Zooplankton for Monitoring and Assessing Lake Ecosystem Health

Subjects: Environmental Sciences

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For the sustainable use of lake ecosystem services—water resources, aquatic habitats for biodiversity conservation, and aesthetic values as waterfront space—ecosystem health assessments using biota are implemented as important national environmental monitoring projects. Zooplankton play a key role as an important linkage in the material circulation as secondary producers in lake ecosystems. At the same time, they influence the composition and biomass of other communities through biological interactions.

Keywords: zooplankton ; multi-metric index ; biotic indices ; biomass

1. The Role of Zooplankton in Lake Ecosystems

Lake ecosystems are enclosed areas with a long residence time and endogenous organic matter produced by macrophytes, sessile algae, and phytoplankton ^[1]. Externally introduced organic matter and pollutants are circulated within the lake through biological food webs ^[2]. Therefore, information related to energy and material flow, acquired by identifying changes in biological interactions among lake organisms within the food web, can be effectively used to represent the health status of the overall lake ecosystem ^[3].

Within lake food webs, zooplankton contains key species, which have a major influence on ecosystem function and stability. Zooplankton are a food source not only for planktivorous fish species, but also for juveniles of most fish species, including piscivorous fish, which are top predators, as well as grazers that control phytoplankton and bacterial populations [4][5][6]. Therefore, identifying changes in zooplankton communities (individual size, biomass, population, community structure, etc.) within lake ecosystems is an important task for effective lake management [Z][8].

In a broad sense, zooplankton may include various Protozoa, such as Ciliophora and Amoebozoa, but zooplankton that can be easily quantified using a 60 µm zooplankton net-a common collecting tool-includes Rotifera, Cladocera, and Copepoda ^[9]. Rotifera are the most common taxa, appearing in almost all lake and river ecosystems. They have a wide variety of forms, including thick-shelled, non-shelled, and soft-shelled species ^[10]. Rotifera have potential as biological indicators of a lake's trophic state, as they have a short life cycle and respond rapidly to environmental changes, making them well-adapted to dynamic environments [11][12]. Cladocera are Crustacea that swim using long tentacles and branched antennae that extend like arms [13]. They consume phytoplankton through filter-feeding; at the same time, they are a preferred food source for planktivorous fish. Through these biological interactions, Cladocera occupies an intermediate trophic level in the lake food web and plays an important role in material and energy circulation and water quality in lake ecosystems [14][15][16][17][18]. There are approximately 2800 freshwater Copepoda species, over a total of 13,000 described species [19]. Copepoda exhibit a very wide range of lifestyles, from predation to parasitism and, like Cladocera, serve as a food source for planktivorous fish. As such, these species also play an important role in material and energy circulation in aquatic ecosystems ^[20]. As mentioned above, zooplankton vary in size and swimming ability, are found at various trophic levels, and include species with different feeding characteristics and food selectivity. The distribution of zooplankton is influenced by abiotic limits, methods and means of distribution, and biological interactions [21] [22][23][24]. In the event of an ecosystem disturbance, the structure of the community changes rapidly, leading to a simplification of the community and the disappearance of species. However, the resting ability of all zooplankton allows them to remain inactive in each lake and reestablish themselves in the water column when favorable conditions return. Using these changes in community composition, it becomes possible to effectively evaluate the health of the lake ecosystem [12][25][26][27].

Owing to their sensitivity to environmental changes and their important role in the freshwater food web, zooplankton species have been used effectively to assess the trophic state of water bodies ^{[12][16][23][28]}. In aquatic ecosystem monitoring, zooplankton indices can be used to indicate the overall health of the system. Information about the lake

environment related to energy flow and material flow through biological interactions within the food web can be identified through zooplankton, (1) the biomass of important secondary producers; (2) changes in the microbial food web and grazing food web as the zooplankton community changes; (3) the quality improvement of total organic C circulating within the aquatic ecosystem food web; and (4) ecological shifts as a result of climate change or toxic substances ^{[29][30][31][32][33]} ^[34]. In addition, for biomanipulated lakes where planktivorous were removed to maximize the zooplankton grazing effect for water quality maintenance, indices regarding zooplankton composition (indices representing the recovery of Cladocera, particularly genus *Daphnia*) can be used as evidence to confirm the success of biomanipulation ^{[35][36]}.

2. Zooplankton Communities' Response Patterns to Environmental Change

Studies on zooplankton community-based indices to diagnose ecological and environmental status of lakes have long been conducted ^{[4][16]}. Broadly, zooplankton species are used as indicators of (i) eutrophication in a comprehensive sense, including water quality; (ii) watershed development and land use; and (iii) biological interactions (bottom-up and top-down pressures). Changes in zooplankton communities are measured in terms of the abundance, richness, and biomass of specific taxa. In recent studies on zooplankton indicators, the use of biomass has become more common ^[12] [26][27][28].

The responses of zooplankton communities to lake eutrophication can be classified in a taxon-specific manner. Zooplankton community characteristics suggested to be inversely proportional to the progress of eutrophication include species richness, average size, and the Calanoida/(Cyclopoida + Cladocera) abundance ratio ^[21]. Potential indicator species that appear in eutrophic lakes and increase in abundance with eutrophication include *Keratella cochlearis* f. *tecta*, *K. tropica*, *Brachionus budapestinensis*, and *B. calyciflorus*. In contrast, the abundances of *Conochilus unicornis*, *C. dossuarius*, *C. coenobasis*, and *Ascomorpha ovalis* increase as the trophic state is lowered, and these species have been suggested as oligotrophic indicators ^{[11][21][37]}.

Zooplankton community indicators that are reportedly inversely proportional to the total P concentration in a water body include species richness, body weight of planktonic Crustacea, the biomass ratio of *Daphnia* spp. among Crustacea, and the biomass and abundance of Calanoida among Copepoda ^[38]. Factors suggested to be proportional to the total P concentration include the proportions of biomass and the abundance of Cyclopoida among Copepoda. In an analysis of zooplankton samples from 146 lakes in the northeastern United States, the Ca concentration (hardness) of the water appeared to be proportional to the abundance of large Cladocera (e.g., *Daphnia pulex*, *D. pulicuria*, *D. schodleri*, and *D. galeata mendotae*), and inversely proportional to that of small Rotifera (<0.2 mm) ^[39].

It has also been suggested that increases in the total P concentration, Chlorophyll *a* concentration, and anthropogenic land use in a watershed can lead to a decrease in the species richness of Calanoida and large Cladocera, and the abundance of *Daphnia pulicaria*, a large crustacean. Increases in the abundance and richness of small Cladocera (e.g., Bosminidae, Chidoridae), Rotifera, Cyclopoida, Nauplius and *Skistodiaptomus pallidus* have also been reported ^{[24][40]}.

In the western United States, the biomass of large Cladocera (*D. pulex complex*) and Cyclopoida (*Diacyclops thomasi*) reportedly increases as lakes become deeper and cooler, and with an increase in unproductive land use and forested catchment in the watershed ^[41]. The biomass of small Cladocera (*Daphnia retrocurva*, *Diaphanosoma* spp., *Cydorus sphaericus*) and Cyclopoida (*Tropocyclops prasinus*) increases as watershed land becomes more productive and is affected by agriculture ^[41]. It has also been suggested that the body length of Cladocera and the biomass of Daphnidae decrease as water temperature increases and latitude decreases ^[41].

In terms of biological interactions, the specialist Cladocera and Calanoida species, commonly found in eutrophic lakes, were identified $^{[24]}$. The biomass of Rotifera and Cyclopoida reportedly increases as bottom-up pressures increase, including both direct and indirect pressures caused by nutrient salts (total P) and Chlorophyll *a*, while the biomass of Calanoida and Cladocera decreases $^{[42]}$.

The average body weight of Cladocera and the biomass ratio of zooplankton/phytoplankton reportedly decrease as the biomass of planktivorous fish increases ^[38]. At low latitudes, the predation pressure on zooplankton by planktivorous fish, such as salmon (*Oncorhynchus* spp.), lake trout (*Salvelinus* spp.), and gizzard shad (*Dorosoma cepedianum*), is high, affecting the size and biomass of Cladocera and Copepoda ^[41].

In previous studies related to zooplankton indicators, regardless of region or disturbance factors, the abundance or biomass of large Cladocera, Calanoida Copepoda, decreased as disturbances increased (total P concentration, predation

pressure of fish, commercial land use in watersheds, etc.). In contrast, the abundance or biomass of small Cladocera, Rotifera, and Cyclopoida Copepoda showed an increasing trend.

Although there is a common zooplankton community that responds to disturbance, zooplankton species may respond differently, depending on the area of occurrence and the disturbance. When developing a lake health evaluation index using the results from previous studies, an understanding of the zooplankton species inhabiting the target area is needed before a "good" index can be selected ^[43].

Alongside studies of changes in zooplankton communities in response to environmental change, research on groupspecific separations of response factors and the formation of formulae have also been conducted. In a previous study of the degree of eutrophication of a lake ecosystem, the number of species of specific zooplankton taxa (Rotifera, Copepoda, and Cladocera) and a eutrophication index were used together. Index values < 0.2 indicated oligotrophic lakes, greater than 0.2 and less than 1 were mesotrophic, greater than 1 and less than 4 were eutrophic, and greater than 4 indicated hypertrophic lakes $\frac{[44]}{2}$.

In summary, assessments of zooplankton community changes to evaluate the health of lake ecosystems have been conducted, but with variations in the zooplankton indicators, type of environmental disturbance, and zooplankton characteristics used. Inconsistencies in evaluation results using specific taxa have also been reported, with conflicting trends in the abundance or species composition ^[10]. Recently, a more comprehensive metric to evaluate a specific lake has been developed.

3. Zooplankton Indices for Freshwater Ecosystem Health Assessment

Recently, many multi-metric indices (MMI) for freshwater (especially lake) ecosystem health assessments have been developed and used. The multi-metric method using an integrated index responding to multiple co-varying stressors provides a more comprehensive assessment, and increases confidence in assessment $^{[45][46]}$. This is obtained by eliminating the possible dilemmas arising when only a single index or assemblage attributed to the different types of indices responds differently to diverse stressor types $^{[47]}$.

The European ECOFRAME project (for the ecological quality and functioning of shallow lake ecosystems with respect to the needs of the European Water Framework Directive 2003), through the development of a zooplankton index for assessing shallow lakes, has shown that the ratio of specific taxonomic groups in zooplankton communities is more effective than using absolute values (e.g., number of species and abundance) for ecosystem assessment [48]. In this index, the zooplankton species were categorized according to their body size, and simple coefficients calculated based on the proportion of large-bodied zooplankton biomass were used. Zooplankton body size is an important factor in determining their susceptibility to fish predation and a prey selection spectrum for phytoplankton assemblages. In ecologically healthy lakes, riparian aquatic plant communities function as effective shelters for large zooplankton, which can shed predatory pressure of fish [49]. Thus, the proportion of large Cladocera is higher in ecologically healthy lakes than in unhealthy lakes. species between 0.2 and 5 mm (Diaphanosoma, Moina, Leydigia leydigii, Holopedium gibberum, and Simocephalus vetulus) were classified as large Cladocera. The large bodied-zooplankton category generally includes Cladocera and Copepoda larger than 0.48 mm in size; specifically, among Cladocera species, genera Ceriodaphnia, Moina, Diaphanosoma, and Daphnia are included, while Bosmina, Alona, and Chydorus are excluded [50]. As zooplankton are grazers of phytoplankton, the "zooplankton biomass/Chlorophyll a concentration" equation was created to calculate the predation pressure of zooplankton, and the phytoplankton biomass was replaced by the Chlorophyll a concentration. This index calculates the impact of zooplankton on phytoplankton as the ratio of producers to primary consumers [48].

Kane et al. $\frac{[25]}{2}$ presented the Planktonic Index of Biotic Integrity(P-IBI) that can assess the health of Lake Erie at the catchment scale using appropriate metrics selected through factor analysis among candidate metrics suggested in the available literature. The study utilized seasonally varying scores to represent different environmental conditions throughout the year. The mean site score for each year was determined by summing the individual planktonic metric scores for June, July, and August and dividing by the number of metrics. Furthermore, the basin mean score was calculated by summing the average site scores and dividing by the number of sites. Subsequently, the lake-wide plankton IBI scores were obtained by summing the basin average scores and dividing by the number of basins. This index demonstrates a strong correspondence with traditional measures of lake nutrient status, reflecting the degree of eutrophication. The ratio of the total June phytoplankton biomass index scores (CB_{jk}) directly uses the percentage values of Cyanobacteria abundance, *Anabaena*, *Aphanizomonon*, and *Microcystis*, which reduce water quality as undesirable algae, and the usefulness of the phytoplankton community as a food source. The *Limnocalanus macrurus* abundance metric score (LM_{ik}) in July and the (Calanoida/[Cladocera + Cyclopoida]) ratio (RJ_{ik}) in June decline together, indicating

the food environment for the zooplankton community had deteriorated. Individual metrics using different plankton groups can provide information on benefit use impairment (BUI), which indicates the degree of eutrophication at different trophic levels. BUI can explain the damage to water resources by humans and assess the degree of impact on human health (e.g., water restriction), ecosystem function, or both ^[25].

Ejsmont-Karabin ^{[12][28]} developed an index to calculate TSI_{ROT} and TSI_{CR} (Indices of the trophic state of lakes), which are health status values of lakes using Rotifera and Crustacea. Through statistical analysis, the relationship between $TSI_{(SD+CHL)}$ —which is the average of TSI_{SD} (trophic state index calculated by transparency) and TSI_{CHL} (trophic state index calculated by the concentration of Chlorophyll a)—and zooplankton indicators that can indicate the trophic state of the lake (e.g., Rotifera abundance, Crustacea abundance, and Cyclopoida biomass) was identified, and a relational equation was presented. This equation indicates the trophic state of the lake using zooplankton. Lakes with TSI_{ROT} and TSI_{CR} values <45 were presented as oligotrophic, 45–55 as mesotrophic, 55–65 as eutrophic, and >65 as hypertrophic. *Keratella cochlearis*, which is the most common rotifer that appears in most lakes, shows a tendency to increase the abundance of *Keratella cochlearis* f. *tecta* type, in which the posterior spine disappears when the lake is eutrophic ^{[51][52]}. The following data are presented as equations that can explain the trophic state of lakes: percentage of form *tecta* in the population of *Keratella cochlearis*, Rotifera numbers, Rotifera biomass, percentage of bacterivores in the total number of Rotifera, ratio of biomass to number, percentage of species indicative of high trophy in the indicative Rotifera group's number, number of Crustacea, biomass of Cyclopoida, percentage of Cyclopoida biomass in the total biomass of Crustacea, ratio of the Cyclopoida biomass to the biomass of Cladocera, ratio of Cyclopoida to Calanoida numbers, and percentage of species indicative of high trophy in the indicative number of Crustacea I12I[28].

A more specific index focusing on the biological interaction between zooplankton and phytoplankton, the grazing potential (GP) index, combining the zooplankton and phytoplankton indices, has been proposed ^[26]. The GP value, formed using the sum of specific species, provides important information on food web function in an easy and cost-effective manner by combining zooplankton and phytoplankton metrics. Lakes with high GP values show that large Cladocera and Copepoda occupy a large proportion of the total zooplankton biomass. However, lakes with low GP values show a trend toward increased phytoplankton biomass, or increased population density of small zooplankton. The variables used in GP are identified at the genus level rather than at the species level, making it easier to identify and calculate than an index that uses species-level information. In the B_{ED} value calculation, a total phytoplankton index, a multiple (very good [1]; very bad [0]) was set in front of the variable to account for the relative edibility of phytoplankton ^[26].

On the other hand, index based on local biological assemblages and their interactions with environmental variables also has been developed. Ochocka [43] presented the Zooplankton Index of Polish Lakes' Assessment (ZIPLAs), which showed the strongest correlation with transparency. Of the 31 candidate metrics, five were selected for use in the index, showing the strongest correlations with TP, TN, transparency, and PCATOT (a cumulative nutrient load index calculated based on principal component analysis). The ecologically healthiest reference lakes were those that had no sources of pollution entering the water body, had the highest water quality status according to existing data, and had at least 80% natural land use within the watershed. Additionally, the dividing values of high (H), good (G), normal (M), poor (P), and very poor (B) indices were 75%, 50%, and 25% of the reference lake. CA/CY is the second most strongly correlated metric with TP for the ratio of Calnoida to Cyclopoida individual numbers, and the value of this index decreases with increasing eutrophication. Based on these results, it is clear that Calanoida prefer oligotrophic lakes. Zooplankton abundance (NZOL) is a commonly used index for assessing the trophic state of lakes [53][54][55]. This index is easy to calculate, decreases as TP concentrations increase, and is highly correlated with the nutrient status of the lake. The percentage of tecta in the population of Keratella cochlearis (TECTA) is an available indicator of the ecological status of lakes, particularly for lakes where Rotifera dominate and Cladocera and Copepoda are rare. Keratella cochlearis f. tecta, K. quadrata, Pompholyx sulcata, Filinia longicata, Anualeopsis fissa, Trichocerca pulchella, Brachionus undularis, and Brachionus dirsicornis were used as indicative species of high trophic levels. Rotifera species indicative of high trophic levels in the group that frequently occur in high-nutrient lakes can differ from country to country. Therefore, a list of national and regional indicator species should be used when assessing lakes using this index. The two diversity indices were the Shannon-Weaver index and the Margalef index; however, of the two indices, the Margalef (d) index, which represents the number of species relative to the total number of individuals, in contrast to the Shannon-Weaver index, which has a statistically highly significant correlation with environmental variables. Consequently, ZIPLAs decreased as the pollution level of the lakes increased [43].

Stamou ^[27] presented the Zoo-IQ index, which includes zooplankton biomass and body size information. The metrics used in this index are the total abundance of zooplankton (A_{zoo}) and total dry biomass of zooplankton (B_{zoo}), which are indices of the response of zooplankton to phytoplankton. Under eutrophic conditions characterized by increased nutrient content and bottom-up pressure, A_{zoo} and B_{zoo} exhibit a tendency to rise, potentially impacting zooplankton population

growth. The morphometric mean size of zooplankton (MW_{zoo}) provides insights into the functioning of the pelagic food web in lakes, where the presence of large Cladocera (particularly *Daphnia*) and Calanoida can effectively control the entire size range of phytoplankton and even induce clear water phases. The mean size was not measured directly, but was calculated as the ratio of dry biomass to abundance. The ratio of large Cladocera to total Cladocera abundance (R_{clad}) serves as an estimate of the changes in dominance among different functional groups, determined by Cladocera feeding mode. However, Rclad and MW_{zoo} decrease as eutrophication processes, reflecting changes in zooplankton groups' domination patterns, with small-bodied species dominating in eutrophic lakes with Cyanobacteria blooms, but also the predation pressure by fish on large- bodied species. The metric value of the healthiest lake in the dataset was set as the reference value; the metric value was rated as good (5) if it was similar to the reference value; normal (3) if different from the reference value. The sum of all metric scores was set as the Zoo-IQ value. This index is the result of synthesizing various stress factors, and can be used as an index to evaluate the decline in the general lake status ^[27].(**Table 1**)

 Table 1. Zooplankton metrics developed for lake health assessment.

Zooplankton Metrics	Description	Parameter	Ref.
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$Zoopl~lge:total=rac{x}{y}$	Ratio of large Cladocera frequently appearing in healthy lake	x = Large Cladocera (>0.5 mm) individual number y = Cladocera individual number	[<u>48]</u>
$Zoopl: phyto = rac{a}{b}$	Effects of zooplankton predation on phytoplankton	a = Cladocera and copepod biomass b = Chlorophyll <i>a</i> concentration	[48]
$P - IBI = rac{1}{B} \sum_{k=1}^{B} rac{1}{S} \sum_{j=1}^{S} rac{1}{M} (EA_{jk} + CB_{jk} + RJ_{jk} + LM_{jk} + RA_{jk} + RA_{jk} + ZB_{jk})$	Eutrophic < 3 3 ≤ Mesotrophic ≤ 4 4 < Oligotrophic	EA _{jk} = June biomass of edible algae taxa metric score CB _{jk} = June% <i>Mycrocystis</i> , <i>Anabaena</i> , <i>Aphanizomenon</i> of total phytoplankton biomass metric score RJ _{jk} = June zooplankton ratio (Calanoida/(Cladocera + Cyclopoida)) metric score LM _{jk} = July <i>Limnocalanus macrurus</i> density metric score RA _{jk} = August zooplankton ratio (Calanoida/(Cladocera + Cyclopoida)) metric score ZB _{jk} = August Crustacea zooplankton biomass metric score M = Number of metrics S = Number of sites (within a basin) B = Number of basins	[<u>25]</u>
$TSI_{ROT1} = 5.38Ln(N) + 19.28$ $TSI_{ROT2} = 5.63Ln(B) + 64.47$ $TSI_{ROT3} = 0.23BAC + 44.30$ $TSI_{ROT4} = 3.85(B:N)^{-0.318}$ $TSI_{ROT5} = 0.187TECTA + 50.38$ $TSI_{ROT6} = 0.203IHT + 40.0$	A lake trophic state evaluation index using the Rotifera community	N = Rotifera numbers (ind./L) B = Total biomass (mg w.wt./L) BAC = Percentage of bacterivores in total numbers (%) TECTA = Percentage of form <i>tecta</i> in the population of <i>Keratella cochlearis</i> (%) B:N = Ratio of biomass to numbers (mg w.wt./ind.) IHT = Percentage of species indicative of high trophy in the indicative group's numbers (%)	[12]
$TSI_{CR1} = 25.5N^{0.142}$ $TSI_{CR2} = 57.6B^{0.081}$ $TSI_{CR3} = 40.9CB^{0.097}$ $TSI_{CR4} = 58.3(CY/CL)^{0.071}$ $TSI_{CR7} = 5.08Ln(CY/CA) + 46.6$ $TSI_{CR8} = 43.8e^{0.004(IHT)}$	A lake trophic state evaluation index using the Crustacea community	N = Numbers of Crustacea (ind./L) B = Biomass of Cyclopoida (mg w.wt./L) CB = Percentage of Cyclopoida biomass in total biomass of Crustacea (%) CY/CL = Ratio of the Cyclopoida biomass to the biomass of Cladocera CY/CA = Ratio of Cyclopoida to Calanoida numbers IHT = Percentage of species indicative of high trophy in the indicative group's numbers (%)	[<u>16]</u>
$GP = rac{B_{ROT} + B_{CLAD} + 0.5B_{COP}}{B_{ED}}$	An index that measures the ecological water	B = dry biomass(mg/L) ROT = Rotifera CLAD = Cladocera	[<u>26</u>]

$B_{ED} = 0.3B_{CYANO} + 0.5B_{CHRYSO} + 1B_{CRYPTO}$ $+ 1B_{PRYMNESIO} + 0.7B_{DIATOMS} + 0B_{DINO}$ $+ 0.3B_{CONJ}$	quality of a lake by combining the dry biomass of plankton Low GP values: high zooplankton biomass dominated High GP values: increased phytoplankton biomass	COP = Copepoda CYANO = Cyanobacteria CHLORO = Chlorophyta CHRYSO = Chrysophyta CHYPTO = Cryptophyta PRYMNESIO = Prymnesiophyta DIATOMS = Bacillariophyta DINO = Dinophyta CONJ = Conjugatophyta	
$ZIPLAs = \frac{\frac{CA}{CY} + NZOL + TECTA + IHYROT + d}{5}$	$bad \le 0.189$ $0.189 < poor \le 0.376$ $0.377 \le moderate$ ≤ 0.565 $0.566 \le good \le 0.754$ $0.755 \le High$	CA/CY = Ratio of Calanoida to Cyclopoida individual numbers(ind./L) NZOL = Zooplankton abundance(ind./L) TECTA = Percentage of form <i>tecta</i> in the population of <i>Keratella cochlearis</i> (%) IHTROT = Percentage of species indicative of high trophy in the indicative group's number (%) D = Margalef's diversity index	<u>[43]</u>
$Zoo - IQ = A_{zoo} + B_{zoo} + MW_{zoo} + R_{Clad}$	Bad ≤ 6 6 < Poor ≤ 10 10 < Moderate \leq 14 14 < Good ≤ 18 18 < High	A _{Zoo} = Abundance (ind./L) B _{Zoo} = Biomass (μg/L) MW _{Zoo} = Mean body size (ind./μg) R _{Clad} = Cladocera ratio	[27]

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