<i>F.terminalis</i>	-.129	-.333	-.333	-.333	-.333	-.577	-.333	-.333	-.333	-.511	.577	-.129	-.333	-.180
<i>K.cochlearis</i>	-.139	-.166	-.410	-.410	-.166	-.498	-.410	-.166	-.410	-.535	.498	-.042	-.166	-.008
<i>K.tecta</i>	.883	-.333	1.000**	-.333	-.333	.577	1.000**	-.333	1.000**	-.429	.577	.511	-.333	-.404

<i>K.valga</i>	-129	-.333	-.333	-.333	-.333	-.333	-.577	-.333	-.333	-.333	-.511	.577	-.129	-.333	-.180
<i>L.bulla</i>		-.022	.883	-.732	-.022	.746	.883	-.022	.883	-.752	.653	.832	-.022	-.044	
<i>L.closterocerca</i>			-.333	-.333	1.000**	.577	-.333	1.000**	-.333	-.002	-.577	.511	1.000**	.987*	
<i>L.elsa.</i>				-.333	-.333	.577	1.000**	-.333	1.000**	-.429	.577	.511	-.333	-.404	
<i>L.hamata</i>					-.333	-.577	-.333	-.333	-.333	.942	-.577	-.892	-.333	-.404	
<i>L.lunaris</i>						.577	-.333	1.000**	-.333	-.002	-.577	.511	1.000**	.987*	
<i>Lecane sp</i>							.577	.577	.577	-.374	0.000	.884	.577	.506	
<i>L.ungulata</i>								-.333	1.000**	-.429	.577	.511	-.333	-.404	
<i>Lepadella sp</i>									-.333	-.002	-.577	.511	1.000**	.987*	
<i>M.angusta</i>										-.429	.577	.511	-.333	-.404	
<i>Naupilus</i>											-.814	-.746	-.002	-.088	
<i>P.quadricornis</i>												.330	-.577	-.506	
<i>P.dolichoptera</i>													.511	.511	
<i>P.major</i>														.987*	

	<i>Synchaeta spp</i>	<i>T.rattus</i>	<i>T.patina</i>	<i>T.tetractis</i>	<i>A.angustatus</i>	<i>A.guttata</i>	<i>A.exigua</i>	<i>C.runcinatus</i>	<i>D.magna</i>	<i>Daphniidae</i>	<i>E.longispina</i>	<i>Cladocera</i>	Air Temperature	Water Temperature	pH	E.C	TDS	Salinity	D.O	Total No of Taxa
<i>Bdelloidea</i>	.413	-.442	-.550	.449	.413	.158	-.984*	-.984*	.500	.413	.315	.684	-.090	-.197	.468	-.452	-.446	-.608	.320	.451
<i>B.forficula</i>	-.333	.093	.554	-.021	-.333	1.000**	-.333	-.333	-.404	-.333	-.883	-.552	.799	.728	-.132	-.046	.112	-.066	-.507	.463
<i>B.calyciflorus</i>	1.000**	-.969*	-.059	.949	-.333	-.333	-.333	-.333	-.180	-.333	.434	.312	-.827	-.888	-.423	.531	.457	.385	.981*	.679
<i>C.gibba</i>	.123	-.359	.508	.424	-.462	.893	-.554	-.554	-.461	-.462	-.697	-.386	.453	.346	-.291	.156	.286	.061	-.070	.804
<i>Collotheca sp</i>	-.333	.471	-.919	-.502	1.000**	-.333	-.333	-.333	.987*	1.000**	.589	.792	.088	.105	.979*	-.925	-	-.911	-.261	-.626
<i>C.adriatica</i>	1.000**	-.969*	-.059	.949	-.333	-.333	-.333	-.333	-.180	-.333	.434	.312	-.827	-.888	-.423	.531	.457	.385	.981*	.679
<i>E.dilatata</i>	-.565	.526	-.498	-.505	.735	.394	-.565	-.565	.672	.735	-.061	.375	.660	.626	.859	-.935	-.869	-.936	-.619	-.278
<i>F.opoliensis</i>	-.333	.405	.423	-.425	-.333	-.333	1.000**	1.000**	-.404	-.333	-.139	-.552	-.060	.055	-.423	.440	.405	.592	-.213	-.515
<i>F.terminalis</i>	-.333	.405	.423	-.425	-.333	-.333	1.000**	1.000**	-.404	-.333	-.139	-.552	-.060	.055	-.423	.440	.405	.592	-.213	-.515
<i>K.cochlearis</i>	-.166	.246	.432	-.271	-.410	-.410	.985*	.985*	-.455	-.410	-.067	-.520	-.214	-.105	-.520	.557	.507	.690	-.043	-.415
<i>K.tecta</i>	-.333	.093	.554	-.021	-.333	1.000**	-.333	-.333	-.404	-.333	-.883	-.552	.799	.728	-.132	-.046	.112	-.066	-.507	.463
<i>K.valga</i>	-.333	.405	.423	-.425	-.333	-.333	1.000**	1.000**	-.404	-.333	-.139	-.552	-.060	.055	-.423	.440	.405	.592	-.213	-.515
<i>L.bulla</i>	-.022	-.221	.831	.288	-.732	.883	-.129	-.129	-.768	-.732	-.901	-.752	.500	.433	-.577	.427	.562	.393	-.188	.690
<i>L.closterocerca</i>	1.000**	-.969*	-.059	.949	-.333	-.333	-.333	-.333	-.180	-.333	.434	.312	-.827	-.888	-.423	.531	.457	.385	.981*	.679

<i>L.elsa</i>	-.333	.093	.554	-.021	-.333	1.000**	-.333	-.333	-.404	-.333	-.883	-.552	.799	.728	-.132	-.046	.112	-.066	-.507	.463
<i>L.hamata</i>	-.333	.471	-.919	-.502	1.000**	-.333	-.333	-.333	.987*	1.000**	.589	.792	.088	.105	.979*	-.925	-	-.911	-.261	-.626
<i>L.lunaris</i>	1.000**	-.969*	-.059	.949	-.333	-.333	-.333	-.333	-.180	-.333	.434	.312	-.827	-.888	-.423	.531	.457	.385	.981*	.679
<i>Lecane sp</i>	.577	-.759	.429	.803	-.577	.577	-.577	-.577	-.506	-.577	-.389	-.208	-.024	-.139	-.481	.420	.492	.276	.411	.989*
<i>L.ungulata</i>	-.333	.093	.554	-.021	-.333	1.000**	-.333	-.333	-.404	-.333	-.883	-.552	.799	.728	-.132	-.046	.112	-.066	-.507	.463
<i>Lepadella sp</i>	1.000**	-.969*	-.059	.949	-.333	-.333	-.333	-.333	-.180	-.333	.434	.312	-.827	-.888	-.423	.531	.457	.385	.981*	.679
<i>M.angusta</i>	-.333	.093	.554	-.021	-.333	1.000**	-.333	-.333	-.404	-.333	-.883	-.552	.799	.728	-.132	-.046	.112	-.066	-.507	.463
<i>Naupilus</i>	-.002	.150	-.990*	-.187	.942	-.429	-.511	-.511	.983*	.942	.753	.948	-.170	-.179	.897	-.809	-.880	-.850	.059	-.395
<i>P.quadricornis</i>	-.577	.431	.846	-.387	-.577	.577	.577	.577	-.699	-.577	-.885	-.956*	.640	.678	-.481	.342	.448	.456	-.624	-.045
<i>P.dolichoptera</i>	.511	-.690	.763	.732	-.892	.511	-.129	-.129	-.845	-.892	-.552	-.568	-.064	-.137	-.826	.762	.830	.674	.377	.609
<i>P.major</i>	1.000**	-.969*	-.059	.949	-.333	-.333	-.333	-.333	-.180	-.333	.434	.312	-.827	-.888	-.423	.531	.457	.385	.981*	.679
<i>Rotifers</i>	.987*	-.943	.010	.919	-.404	-.404	-.180	-.180	-.256	-.404	.429	.233	-.873	-.918	-.512	.627	.545	.501	.988*	.622
<i>Synchaeta sp</i>		-.969*	-.059	.949	-.333	-.333	-.333	-.333	-.180	-.333	.434	.312	-.827	-.888	-.423	.531	.457	.385	.981*	.679
<i>T.rattus</i>			-.115	-.997**	.471	.093	.405	.405	.329	.471	-.211	-.153	.670	.750	.516	-.582	-.545	-.426	-.905	-.839
<i>T.patina</i>			.160	-.919	.554	.423	.423	-.968*	-.919	-.836	-.964*	.288	.286	-.845	.735	.826	.772	-.142	.430	
<i>T.tetractis</i>				-.502	-.021	-.425	-.425	-.365	-.502	.146	.108	-.615	-.701	-.533	.586	.561	.429	.873	.876	
<i>A.angustatus</i>					-.333	-.333	-.333	.987*	1.000**	.589	.792	.088	.105	.979*	-.925	-	-.911	-.261	-.626	
<i>A.guttata</i>						-.333	-.333	-.404	-.333	-.883	-.552	.799	.728	-.132	-.046	.112	-.066	-.507	.463	
<i>A.exigua</i>							1.000**	-.404	-.333	-.139	-.552	-.060	.055	-.423	.440	.405	.592	-.213	-.515	
<i>C.uncinatus</i>								-.404	-.333	-.139	-.552	-.060	.055	-.423	.440	.405	.592	-.213	-.515	
<i>D.magna</i>									.987*	.687	.878	-.047	-.039	.950*	-.876	-.939	-.886	-.108	-.540	
<i>Daphniidae</i>										.589	.792	.088	.105	.979*	-.925	-	-.911	-.261	-.626	
<i>E.longispina</i>											.874	-.753	-.730	.426	-.252	-.397	-.295	.562	-.309	
<i>Cladocera</i>												-.447	-.469	.712	-.589	-.685	-.669	.373	-.192	
Air Temperature													.992**	.267	-.437	-.299	-.365	-.920	-.162	
Water Temperature														.269	-.433	-.304	-.340	-.960*	-.270	
pH															-.983*	-.999*	-.972*	-.385	-.558	
E.C																.987*	.983*	.519	.519	
TDS																	.972*	.420	.572	
Salinity																		.392	.372	
D.O																				-.528

**Appendix (5): The Correlation between some environmental parameters , species density and number of taxa among station five**

	<i>B.forficula</i>	<i>B.rubens</i>	<i>B.angularis</i>	<i>B.budapestinensis</i>	<i>B.calyciflorus</i>	<i>B.quadridentatus</i>	<i>C.adriatica</i>	<i>D.prionacis</i>	<i>E.porsildi</i>	<i>E.dilatata</i>	<i>F.opoliensis</i>	<i>H.brehmi</i>	<i>K.cochlearis</i>	<i>K.recta</i>
<i>Bdelloidea</i>	-.530	-.530	-.530	-.530	-.103	.411	-.602	-.981*	-.602	-.228	-.530	.411	.411	.411
<i>B.forficula</i>		1.000**	1.000**	1.000**	.577	-.333	-.333	.577	-.333	.744	1.000**	-.333	-.333	-.333
<i>B.rubens</i>			1.000**	1.000**	.577	-.333	-.333	.577	-.333	.744	1.000**	-.333	-.333	-.333
<i>B.angularis</i>				1.000**	.577	-.333	-.333	.577	-.333	.744	1.000**	-.333	-.333	-.333
<i>B.budapestinensis</i>					.577	-.333	-.333	.577	-.333	.744	1.000**	-.333	-.333	-.333
<i>B.calyciflorus</i>						.577	-.577	0.000	-.577	.975*	.577	.577	.577	.577
<i>B.quadridentatus</i>							-.333	-.577	-.333	.382	-.333	1.000**	1.000**	1.000**
<i>C.adriatica</i>								.577	1.000**	-.563	-.333	-.333	-.333	-.333
<i>D.prionacis</i>									.577	.157	.577	-.577	-.577	-.577
<i>E.porsildi</i>										-.563	-.333	-.333	-.333	-.333
<i>E.dilatata</i>											.744	.382	.382	.382
<i>F.opoliensis</i>												-.333	-.333	-.333
<i>H.brehmi</i>													1.000**	1.000**
<i>K.cochlearis</i>														1.000**

**Continue to Appendix (5):**

	<i>L.bulla</i>	<i>L.closteroerca</i>	<i>L.elsa</i>	<i>L.inermis</i>	<i>L.lunaris</i>	<i>L.nana</i>	<i>Nauplius</i>	<i>P.quadricornis</i>	<i>P.major</i>	<i>P.minor</i>	<i>Rotifers</i>
<i>Bdelloidea</i>	-.510	.968*	.166	-.602	.411	-.530	.391	-.530	.615	.721	-.530
<i>B.forficula</i>	.997**	-.522	.577	-.333	-.333	1.000**	-.289	1.000**	-.331	-.333	1.000**
<i>B.rubens</i>	.997**	-.522	.577	-.333	-.333	1.000**	-.289	1.000**	-.331	-.333	1.000**
<i>B.angularis</i>	.997**	-.522	.577	-.333	-.333	1.000**	-.289	1.000**	-.331	-.333	1.000**
<i>B.budapestinensis</i>	.997**	-.522	.577	-.333	-.333	1.000**	-.289	1.000**	-.331	-.333	1.000**
<i>B.calyciflorus</i>	.637	-.302	0.000	-.577	.577	.577	.615	.577	.548	-.577	.577

<i>B.quadridentatus</i>	-.262	.174	-.577	-.333	1.000**	-.333	.999**	-.333	.964*	-.333	-.333
<i>C. adriatica</i>	-.368	-.522	-.577	1.000**	-.333	-.333	-.355	-.333	-.533	-.333	-.333
<i>D.prionacis</i>	.545	-.905	0.000	.577	-.577	.577	-.558	.577	-.748	-.577	.577
<i>E.porsildi</i>	-.368	-.522	-.577	1.000**	-.333	-.333	-.355	-.333	-.533	-.333	-.333
<i>E.dilatata</i>	.792	-.389	.157	-.563	.382	.744	.425	.744	.359	-.563	.744
<i>F.opoliensis</i>	.997**	-.522	.577	-.333	-.333	1.000**	-.289	1.000**	-.331	-.333	1.000**
<i>H.brehmi</i>	-.262	.174	-.577	-.333	1.000**	-.333	.999**	-.333	.964*	-.333	-.333
<i>K.cochlearis</i>	-.262	.174	-.577	-.333	1.000**	-.333	.999**	-.333	.964*	-.333	-.333
<i>K.tecta</i>	-.262	.174	-.577	-.333	1.000**	-.333	.999**	-.333	.964*	-.333	-.333
<i>L.bulla</i>		-.521	.545	-.368	-.262	.997**	-.216	.997**	-.262	-.368	.997**
<i>L.closterocerca</i>			.302	-.522	.174	-.522	.151	-.522	.398	.870	-.522
<i>L.elsa</i>				-.577	-.577	.577	-.558	.577	-.374	.577	.577
<i>L.inermis</i>					-.333	-.333	-.355	-.333	-.533	-.333	-.333
<i>L.lunaris</i>						-.333	.999**	-.333	.964*	-.333	-.333
<i>L.nana</i>							-.289	1.000**	-.331	-.333	1.000**
<i>Naupilus</i>								-.289	.963*	-.355	-.289
<i>P.quadricornis</i>									-.331	-.333	1.000**
<i>P.major</i>										-.101	-.331
<i>P.minor</i>											-.333

	<i>Synchaeta spp</i>	<i>T.rattus</i>	<i>T.patina</i>	<i>T.tetractis</i>	<i>A.curvirostris</i>	<i>A.angustatus</i>	<i>Alona spp</i>	<i>A.verrucosa</i>	<i>Campocercus sp</i>	<i>D.magna</i>	<i>Daphniidae</i>	<i>E.longispina</i>	<i>Cladocera</i>	Air Temperature	Water Temperature	pH	E.C	TDS	Salinity	D.O	Total No of Taxa
<i>Bdelloidea</i>	-.166	-.619	.068	-.530	-.508	.991**	.103	-.602	-.530	-.486	.411	.843	.908	-.714	-.757	-.681	-.134	-.246	-.100	.807	-.045
<i>B.forficula</i>	-.577	-.189	.720	1.000**	.997**	-.527	-.577	-.333	1.000**	.988*	-.333	-.878	-.322	.825	.755	.223	-.009	.029	-.215	-.519	.671
<i>B.rubens</i>	-.577	-.189	.720	1.000**	.997**	-.527	-.577	-.333	1.000**	.988*	-.333	-.878	-.322	.825	.755	.223	-.009	.029	-.215	-.519	.671
<i>B.angularis</i>	-.577	-.189	.720	1.000**	.997**	-.527	-.577	-.333	1.000**	.988*	-.333	-.878	-.322	.825	.755	.223	-.009	.029	-.215	-.519	.671
<i>B.budapestinensis</i>	-.577	-.189	.720	1.000**	.997**	-.527	-.577	-.333	1.000**	.988*	-.333	-.878	-.322	.825	.755	.223	-.009	.029	-.215	-.519	.671
<i>B.calyciflorus</i>	0.000	.074	.892	.577	.642	.000	-	-.577	.577	.695	.577	-.265	-.270	.758	.728	.527	-.815	-.799	-.921	-.603	.978*
<i>B.quadridentatus</i>	.577	.274	.310	-.333	-.255	.527	-.577	-.333	-.333	-.185	1.000**	.572	.010	.050	.086	.385	-.931	-	-.849	-.177	.459
<i>C.adriatica</i>	.577	.738	-.822	-.333	-.371	-.624	.577	1.000**	-.333	-.401	-.333	-.153	-.613	-.073	.029	.385	.362	.443	.502	-.281	-.697

<i>D.prionacis</i>	0.000	.475	-.089	.577	.542	-.996**	0.000	.577	.577	.508	-.577	-.893	-.809	.652	.679	.527	.305	.409	.249	-.692	-.023	
<i>E.porsildi</i>	.577	.738	-.822	-.333	-.371	-.624	.577	1.000**	-.333	-.401	-.333	-.153	-.613	-.073	.029	.385	.362	.443	.502	-.281	-.697	
<i>E.dilatata</i>	-.157	.009	.925	.744	.796	-.143	-.975*	-.563	.744	.837	.382	-.456	-.308	.845	.801	.492	-.669	-.646	-.812	-.634	.983*	
<i>F.opoliensis</i>	-.577	-.189	.720	1.000**	.997**	-.527	-.577	-.333	1.000**	.988*	-.333	-.878	-.322	.825	.755	.223	-.009	.029	-.215	-.519	-.671	
<i>H.brehmi</i>	.577	.274	.310	-.333	-.255	.527	-.577	-.333	-.333	-.185	1.000**	.572	.010	.050	.086	.385	-.931	-	.952*	-.849	-.177	.459
<i>K.cochlearis</i>	.577	.274	.310	-.333	-.255	.527	-.577	-.333	-.333	-.185	1.000**	.572	.010	.050	.086	.385	-.931	-	.952*	-.849	-.177	.459
<i>K.tecta</i>	.577	.274	.310	-.333	-.255	.527	-.577	-.333	-.333	-.185	1.000**	.572	.010	.050	.086	.385	-.931	-	.952*	-.849	-.177	.459
<i>L.bulla</i>	-.545	-.172	.761	.997**	1.000**	-.497	-.637	-.368	.997**	.997**	-.262	-.854	-.329	.849	.779	.259	-.084	-.046	-.287	-.545	.723	
<i>L.closterocerc a</i>	-.302	-.717	-.055	-.522	-.520	.927	.302	-.522	-.522	-.516	.174	.779	.971*	-.812	-.863	-.836	.118	.004	.143	.928	-.213	
<i>L.elsa</i>	-	-.877	.443	.577	.542	.084	0.000	-.577	.577	.508	-.577	-.363	.522	.020	-.099	-.667	.493	.441	.300	.397	-.206	
<i>L.inermis</i>	1.000**	.577	.738	-.822	-.333	-.371	-.624	.577	1.000**	-.333	-.401	-.333	-.153	-.613	-.073	.029	.385	.362	.443	.502	-.281	-.697
<i>L.lunaris</i>	.577	.274	.310	-.333	-.255	.527	-.577	-.333	-.333	-.185	1.000**	.572	.010	.050	.086	.385	-.931	-	.952*	-.849	-.177	.459
<i>L.nana</i>	-.577	-.189	.720	1.000**	.997**	-.527	-.577	-.333	1.000**	.988*	-.333	-.878	-.322	.825	.755	.223	-.009	.029	-.215	-.519	.671	
<i>Naupilus</i>	.558	.269	.351	-.289	-.210	.509	-.615	-.355	-.289	-.139	.999**	.537	-.005	.092	.125	.402	-.946	-	.965*	-.873	-.206	.500
<i>P.quadricorni s</i>	-.577	-.189	.720	1.000**	.997**	-.527	-.577	-.333	1.000**	.988*	-.333	-.878	-.322	.825	.755	.223	-.009	.029	-.215	-.519	.671	
<i>P.major</i>	.374	.013	.390	-.331	-.256	.714	-.548	-.533	-.331	-.189	.964*	.658	.262	-.079	-.071	.144	-.860	-.909	-.804	.039	.477	
<i>P.minor</i>	-.577	-.823	-.208	-.333	-.371	.624	.577	-.333	-.333	-.401	-.333	.459	.924	-.803	-.869	-.993**	.579	.480	.561	.977*	-.433	
<i>Rotifers</i>	-.577	-.189	.720	1.000**	.997**	-.527	-.577	-.333	1.000**	.988*	-.333	-.878	-.322	.825	.755	.223	-.009	.029	-.215	-.519	.671	
<i>Synchaeta sp</i>		.877	-.443	-.577	-.542	-.084	0.000	.577	-.577	-.508	.577	.363	-.522	-.020	.099	.667	-.493	-.441	-.300	-.397	-.206	
<i>T.rattus</i>			-.365	-.189	-.170	-.547	-.074	.738	-.189	-.153	.274	-.126	-.862	.349	.463	.874	-.347	-.251	-.213	-.721	-.119	
<i>T.patina</i>				.720	.765	.126	-.892	-.822	.720	.800	.310	-.318	.062	.627	.545	.128	-.535	-.553	-.711	-.301	.966*	
<i>T.tetractis</i>					.997**	-.527	-.577	-.333	1.000**	.988*	-.333	-.878	-.322	.825	.755	.223	-.009	.029	-.215	-.519	.671	
<i>A.curvirostris</i>						-.495	-.642	-.371	.997**	.997**	-.255	-.851	-.329	.851	.781	.262	-.090	-.052	-.293	-.547	.727	
<i>A.angustatus</i>							.000	-.624	-.527	-.464	.527	.860	.850	-.648	-.684	-.581	-.263	-.370	-.223	.723	.040	
<i>Alona sp</i>								.577	-.577	-.695	-.577	.265	.270	-.758	-.728	-.527	.815	.799	.921	.603	-.978*	
<i>A.verrucosa</i>									-.333	-.401	-.333	-.153	-.613	-.073	.029	.385	.362	.443	.502	-.281	-.697	
<i>Camptocercus sp</i>										.988*	-.333	-.878	-.322	.825	.755	.223	-.009	.029	-.215	-.519	.671	
<i>D.magna</i>											-.185	-.823	-.334	.868	.800	.295	-.161	-.124	-.361	-.570	.773	
<i>Daphniidae</i>												.572	.010	.050	.086	.385	-.931	-	.952*	-.849	-.177	.459
<i>E.longispina</i>													.605	-.790	-.763	-.368	-.236	-.313	-.087	.634	-.314	
<i>Cladocera</i>														-.722	-.797	-.916	.230	.115	.203	.930	-.139	

Air Temperature	.992**	.730	-.409	-.331	-.530	-.900	.731
Water Temperature		.807	-.435	-.348	-.531	-.948	.676
pH			-.597	-.500	-.554	-.947	.366
E.C				.993*	.974*	.475	-.703
TDS					.970*	.373	-.700
Salinity						.501	-.845
D.O							-.493

**Appendix (6): The Correlation between some environmental parameters, total density, number of taxa and diversity indices among station one**

	Water Temperature	pH	EC	TDS	Salinity	DO	Total No. of Taxa	Total Density	Simpson Index	Shannon Index	Menhinick Index	Equitability Index	Berger-Parker Index	Margalef Index
Air Temperature	.991**	-.105	-.521	-.498	-.289	-.936	.939	.870	-.950	.934	.938	.963*	-.968*	.931
Water Temperature		-.206	-.515	-.481	-.255	-.972*	.925	.839	-.967*	.938	.976*	.970*	-.991**	.934
pH			-.499	-.564	-.740	.271	.149	.334	.086	.048	-.326	-.041	.233	.066
EC				.997**	.946	.588	-.782	-.851	.705	-.779	-.529	-.706	.569	-.787
TDS					.965*	.543	-.766	-.848	.671	-.754	-.482	-.677	.530	-.763
Salinity						.307	-.598	-.721	.461	-.567	-.239	-.473	.298	-.580
DO							-.908	-.815	.982*	-.948	-.997**	-.971*	.994**	-.943
Total No. of Taxa								.982*	-.969*	.991**	.885	.982*	-.927	.993**
Total Density									-.909	.956*	.782	.928	-.838	.960*
Simpson Index										-.991**	-.969*	-	.984*	-.988*
Shannon Index												.998**		
Menhinick Index											.927	.994**	-.954*	1.000**
Equitability Index												.958*	-.994**	.920
Berger-Parker Index													-.980*	.992**
Margalef Index														-.949

**Appendix (7): The Correlation between some environmental parameters, total density, number of taxa and diversity indices among station two**

	Water Temperature	pH	EC	TDS	Salinity	DO	Total No. of Taxa	Total Density	Simpson Index	Shannon Index	Menhinick Index	Equitability Index	Berger-Parker Index	Margalef Index
Air Temperature	.991**	.201	-.424	-.450	-.283	-.913	.898	.773	-.868	.880	.749	-.532	-.761	.901
Water Temperature		.166	-.404	-.425	-.254	-.951*	.831	.724	-.797	.812	.691	-.536	-.666	.836
pH			-.966*	-.963*	-.995**	-.319	.378	.759	-.103	.083	-.282	-.895	-.433	.140
EC				.999**	.986*	.553	-.520	-.866	.249	-.236	.132	.978*	.517	-.297
TDS					.983*	.560	-.558	-.887	.294	-.280	.088	.973*	.559	-.340
Salinity						.407	-.430	-.803	.154	-.137	.232	.933	.464	-.196
DO							-.698	-.733	.595	-.612	-.434	.698	.511	-.651
Total No. of Taxa								.877	-.957*	.953*	.777	-.537	-	.970*
Total Density									-.700	.691	.382	-.867	-.853	.736
Simpson Index										-.999**	-.925	.274	.925	-
Shannon Index											.932	-.269	-.913	.998**
Menhinick Index												.086	-.731	.907
Equitability Index													.482	-.332
Berger-Parker Index														-.925

**Appendix (8): The Correlation between some environmental parameters, total density, number of taxa and diversity indices among station three**

	Water Temperature	pH	EC	TDS	Salinity	DO	Total No. of Taxa	Total Density	Simpson Index	Shannon Index	Menhinick Index	Equitability Index	Berger-Parker Index	Margalef Index
<b>Air Temperature</b>	.982*	.903	-.515	-.490	-.681	-.927	.878	.622	-.926	.874	.369	.544	-.759	.696
<b>Water Temperature</b>		.913	-.567	-.520	-.693	-.980*	.788	.615	-.877	.774	.204	.690	-.799	.550
<b>pH</b>			-.832	-.816	-.926	-.916	.844	.882	-.947	.797	.061	.640	-.963*	.499
<b>EC</b>				.991**	.964*	.652	-.527	-.946	.682	-.439	.389	-.619	.948	-.065
<b>TDS</b>					.971*	.589	-.568	-.971*	.698	-.482	.316	-.509	.926	-.130
<b>Salinity</b>						.728	-.734	-.990*	.847	-.661	.133	-.538	.975*	-.327
<b>DO</b>							-.684	-.636	.816	-.655	-.003	-.818	.849	-.376
<b>Total No. of Taxa</b>								.744	-.971*	.995**	.565	.165	-.704	.878
<b>Total Density</b>									-.834	.673	-.081	.417	-.935	.363
<b>Simpson Index</b>										-.948	-.366	-.383	.846	-.747
<b>Shannon Index</b>											.641	.111	-.635	.919
<b>Menhinick Index</b>												-.540	.185	.891
<b>Equitability Index</b>													-.700	-.225
<b>Berger-Parker Index</b>														-.279

**Appendix (9): The Correlation between some environmental parameters, total density, number of taxa and diversity indices among station four**

	Water Temperature	pH	EC	TDS	Salinity	DO	Total No. of Taxa	Total Density	Simpson Index	Shannon Index	Menhinick Index	Equitability Index	Berger-Parker Index	Margalef Index
Air Temperature	.992**	.267	-.437	-.299	-.365	-.920	-.162	.288	.134	-.256	-.426	-.224	-.376	-.326
Water Temperature		.269	-.433	-.304	-.340	-.960*	-.270	.164	.228	-.341	-.458	-.339	-.255	-.397
pH			-.983*	-.999**	-.972*	-.385	-.558	.227	.702	-.755	-.954*	-.439	-.191	-.835
EC				.987*	.983*	.519	.519	-.302	-.653	.727	.961*	.418	.284	.819
TDS					.972*	.420	.572	-.218	-.713	.767	.964*	.458	.186	.847
Salinity						.392	.372	-.442	-.530	.605	.897	.255	.413	.712
DO							.528	.057	-.482	.577	.606	.588	.024	.609
Total No. of Taxa								.655	-.980*	.964*	.734	.986*	-.654	.915
Total Density									-.524	.433	-.031	.717	-.994**	.295
Simpson Index										-.991**	-.831	-.935	.539	-.964*
Shannon Index											.888	.920	-.440	.989*
Menhinick Index												.654	.020	.946
Equitability Index													-.701	.860
Berger-Parker Index														-.302

**Appendix (10): The Correlation between some environmental parameters, total density, number of taxa and diversity indices among station five**

	Water Temperature	pH	EC	TDS	Salinity	DO	Total No. of Taxa	Total Density	Simpson Index	Shannon Index	Menhinick Index	Equitability Index	Berger-Parker Index	Margalef Index
Air Temperature	.992**	.730	-.409	-.331	-.530	-.900	.731	.491	-.565	.705	.979*	.119	-.631	.901
Water Temperature		.807	-.435	-.348	-.531	-.948	.676	.434	-.498	.651	.997**	.021	-.578	.873
pH			-.597	-.500	-.554	-.947	.366	.200	-.190	.357	.853	-.324	-.315	.616
EC				.993**	.974*	.475	-.703	-.773	.697	-.726	-.474	-.419	.763	-.687
TDS					.970*	.373	-.700	-.801	.720	-.727	-.384	-.496	.773	-.647
Salinity						.501	-.845	-.886	.836	-.862	-.553	-.558	.888	-.812
DO							-.493	-.262	.297	-.471	-.969*	.224	.406	-.744
Total No. of Taxa								.951*	-.975*	.999**	.654	.737	-.989*	.947
Total Density									-.990*	.963*	.418	.861	-.986*	.818
Simpson Index										-.982*	-.472	-.862	.991**	-.854
Shannon Index											.630	.753	-.994**	.937
Menhinick Index												-.020	-.561	.862
Equitability Index													-.792	.484
Berger-Parker Index														-.902

## الخلاصة

تضمنت الدراسة اختيار خمس محطات موزعة على طول نهر الحلة لدراسة التنوع الإحيائي المظهري و الوراثة للهائمات الحيوانية ( الدولابيات و متفرعة اللوامس). مثلت المحطة الأولى نهر الحلة عند دخوله محافظة بابل عند قرية عانة ، و مثلت المحطة الثانية نهر الحلة قبل دخوله مركز المحافظة ( بالقرب من جسر بته) و صورت المحطة الثالثة نهر الحلة فور خروجه من مركز المحافظة بمنطقة الفارسي و المحطة الرابعة هي تفرع ينبع من نهر الحلة و يمر بقرية الغليس أما المحطة الخامسة فهي تفرع ينفصل عن نهر الحلة بالقرب من ناظم دورة و يمر عبر المنطقة بين الدولاب و الدبلة . كما جمعت عينات الهائمات الحيوانية ( الدولابيات و متفرعة اللوامس) فصلياً بوساطة شبكة هائمات قطر فوهتها 30 سم و حجم فتحاتها 50 مايكرومتر و تم عد الهائمات الحيوانية و صورت جميع العينات ثم شخصت باستخدام المفاتيح التصنيفية.

تناولت الدراسة الحالية تقييم بعض الصفات الفيزيائية و الكيميائية للمياه موسمياً خلال فترة الدراسة بين ربيع 2021 و شتاء 2022 و تضمنت هذه الصفات كل من درجة حرارة الهواء التي تراوحت بين ( 11.73 - 46.01 ) م° و حرارة الماء ( 10.51 - 35.21 ) م° و الأس الهيدروجيني (7.4 - 8.38) و التوصيلية الكهربائية (520.66 - 1375.6) مايكروسيمنز/سم و المواد الصلبة الذائبة الكلية (362.66 - 964) ملغم/لتر و الملوحة (347.5 - 691) ملغم/لتر و الأوكسجين المذاب (4.94 - 10.25) ملغم/لتر.

تباينت قيم كثافة الهائمات الحيوانية (الدولابيات و متفرعة اللوامس) بين أدنى قيمة (0.32) فرد/لتر في المحطة الخامسة في فصل الخريف وأعلى قيمة (1.386) فرد/لتر في المحطة الرابعة في فصل الصيف و لوحظ زيادة كثافة الدولابيات في فصل الصيف و متفرعة اللوامس في فصل الربيع ولأغلب محطات الدراسة. في حين سجل أقل عدد مصنفات (5.33) في المحطة الخامسة في فصل الخريف أما أعلى عدد مصنفات (14) فكان في فصل الصيف في المحطة الأولى و الثانية و الخامسة. كما بينت الدراسة الحالية تباين في كثافة الأنواع حيث تمثلت سيادة الدولابيات من خلال سيادة كل من *Bdelloidea* و *Trichocerca rattus* و *Euchlanis dilatata* و *Trichotria tetractis* و *Keratella cochlearis* و *Testudiella patina* أما سيادة متفرعة القرون فتمثلت بسيادة كل من *Eubosmina longispina* و *Acroperus angustatus* و *Daphnia magna* في جميع محطات الدراسة.

درست أيضا بعض أدلة التنوع الحياتي للمقارنة بين غزارة و وفرة الأنواع في المحطات خلال فترة الدراسة مثل دليل شانون للتنوع حيث تراوحت القيم بين ( 1.721 - 2.83 ) و دليل ماركليف ( 1.278 - 3.351 ) و دليل سمبسون ( 0.071 - 0.250 ) و دليل التكافؤ ( 0.717 - 0.913 ) و دليل مينهينك ( 0.343 - 0.776 ) و دليل بركر- باركر ( 0.13 - 0.4 ) و أشارت النتائج إلى أن اغلب المحطات تميزت بمعدل متوسط من التنوع الإحيائي كونها متأثرة بتغير الصفات الفيزيائية و الكيمائية للمياه. أما دليل جكارد للتشابه فظهر أعلى نسبة تشابه بين المحطتين الثانية و الثالثة و اقل نسبة تشابه كانت بين المحطتين الأولى و الخامسة.

تضمنت هذه الدراسة و تبعا للتصنيف المظهري تسجيل (69) وحدة تصنيفية من الدولابيات تضمنت ( 54 نوع و 12 جنس) تنتمي إلى 17 عائلة بالإضافة إلى تسجيل ثلاثة أنواع (*Lecane arcula* و *Horaella brehmi* و *Ascomorpha ovalis*) لأول مرة في العراق و أربعة أنواع (*Lecane quadridentata* و *Lecane tenuiseta* و *Lecane unguulate* و *Scaridium longicaudum*) لأول مرة في شط الحلة. في حين سجل ( 14 ) وحدة تصنيفية من متفرعة اللوامس و تضمنت ( 8 أنواع و 2 جنس) تنتمي إلى 4 عوائل بالإضافة إلى تسجيل خمسة أنواع ( *Acantholeberis curvirostris* و *Acroperus angustatus* و *Anthalona verrucosa* و *Alonella exigua* و *Eubosmina longispina*) لأول مرة في العراق. أما التصنيف الوراثي واعتمادا على تحليل التتابع الجيني و مقارنة النتائج مع المركز الوطني لمعلومات التقانة الإحيائية NCBI فقد سجل (83) نوع من الدولابيات بضمنها تسجيل نوعين (*Bryceella stylata* و *Ascomorpha ovalis*) لأول مرة في العراق و ثلاثة أنواع (*Lecane tenuista* و *Lecane unguulata* و *Scaridium longicaudum*) لأول مرة في شط الحلة كما سجل (40) نوع من متفرعة اللوامس بضمنها تسجيل سبعة أنواع ( *Daphnia dubia* و *Daphnia laevis* و *Daphnia ambigua* و *Daphnia pulicaria* و *Daphnia longicephala* و *Polyphemus pediculus* و *Acantholeberis curvirostris*) لأول مرة في العراق. كما تعد الدراسة الحالية الأولى على نهر الحلة فيما يتعلق في دراسة بيئة وتشخيص متفرعة القرون و كذلك تعد أول دراسة فيما يتعلق بدراسة التنوع الوراثي للهائمات الحيوانية .

كذلك تناولت الدراسة الحالية تحديد الشجرة التطورية للأنواع و المحطات المدروسة حيث أشارت النتائج إلى تقارب وراثي كبير ( 82% ) بين أنواع الدولابيات ( *Trichocerca*

*tenuior* و *Testudinella reflexa* و *Polyarthra dolichoptera* ) و تقارب وراثي كبير أيضا ( 92% ) بين أنواع متفرعة القرون ( *Scapholeberis mucronata* و *Podon* و *Evadne nordmanni* و *Cercopagis pengoi* و *Leptodora kindtii* و *leuckartii* و *Bythotrephes longimanus* و *Daphnia longicephala* و *Daphnia occidentalis* و *Daphnia ambigua* و *Daphniopsis truncata* ). كما أشارت النتائج إلى وجود تقارب بين المحطتين الأولى و الخامسة بنسبة ( 85% ) فيما يخص الدولابيات و نسبة ( 75 % ) فيما يخص متفرعة القرون.

أوضح التحليل الإحصائي SPSS و التحليل التوافقي CCA وجود ارتباطات موجبة وسالبة بين بعض العوامل البيئية المدروسة وأنواع الهائمات الحيوانية . إذ ارتبطت درجة حرارة الهواء و الماء ارتباط معنوي موجب في حين ارتبطت درجة الحرارة ارتباط معنوي عكسي مع الأوكسجين المذاب في الماء. كذلك أشارت النتائج إلى ارتباط التوصيلية الكهربائية و المواد الصلبة الذائبة الكلية و الملوحة ارتباط معنوي موجب بينما سجلت ارتباط معنوي سالب مع الأس الهيدروجيني . أما أنواع الدولابيات و متفرعة اللوامس فسجلت أغلبها ارتباط معنوي موجب مع كل من الأس الهيدروجيني و الأوكسجين المذاب في حين سجلت ارتباط معنوي سالب مع كل من التوصيلية الكهربائية و المواد الصلبة الذائبة الكلية و الملوحة. بالإضافة إلى ذلك ، تباينت علاقات الارتباط الأنواع مع بعضها البعض.

جمهورية العراق

وزارة التعليم العالي والبحث العلمي

جامعة بابل

كلية العلوم

قسم علوم الحياة



## التنوع الإحيائي للدولابيات و متفرعة اللوامس في نهر الحلة ، محافظة بابل - العراق

أطروحة

مقدمة إلى مجلس كلية العلوم - جامعة بابل لجزء من متطلبات نيل درجة  
الدكتوراه فلسفة في علوم الحياة

من قبل

وميض عادل كاظم محمد اليساري

بكالوريوس (2006) - علوم حياة - جامعة بابل

ماجستير (2012) - علوم حياة - جامعة بابل

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