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Response of the rotifer community to human-induced changes in the trophic state of a reservoir

by

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## Abstract

Human disturbance and nutrient runoff lead to water pollution, particularly in downstream waters and reservoirs. We hypothesized that increased human activity in summer would affect the trophic state of downstream reservoirs, affecting the interannual species composition of rotifers. We used long-term data for the Unmun Reservoir in South Korea (2009–2015), which is increasingly affected by human activity. The interannual variation of nitrogen and phosphorus levels was higher in summer and autumn, resulting in eutrophication. This led to a change in species composition of rotifers. Anuraeopsis fissa, Brachionus calyciflorus and Trichocerca gracilis were abundant in the most eutrophic state, while high densities of Ascomorpha ovalis and Ploesoma hudsoni were observed when nutrient concentrations were lower. The trophic state changes in the Unmun Reservoir were largely attributed to summer human activity in tributary streams. Our study location is typical of the stream network in South Korea and we assume that similar trophic state changes in reservoirs will be common. Changes in the density and species diversity of rotifers due to eutrophication indicate the need for active management and conservation, including the restriction of human activity around streams.

**Key words:** nutrient, eutrophication, selforganizing map, human activities, land cover, stream disturbance

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## Introduction

Rotifers play a crucial role in the functioning of freshwater ecosystems and are sentinel species for various changes in the aquatic environment (Oh et al. 2017). Factors such as water temperature, dissolved oxygen (DO) and nutrient concentrations are the main determinants of the spatio-temporal distribution of rotifer communities, which contribute to their population growth and fecundity. Thus, the response of rotifer communities to environmental factors has been central to limnological research. Empirical studies have shown that the effect of seasonal environmental factors on the success of rotifer communities can be complex (Castro et al. 2005; Elliott 1977; Nandini et al. 2005). Water temperature regulates the metabolic rate of rotifer communities and is related to sexual and asexual reproduction or germination from dormant eggs (Xi et al. 2004). Macrophytes cover the water surface of wetlands densely from spring to autumn, affecting the abundance and species diversity of rotifers (Große 1996). Areas with low dissolved oxygen content are often used by rotifer communities as a refuge to avoid fish predators (Larsson & Lampert 2011). Concentrations of nutrients (i.e. nitrogen and phosphorus) reduce water quality but also contribute to the growth and development of bacteria and/ or phytoplankton, which are a source of food for rotifer communities. Therefore, the density and diversity of rotifer communities is strongly affected by environmental conditions. Some empirical studies suggest that the species composition of rotifer communities varies depending on the trophic state of water (i.e. oligotrophic, mesotrophic, and eutrophic) and can be used for indirect water guality control based on habitat preferences of species that are dominant (Mäemets 1983; Singh et al. 2013; Sládeček 1983). For example, zooplankton species such as Brachionus and Polyarthra are abundant in high nutrient concentrations and are therefore considered to be biological indicators of eutrophication (Jafari et al. 2011; Singh et al. 2013; Suthar et al. 2010). This finding suggests that environmental factors must be considered in order to understand the seasonality of rotifer communities.

Rapid population growth as well as urban and industrial development in river basins have exposed waterways to increased environmental stress, causing water pollution and environmental degradation (Goethals & De Pauw 2001; Suthar et al. 2010). River basins generally support high human population densities through their favorable living conditions, including the availability of water for irrigation, industrial and drinking purposes, fertile land and efficiency of transportation. Many anthropogenic activities are part of larger land-cover change processes that can affect water quality in river ecosystems, as well as downstream estuarine and coastal waters. Human activity has a direct impact on the hydrological regime by changing physical and biological characteristics of land (Goethals & De Pauw 2001; Grady & Mullaney 1998; Søndergaard & Jeppesen 2007). Physical modifications such as urbanization, transport, farming (irrigation), land drainage, as well as channelization and damming change hydrological pathways and may change the water quality by modifying the water regime. For example, impervious surfaces created by urbanization produce large amounts of runoff and overland flow even at moderate rainfall intensities (Pappas et al. 2008). In addition, human activity changes water quality by adding substances and waste materials to the landscape. These include surface application of pesticides, herbicides, and fertilizers, as well as leaching from landfills, mine tailings, and irrigated farmland into groundwater and surface water.

Freshwater rotifer communities react to ecosystem changes over time and may reveal sources of pollution (Buikema et al. 1974; Snell et al. 1991). Therefore, the responses of these organisms are used as surrogates to assess the biological integrity of waterways through frequent water-chemistry measurements. The development of bioassessment techniques has led to the concept of ecological indicators used to evaluate the structure and function of ecosystems (Jackson et al. 2000; Gao et al. 2018). The main role of ecological indicators is to measure the ecosystem responses to anthropogenic disturbances as in organisms, i.e. deviations from ecological integrity (Frost et al. 1992). Rotifer species are valuable indicators of environmental conditions (Gannon & Stemberger 1978; Kuczynski 1987) because they respond directly and sensitively to diverse environmental changes occurring in aquatic ecosystems. Rotifers have a short life cycle and easily respond to rapidly changing environments (Allan 1976). Differences in the abundance of rotifers and species composition between lakes are determined mainly by "bottom-up" forces rather than "top-down" predatory interactions, as shown by studies carried out in 34 lakes in Ontario, Canada (Yoshida et al. 2003). Predicting the rotifer community responses to different hydrological forcings and their impact on freshwater ecosystems constitutes a major challenge for the management of freshwater ecosystems.

Research on the effects of environmental factors on rotifer distributions requires simultaneous consideration of miscellaneous data, but this is





often compromised due to the complexity of data. Empirical studies suggest that it has often been difficult to extract clear interactions from long-term data using previous analyses (Fielding 1999). Recent soft computing techniques for ecological analysis may provide an opportunity to address this problem. The self-organizing map (SOM) is one of the available non-linear data ordination processes. It extracts information from multi-dimensional data and maps it into a reduced dimensional space (Kohonen 1997). In recent research, the SOM has been used as a powerful and applicable method due to its easy visual interpretation (Chon et al. 1996) compared to other methods, such as principal component and correspondence analyses (Giraudel & Lek 2001).

In this study, we sought to elucidate the responses of rotifer communities to changes in environmental factors resulting from summer human activities in order to advance our current understanding of the seasonality of rotifers in freshwater ecosystems. In reservoir ecosystems, the trophic state is an important factor that determines the density and diversity of rotifers. The objective of this study was to elucidate: i) interannual changes in the structure of rotifer communities in relation to environmental changes, and (ii) responses of rotifers to changes in the trophic state as a result of human activity. To this end, we surveyed the Unmun Reservoir in South Korea, using a range of environmental factors and rotifer communities in the SOM model to identify the relationships between them. We measured the environmental factors and land cover characteristics of the main tributaries flowing into the reservoir to explain the interannual trophic state change in the Unmun Reservoir. We hypothesized that pollution in the main tributary would have a strong impact on the trophic state, which would be reflected by different rotifer species.

## Materials and methods

## **Study sites**

South Korea has a temperate climate and is governed by four distinct seasons (spring, summer, autumn, and winter). Thus, the succession of biological communities depends on seasonal characteristics. The study sites – the Unmun Reservoir, the Unmun Stream and the Sinwon Stream – are located in southeastern South Korea, in the upper reaches of the Miryang River (Fig. 1). The Unmun Reservoir is designed for water supply, with a basin area of 301.3 km<sup>2</sup>. The lowest and highest water levels are 122 m and 155 m, and the water filled area of the reservoir is 7.8 km<sup>2</sup> with a storage capacity of 1.35 Mt. The Unmun Reservoir



#### Figure 1

Map of the study sites. Location of the study sites in southeastern South Korea are indicated by a solid square (■) in the inset map of the Korean Peninsula. The main map shows the Unmun Reservoir and two tributary streams (Unmun and Sinwon). The sampling points are indicated by open circles (○).



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is surrounded mainly by forests, so the inflow of pollutants from the immediate catchment is low, but the inflow of pollutants from the major tributary streams is common. The Unmun and Sinwon streams are the main tributaries of the Unmun Reservoir. Their basin areas are 91.5 km<sup>2</sup> and 29.5 km<sup>2</sup>, respectively, and their lengths are 19.22 km and 11.48 km, respectively (Lee et al. 2005; RIMGIS: http://www.river.go.kr/). The dominant land covers along the two streams are clearly different and there are continuous inputs of non-point source pollution. The Unmun Temple is located in the middle reach of the Unmun Stream, and other human use and development is limited at this site because of the cultural characteristics of the temple. The area around the Sinwon Stream, on the other hand, has been developed for many years, with a number of residential, accommodation and commercial facilities (e.g. lodgings, camping sites, etc.). The Sinwon Stream is subjected to frequent human activities and inputs from summer tourists, which affect the water quality and distribution of aquatic organisms. It is important to identify changes in environmental factors and land cover patterns of the Sinwon and Unmun streams to understand how they affect the interannual distribution and species diversity of rotifer communities in the Unmun Reservoir and, consequently, how this affects the water quality.

### Monitoring strategy and data analysis

We investigated environmental factors in the Unmun Reservoir and its main tributary streams (Unmun and Sinwon) for seven years (2009-2015). The environmental factors, including water temperature, dissolved oxygen (DO), pH, conductivity, turbidity, total nitrogen (T-N), total phosphorus (T-P) and chlorophyll a were measured in water samples collected from sampling points located in the reservoir and two streams during monthly surveys (84 data records). Water samples were collected using a 20 I column water sampler and kept in the shade at ambient temperature until all analyses were completed. A DO meter (YSI Model 58, YSI Incorporated, Yellow Springs, OH, USA) was used to measure water temperature and DO; pH and conductivity were measured using a pH meter (Orion Model 250 A, Orion Research Inc., Boston, MA, USA) and a conductivity meter (YSI Model 152, Fisher Scientific, Hampton, NH, USA), respectively. To determine the chlorophyll *a* concentration, we filtered water samples through 0.45 µm mixed cellulose ester membrane filters (A045A047A; Advantech Co. Ltd., Taipei, Taiwan). Filter membranes were kept in 90% acetone in darkness at 20°C for 4 h. To improve the extraction, the cells were disintegrated for 2 min in

an ultrasonic bath. To remove cell debris and filter particles, the pigment extract was centrifuged at 5000 rpm for 5–10 min. The extinction was estimated at 600 and 750 nm by a spectrophotometer (Japan Fantec Research Institute, Shizuoka, Japan) using a 1 cm glass cuvette (Wetzel & Likens 2000). The concentration of chlorophyll *a* was estimated using the formula: Chlorophyll *a* = 11.403 × ( $A_{600} - A_{750}$ ) ×  $V_a$  ×  $V_b^{-1}$ , derived on the basis of a factor, where  $V_a$  is the extract volume (ml) and  $V_b$  is the sample volume (ml). T-N and T-P were also determined spectrophotometrically, based on the method of Wetzel & Likens (2000).

To determine the rotifer density, we collected 5 I water samples from the Unmun Reservoir during monthly surveys carried out in 2009–2015, using a 10 I column water sampler (84 data records). The sampler was placed vertically (approximately 0.5–1 m) in the water to collect rotifers. The sampled water was filtered through a plankton net with 32  $\mu$ m mesh size and the filtrate was preserved using sugar formalin (4% for formaldehyde; Haney & Hall 1973). Rotifers were identified at the species level and counted under a microscope (Model Axioskop 40, ZEISS; × 200 magnification) based on the identification keys published by Koste (1978) and Mizuno & Takahashi (1999).

We conducted a long-term data analysis to examine interannual changes in the density of rotifer communities in the Unmun Reservoir. Monthly data on rotifer communities were collected for 7 years (2009–2015). Seasonally (i.e. summer and autumn) summed values were used in the following analyses. Based on the long-term seasonal data, determination coefficients ( $r^2$ ) were calculated for the summer and autumn data to determine whether there is an annual trend in rotifer density changes. We also employed one-way ANOVA from the statistical package SPSS for Windows version 20 (IBM Corp., Armonk, NY, USA) to compare the environmental factors and the density of rotifer species in three clusters distinguished by SOM analysis.

### Land cover data analysis

In order to confirm the land use changes around the Unmun and Sinwon streams, land cover maps with a 500 m radius from the study sites were obtained from the Ministry of Environment in 2003, 2007 and 2013 and used to calculate the area of different land covers using ArcGIS 10.5 (ESRI, Redlands, Ca). We focused on land uses that indicated human disturbance, namely urbanized and agricultural areas. We applied a hierarchical approach, calculating first the area of the two first-level classes and then the ratio



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of second-level classes within each first-level class. The urbanized areas were divided into three second-level classes (residential, commercial, and cultural/ recreational areas) and the agricultural areas were divided into two second-level classes (paddy fields and fields). The area of different land cover classes was analyzed in three different periods. Through this process, we identified time-series land-cover changes in the two tributary streams above the Unmun Reservoir to confirm the level of human disturbance.

#### Self-organizing map (SOM)

The SOM was developed from the Kohonen network (Kohonen 1982; Kohonen 1997), which is an unsupervised learning algorithm (an artificial neural network). Artificial neural networks mimic the intellectual functions of animal brains, learning by example rather than by specific rules. The SOM is widely used as a tool for mapping multi-dimensional data into a two-dimensional representational space (Kohonen 1982). This mapping effectively captures the relationship between the input data, thus describing a topology-preserving representation of input similarities in terms of distances in the output space (Fig. 2). It is therefore possible to visually identify clusters on the map. The main advantage of such mapping is the ease with which the user can interpret the structure of the data.

The possibility of using neural networks in ecosystem simulations was first suggested by Odum (1994), based on the hypothesis that they may be useful in understanding life systems, including many aspects of ecology (Chon et al. 1996). The SOM



### Figure 2



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network is a competitive system in which the neurons (i.e. sample units) in the Euclidean map space compete with one another, converting non-linear relationships into simple geometric relationships. This algorithm is effective in clustering and visualizing essential features of complex data. It has a unique structure that allows multivariate data to be projected non-linearly onto a rectangular grid layout with a rectangular or hexagonal lattice.

In this study, eight dominant rotifer species and environmental factors were used as input variables in the SOM. During the training process, the number of nodes forming the SOM plane was determined as being adjacent to  $5 \times n$  (n indicates the number of samples, i.e. the number of sites in this study; Vesanto & Alhoniemi 2000). From a variety of map structures of different sizes, we selected the optimal structure based on the minimum values for guantization (QE) and topographic errors (TE) (Cèrèghino & Park 2009; Uriarte & Martín 2005). After selecting the optimal SOM structure, each parameter was projected onto the two-dimensional SOM plane with a gray scale gradient. The parameters were then clustered according to the calculated U-matrix. The gradient range was determined using the mean abundance of rotifers. We used MATLAB 6.1 (MathWorks, Inc, Natick, MA, USA) and the SOM Toolbox (Helsinki University of Technology, Helsinki) to develop the SOM model. We examined the differences between each cluster with ANOVA using SPSS version 20.

## Results

## Environmental factors and rotifer communities

Clear interannual variability of environmental factors was observed during the study period (Fig. 3). Water temperature increased from spring to summer, while from summer to winter it showed a downward trend. On the other hand, DO and pH were low in summer and high in winter. The highest values of conductivity, T-N, T-P, turbidity and chlorophyll a were found in autumn and then in summer. The seasonality of these environmental factors was similar throughout the survey period. The conductivity, T-N, T-P, turbidity and chlorophyll *a* showed an increasing trend during the study period.

The density of rotifers exhibited seasonality during the study period, with moderate abundance in spring (March-May) and summer (June-August), followed by frequent peaks in autumn (September-November). A total of 12 rotifer species were identified: Anuraeopsis fissa Gosse, Ascomorpha ovalis Carlin,



## Figure 3

Time-series fluctuations in environmental factors and rotifer density at the study sites in the Unmun Reservoir in 2009–2015. (a) WT (°C), water temperature; (b) DO (mg l<sup>-1</sup>), dissolved oxygen; (c) pH, (d) conductivity ( $\mu$ S cm<sup>-1</sup>), (e) turbidity (NTU), (f) T-N (mg l<sup>-1</sup>), total nitrogen; (g) T-P ( $\mu$ g l<sup>-1</sup>), total phosphorus; (h) chlorophyll *a* ( $\mu$ g l<sup>-1</sup>), and (i) rotifer density (ind. l<sup>-1</sup>)

Brachionus angularis Gosse, Brachionus calyciflorus Pallas, Filnia cornuta Weisse, Keratella cochlearis Gosse, Keratella quadrata O.F. Müller, Ploesoma hudsoni Imhof, Polyarthra spp., Synchaeta oblonga Ehrenberg, Trichocerca capucina Wierzejski & Zacharias, and Trichocerca gracilis Tessin. The most dominant rotifer species was A. fissa, followed by K. cochlearis and B. calyciflorus. All the rotifer species were more abundant in summer and autumn, with a relatively low density in winter and spring.

### **Data classification by SOM**

The SOM model was adaptively fitted to the input data (QE = 0.455; TE = 0.000). The optimal structure of the SOM model consisted of 45 hexagonal nodes (5  $\times$  9 array; Fig. 4c). The U-matrix (Fig. 4a) and distance among nodes (Fig. 4b) identified three distinctive clusters. The horizontal-center area of the U-matrix exhibited high distance scores (i.e. dark gray to black

color), which indicated the existence of distinguishing nodes in the upper lower part of the map plane. The subsequent clustering was analyzed based on relatively dark areas, while boundaries of the nodes in clusters were revealed with the hierarchical clustering method (Fig. 4b). Characteristics of the input variables in each cluster are summarized in Table 1, which presents average values of each variable.

Figure 5 compares the component planes, showing that each rotifer group exhibited different shapes and gradients on the map plane. *Polyarthra* spp. and *S. oblonga* were present in many of the clusters and were sporadically distributed in Clusters 1 and 2. Whereas *A. fissa, B. calyciflorus, K. cochlearis* and *T. gracilis* were concentrated in Cluster 2, while *Ascomorpha ovalis* and *P. hudsoni* were mostly found in Cluster 1. In addition to the eight rotifer species, eight environmental factors were displayed on the SOM. Each factor exhibited a different gradient on the map plane, and these gradients were helpful in

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### Figure 4

Clustering through data learning in the self-organizing map. (a) U-matrix, (b) hierarchical dendrogram, (c) clustering result

Table 1

335

Environmental factors and rotifer density in different clusters defined by SOM. Significant differences between clusters were based on one-way ANOVA. SD – standard deviation; F – F-value; p – probability value; WT (°C) – water temperature; DO (mg l–1) – dissolved oxygen; T-N (mg l–1) – total nitrogen; T-P (µg l–1) – total phosphorus

Fa ata a	Units	Mean	SD	Cluster			F	
Factor				1	2	3	F	p
WT	°C	15.6	8.8	19.6 ± 5.9	19.3 ± 7.1	$4.2 \pm 2.1$	52.3	< 0.01
DO	mg l⁻¹	107.8	17.2	94.6 ± 13.3	$108.1 \pm 10.7$	124.0 ± 16.3	28.5	< 0.01
рН		8.19	0.6	8.04 ± 0.5	8.06 ± 0.5	8.60 ± 0.8	6.9	0.02
Conductivity	µS cm⁻¹	93.9	10.2	85.6 ± 6.9	$100.3 \pm 6.7$	93.5 ± 10.6	25.9	< 0.01
Turbidity	NTU	9.8	2.5	8.0 ± 2.1	$11.2 \pm 1.8$	9.8 ± 2.6	16.6	< 0.01
T-N	mg l⁻¹	1.5	0.3	$1.2 \pm 0.3$	$1.6 \pm 0.2$	$1.5 \pm 0.3$	19.9	< 0.01
T-P	µg l⁻¹	17.8	3.3	15.2 ± 1.8	20.2 ± 2.1	16.9 ± 3.5	33.4	< 0.01
Chlorophyll a		12.3	4.2	8.9 ± 3.0	14.7 ± 2.8	12.6 ± 4.3	22.4	< 0.01
Anuraeopsis fissa		32.9	36.9	12.0 ± 5.9	65.6 ± 34.5	$3.6 \pm 6.8$	61.8	< 0.01
Ascomorpha ovalis		26.0	27.7	59.1 ± 21.4	$13.8 \pm 11.5$	$4.4 \pm 8.6$	96.7	< 0.01
Brachionus calyciflorus	ind. l <sup>-1</sup>	29.6	32.8	$10.4 \pm 4.2$	59.3 ± 29.8	$3.6 \pm 6.6$	69.7	< 0.01
Keratella cochlearis		41.5	30.2	36.9 ± 10.7	65.3 ± 26.1	6.4 ± 7.7	66.0	< 0.01
Ploesoma hudsoni		16.8	23.4	47.6 ± 16.0	2.3 ± 3.3	$1.9 \pm 4.0$	199.3	< 0.01
Polyarthra spp		20.5	12.4	26.7 ± 8.3	25.8 ± 8.3	3.7 ± 5.4	65.3	< 0.01
Synchaeta oblonga		9.2	7.4	$13.8 \pm 6.1$	10.8 ± 5.8	$0.6 \pm 1.8$	39.5	< 0.01
Trichocerca gracilis		22.0	27.3	$5.2 \pm 6.9$	46.8 ± 24.4	$1.1 \pm 2.8$	68.5	< 0.01

interpreting the effect of the environmental factors on the rotifer community. Water temperature was mainly concentrated in the lower left part of the map (Cluster 1 and 2), while DO and pH were concentrated in the upper part of the map (Cluster 3). Conductivity, turbidity, T-N, T-P, and chlorophyll *a* were gathered mainly in the lower right corner of the map (Cluster 2). The clusters of environmental parameters and rotifer group densities were significantly different (one-way ANOVA, Table 1).

Table 2 summarizes some of the primary characteristics such the main environmental factors, dominant rotifer species, and sampling time (initial,

middle, and late) for each cluster. Water temperature was the main factor separating Cluster 3 from Clusters 1 and 2. Higher values of water temperature were mainly concentrated in the lower left part of the map (Clusters 1 and 2), and Cluster 3 showed low values of water temperature (Fig. 5). This mean that Cluster 3 was related to winter time, when low densities of rotifer groups were observed. Clusters 1 and 2 were clearly distinguished by T-N and T-P concentration (Table 2, Fig. 5). The T-N and T-P concentration was higher in Cluster 2 and lower in Cluster 1. The clear distinction between the two clusters is related to the interannual distribution pattern of T-N and T-P. In the



Seong-Ki Kim, Jeong-Cheol Kim, Gea-Jae Joo, Jong-Yun Choi



## Figure 5

Environmental factors and rotifer density measured in the Unmun Reservoir. WT (°C) – water temperature; DO  $(mg I^{-1})$  – dissolved oxygen; T-N  $(mg I^{-1})$  – total nitrogen; T-P  $(\mu g I^{-1})$  – total phosphorus

#### Table 2

Characteristics of each cluster based on the distribution patterns of input variables and the number of study sites in each cluster. WT (°C) – water temperature; DO (mg  $I^{-1}$ ) – dissolved oxygen; T-N (mg  $I^{-1}$ ) – total nitrogen; T-P ( $\mu$ g  $I^{-1}$ ) – total phosphorus

Cluster		Voor	Concorn	Characteristics	No.	
Cluster	Environment	nvironment Rotifer		Season		Characteristics
1	WT	Ascomorpha ovalis, Ploesoma hudsoni, Polyarthra spp., Synchaeta oblonga	2009, 2010, 2011	spring, summer, autumn	low nutrient concentration	27
2	WT, Conductivity, Turbidity, T-N, T-P, Chlorophyll <i>a</i>	Anuraeopsis fissa, Brachionus calyciflorus, Keratella cochlearis, Polyarthra spp., Synchaeta oblonga, Trichocerca gracilis	2012, 2013, 2014, 2015	spring, summer, autumn	eutrophication state, most rotifers are abundant	36
3	DO, pH, T-N	-	-	winter	low water temperature, low rotifer density	21





initial survey (2009–2011), the Unmun Reservoir was characterized by low concentrations of T-N and T-P, after which T-N and T-P values gradually increased (Fig. 3). Thus, Cluster 1 was located according to the initial sampling data, and Cluster 2 was positioned based on the later surveys.

According to the map plane, rotifer species were largely affected by the concentration of T-N and T-P. Some rotifer species (Fig. 5), such as *A. fissa*, *B. calyciflorus*, *K. cochlearis* and *T. gracilis*, were positively correlated with T-N and T-P, while *Ascomorpha ovalis* and *P. hudsoni* were negatively correlated with T-N and T-P. This interannual distribution of rotifers was clearly related to the time-series fluctuations of T-N and T-P during the study period (Fig. 3). However, *Polyarthra* spp. and

*S. oblonga* were abundant both in Cluster 1 and Cluster 2 with high water temperatures, regardless of nutrient concentrations such as T-N and T-P.

# Interannual distribution of rotifers in summer and autumn

Summer and autumn densities of rotifer communities were observed to have different interannual distribution patterns in the eight dominant species (Fig. 6). *Anuraeopsis fissa*, *B. calyciflorus*, *K. cochlearis* and *T. gracilis* showed an upward trend in the study period, while *Ascomorpha ovalis* and *P. hudsoni* gradually decreased. The trends were more pronounced in autumn (opened circles, Fig. 6) than in summer (closed circles, Fig. 6). However, *Polyarthra* spp.



#### Figure 6

Interannual summer and autumn distribution of eight dominant rotifer species in the Unmun Reservoir. (a) *Anuraeopsis fissa*, (b) *Ascomorpha ovalis*, (c) *Brachionus calyciflorus*, (d) *Keratella cochlearis*, (e) *Ploesoma hudsoni*, (f) *Polyarthra* spp., (g) *Synchaeta oblonga*, (h) *Trichocerca gracilis*. ●: summer,  $\circ$ : autumn



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and *S. oblonga* showed no change during the survey period, with varying annual summer and autumn densities.

# Environmental factors and land cover in two tributary streams

The interannual summer changes in environmental factors measured in the Unmun and Sinwon streams clearly differed (Fig. 7). While most of the environmental factors showed no noticeable interannual changes in the Unmun Stream, there was a marked upward trend in the Sinwon Stream. Some environmental factors, such as conductivity ( $r^2 = 0.98$ , p < 0.01), turbidity ( $r^2 = 0.97$ , p < 0.01), T-N ( $r^2 = 0.99$ , p < 0.01), T-P ( $r^2 = 0.94$ , p < 0.01), and chlorophyll a ( $r^2 = 0.99$ , p < 0.01), increased noticeably during the study period (2009–2015). Moreover, most environmental factors showed higher values in the Sinwon Stream than in the Unmun Stream.

Trends in the land cover area around the Unmun and Sinwon streams were clearly different (Table 3). More specifically, an increase in the commercial and cultural/recreational areas was observed in both streams, but this increase was greater in the Sinwon Stream compared to the Unmun Stream. In 2007, the commercial area around the Sinwon Stream was 6186 m<sup>2</sup>, similar to the commercial area of the Unmun Stream, but in 2013 the Sinwon Stream increased more than seven times – up to 44 684 m<sup>2</sup>, whereas the Unmun Stream increased only to 10 734 m<sup>2</sup>. This trend was also found in the cultural/recreational areas of the two streams. There was no cultural/recreational area in 2003 and 2007 in either stream, but by 2013 the cultural/recreational land use in the Sinwon Stream covered an area of 40 504 m<sup>2</sup>, while in the Unmun Stream – only to 4209 m<sup>2</sup>. The area of the remaining land cover types (residential, paddy, and field) was irregular, as it showed slight increases or decreases.

## Discussion

## **Cluster characterization of the SOM Network**

Our objective was to explain i) interannual changes in the rotifer community structure in relation to environmental changes, and (ii) responses of rotifers to changes in the trophic state caused by human activity. We hypothesized that the pollution in the main



## Figure 7

Interannual summer change of environmental factors in the Unmun and Sinwon streams. (a) DO (mg  $I^{-1}$ ) dissolved oxygen; (b) conductivity ( $\mu$ S cm<sup>-1</sup>); (c) turbidity (NTU); (d) T-N (mg  $I^{-1}$ ) total nitrogen; (e) T-P ( $\mu$ g  $I^{-1}$ ) total phosphorus; (f) chlorophyll *a* ( $\mu$ g  $I^{-1}$ )





Table 3

Land cover (m<sup>2</sup>) change of the Unmun and Sinwon streams. Res. – residential area; Com. – commercial area; Cul. – cultural/recreational area; Paddy – paddy area; Field – field

	Unmun Stream				Sinwon Stream					
	Res.	Com.	Cul.	Paddy	Field	Res.	Com.	Cul.	Paddy	Field
2003	108 132	0	0	352 613	197 180	139 634	5082	0	329 444	326 708
2007	124 809	6066	0	362 887	186 557	167 932	6186	0	302 955	308 158
2013	195 124	10 734	4209	218 754	266 878	154 048	44 684	40 504	112 260	256 991

tributary would have a strong impact on the trophic state, which in turn would be manifested in different rotifer species. Our findings confirm that rotifers actually respond to nutrient concentrations and can be used as indicators of water quality in the Unmun Dam.

Based on the SOM cluster results, we suggest that nutrient concentrations (T-N and T-P) affect not only water quality, such as conductivity and chlorophyll *a*, but also rotifer groups. *A. fissa*, *B. calyciflorus* and *T. gracilis* were abundant in the eutrophic state with high nutrient concentrations, while high densities of *Ascomorpha ovalis* and *P. hudsoni* were observed with low nutrient concentrations. However, *K. cochlearis, Polyarthra* spp. and *S. oblonga* were abundant during all seasons except winter, regardless of the trophic status.

Although empirical studies have suggested the influence of environmental factors on rotifer communities over time (Dejen et al. 2004; Duggan et al. 2002; Wang et al. 2010), we found that some environmental factors (e.g. DO and pH) were not significantly correlated with the distribution of rotifers. These factors were evenly distributed on the trained SOM plane, which indicated that they did not have any relationship with the well-clustered rotifer patterns. In addition, the low concentration of DO was found during the development of rotifers (April-October, due to the excessive development of phytoplankton on the water surface), and thus was not associated with the distribution patterns of rotifers. Consequently, we consider that shallow wetlands are directly governed by the trophic state (caused by T-N and T-P) and indirectly by water physico-chemistry.

### Impact of the trophic state on rotifer groups

In this study, the eutrophication changed the species composition and increased the abundance of rotifers. The Unmun Reservoir showed low nutrient levels (i.e. mesotrophic) in the early period of the research (2009–2011), but gradually turned to eutrophication in the later period of the survey. The dramatic change in nutrient concentration was sufficient to lead to an interannual change in

the species composition of rotifers. Rotifer species, such as A. fissa, B. calyciflorus, K. cochlearis and T. gracilis, were more abundant in the later period of the study when the nutrient levels were higher. Previous studies also reported that these species were mainly observed in eutrophic lakes (Gao et al. 2019; Mäemets 1983; Singh et al. 2013). On the other hand, Yin et al. (2018) suggested that A. fissa and T. gracilis prefer the oligotrophic status, however, different environmental features were involved in their research on backshore wetlands compared with this study. The density of Ascomorpha ovalis and P. hudsoni was higher in the early part of the survey and gradually decreased in the latter part of the survey. Based on this, we suggest that these two rotifer species prefer an oligotrophic environment. Similar to our study, Erdoğan & Güher (2005) found that Ascomorpha ovalis and P. hudsoni were abundant in lakes with low nutrient concentrations. Duggan et al. (2001) stated that rotifers have the potential as bioindicators of the trophic state of lakes. Beaver & Crisman (1990) also suggested that rotifer communities are sensitive indicators of small changes in the trophic state of lakes. This supports the observations made by Balvay (1989) and Balvay & Laurent (1990) on changes in the rotifer communities in Lake Geneva over a period of three decades. Short-term bag experiments (Hessen & Nilssen 1985) have shown that factors like predation from Asplanchna and suppression by cladocerans are important inhibitors of rotifers in spring, whereas strictly eutrophication factors (i.e. chlorophyll content, phytoplankton biomass, phosphorus concentrations) are important in summer.

The increase in the density of rotifers with an increase in eutrophic status has been reported extensively (e.g. Bays & Crisman 1983; Pace 1986; Yoshida et al. 2003). It is believed that the regional distribution of rotifer species is strongly correlated with trophic status (Castro et al. 2005; Duggan et al. 2001; Duggan et al. 2002; Yoshida et al. 2003). Previous studies have reported that rotifer abundance in different lakes was also related to the species composition of algae (Bays & Crisman 1983; Gasol et al. 1995; Pace 1986). Edmondson (1965) found



that the reproduction rates of three rotifer species (K. cochlearis, Kellicottia longispina Kellicott and Polyarthra vulgaris Carlin) were clearly related to the abundance and species diversity of algal food. Bacteria are known to be an important food source for Keratella spp. and K. longispina (Arndt 1993; Bogdan et al. 1980; Ooms-Wilms 1991; Walz 1995), but not for Polyarthra and Synchaeta, which are specialist herbivores (Gilbert & Bogdan 1981; Pourriot 1997; Walz 1995). Furthermore, Pourriot (1997) suggests that Chrysophyceae is the dominant food of Synchaeta lakowitziana Lucks, while Cryptophyceae is preferred by Polyarthra species. Keratella are also polyphagous species that mainly feed on Chlorococales, Volvocales and Chrysomonadales smaller than 10 µm (Walz 1995). According to Pourriot (1997), Chrysomonadales and diatoms are suitable food for Kellicottia. Devetter (1998) showed that the abundance of Chrysophyceae, ciliates and heterotrophic nanoflagellates (HNF) is the most important food factors structuring the rotifer community in the Římov Reservoir. On this basis, it is estimated that different food composition related to trophic conditions in lakes and reservoirs leads to different species in rotifer communities. Our results show that it is more effective to evaluate changes in the overall species composition of rotifers in order to understand the effects of environmental changes than to monitor certain rotifer species as indicators, as was the case in previous studies (Gannon & Stemberger 1978; Sládeček 1983).

# Effect of tributary streams on environmental characteristics of the Unmun Reservoir

In this study, environmental changes in the tributary Sinwon Stream caused by human activity strongly affected the trophic state and species composition of rotifers in the downstream Unmun Reservoir. During the study period, as in the case of the Unmun Reservoir, the trophic state of the Sinwon Stream increased gradually due to changes in the surrounding land cover. It has already been suggested in many limnological studies that human activities affect the water quality of streams (Angelidis et al. 1995; Gupta et al. 2017; Olajire & Imeokparia 2001). Due to the aesthetic qualities of water, diverse human activities are carried out around the streams, which have a negative effect on water quality (Goethals & De Pauw 2001; King & Mace 1974; Søndergaard & Jeppesen 2007). For example, residential areas, accommodation and commercial facilities built around the streams can increase water pollution because their inhabitants generate sewage (King & Mace 1974). In this study, land cover changes around the

Sinwon Stream are a recent example of active stream utilization by humans. In 2003, the area of commercial and cultural/recreational land near the Sinwon Stream was similar to that of the Unmun Stream. In 2013, i.e. after 10 years, it increased by about 17 times, i.e. up to about 85 000 m<sup>2</sup>. The increase in human use led to a gradual increase in the nutrient level in the Sinwon Stream. The nutrient status of the Unmun Reservoir, which is located downstream, appears to have been caused by an increase in nutrient concentration in the Sinwon Stream. This is demonstrated by the fact that the land cover around the Unmun Reservoir is mainly forested and there are few non-point sources of nutrients that can feed the reservoir directly.

The rapid population growth and economic development have accelerated the rate of changes in land cover around the streams. Land cover change has led to diverse disturbances, including habitat loss and fragmentation, biodiversity loss, soil degradation and species invasion (Grau et al. 2003). Land cover change is particularly dramatic in tropical developing countries, which tend to be characterized by rapidly growing human populations (Grau et al. 2003; Watson et al. 2001). Land cover changes can induce changes in precipitation (Snyder et al. 2004), evapotranspiration (Yang et al. 2012), soil moisture (Chen et al. 2009), the type of vegetation and its water use, as well as stream-flow generation.

We assume that tributary streams will largely influence the trophic state of reservoirs in South Korea, based on the structure and form of the stream network. In the regions of East Asia, including South Korea, rainfall is concentrated mainly in the summer due to the impact of the monsoon climate. Therefore, dams or weirs have been constructed in upstream areas of rivers or streams to efficiently utilize summer rainfall. The reservoirs retain the quantity of water from the upstream parts of their catchments, but they are very vulnerable to water contaminants as well as nutrient concentrations such as phosphorus and nitrogen. In this study, we found that changes in the land cover types, such as commercial and cultural/recreational areas, in the tributary streams greatly contributed to the water quality status of the downstream reservoir, which affected the distribution of aquatic organisms, including rotifer communities. At present, the Unmun Reservoir is mostly characterized by eutrophication conditions in summer and autumn when the water temperature is high, causing changes in environmental factors such as light penetration through the water or reduction of DO. The changing trend of these environmental factors is predicted to lead to changes in the species composition of rotifers.



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# Changes in leisure activities and water quality management

In South Korea, changes in land cover types around the streams associated with human activities are largely related to recently adopted leisure activities. After the Korean War in 1951, South Korea sought to improve its living standards and to increase its revenue through gradual economic growth, which until recently gave little time for leisure activities. However, the recent rapid economic growth has led to increased leisure activities, accelerating the land development in areas surrounding the waterways. This has negatively affected the environment of stream basins. The change is largely attributed to the growing aesthetic desire to visit a natural environment as more residents live in urban areas. Leisure activities in South Korea are concentrated mainly in the summer, with a cultural preference for summer vacations. The summer temperature in South Korea has also been gradually increasing (Boo et al. 2006), which further promotes the recreational use of water-related natural environments such as rivers and oceans. In this study, we found gradual changes in land cover, such as the growth of commercial facilities in the Sinwon Stream and surrounding areas. The most noticeable change around the stream was the increase in the number of beds (accommodation facilities, i.e. guesthouses and campgrounds), which led to severe disturbance in the stream by increasing temporary occupation in summer or autumn. Empirical studies have reported that the impact of human activity on streams is observed mainly in developing countries (Alvani et al. 2011), but the increase in nutrient concentrations in South Korean streams is caused by the growth of facilities related to human leisure activities, rather than land cover changes resulting from economic growth, which is a clear difference compared to other cases. Moreover, unlike other countries, leisure activities have led to the eutrophication of streams and downstream reservoirs only in certain seasons (especially summer). Levels of eutrophication are sufficient to lead to changes in the interannual distribution of aquatic organisms, including rotifer communities.

The list of endangered species associated with aquatic ecosystems of South Korea (i.e. fresh water, marine, etc.) includes 19 species that occur in the upper reaches of streams; they account for 70% of all species on the list. On the other hand, relatively few endangered species are found in the middle and lower parts of the streams. The decline of aquatic organisms in the upper reaches of the streams is strongly related to human activity. For example, endangered fish species, such as *Pseudobagrus brevicorpus* Mori

and *Microphysogobio rapidus* Chae and Yang, are found mostly in areas where water flows are well maintained and rich in DO (Chae & Yang 1999; Kang et al. 2007), but the construction of upstream weirs has led not only to a decrease in water flow but also a decrease in DO due to the continuous accumulation of nutrients (Ogbeibu & Oribhabor 2002). Given the aforementioned structure of the Korean stream network, we assume that the negative impact found in our study is not limited to the Sinwon Stream.

Our results indicate that the upper part of the South Korean stream network needs to be actively managed to improve water quality, as further deterioration may occur in the future due to the gradual increase in human activity. Therefore, it is necessary to introduce appropriate human activities around the streams and restrict the growth of recreational facilities. Moreover, we suggest that leisure activities concentrated in summer should be spread over other seasons. This would reduce seasonal disturbance around the streams. However, such a measure could not be continued in the future. The best approach is to reduce human activity around the streams. The conservation of freshwater ecosystems to enhance their ecological functions is more urgent than in the case of terrestrial or marine environments. Since freshwater ecosystems have clearer boundaries than other ecosystems, even small changes can strongly affect the distribution of aquatic organisms. In particular, since the stream areas in South Korea are relatively large compared with other countries, it is necessary to improve water quality and habitat conservation through effective management.

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Seong-Ki Kim, Jeong-Cheol Kim, Gea-Jae Joo, Jong-Yun Choi

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