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Use of rotifer trophic state indices to show the effect of hydrobionts and nutrients on water trophic status in mesocosms

by

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Abstract

We have determined how Crustacea, zebra mussels (*Dreissena polymorpha*), fish and nutrients affect the trophic state in mesocosms filled with water collected from the pelagic zone of a eutrophic lake. We hypothesized that the pool of nutrients would increase both directly due to the input of phosphorus and nitrogen and/or indirectly due to the introduction of animal biomass. We used trophic state indices based on the abundance and species structure of rotifers to assess changes in the trophic state in mesocosms. The role of small detritophages in rotifer communities increased as a result of treatments. Our results clearly demonstrated that *D. polymorpha* was able to reduce the trophic status of mesocosm waters as indicated by reduced rotifer trophic state indices regardless of interactions with other treatment factors. Contrary to our expectations, neither the nutrients added at the beginning of the experiment nor the introduction of crustaceans or fish affected the rotifer trophic indices.

Key words: Rotifera, eutrophic lake, effects of Dreissena's, fish, Crustacea

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Introduction

Eutrophication is a process where the pool of nutrients in a lake increases as a result of an influx from outside the ecosystem. This increase in the pool of nutrients is then followed by changes in internal nutrient cycles (Ejsmont-Karabin 1983). Mineralization of phosphorus (P) by heterotrophs generally provides a sufficient amount of P to sustain primary production during periods of low P availability, but external pulses are required to induce algal blooms (Kamarainen et al. 2009). In eutrophic lakes, a smaller proportion of algal biomass is directly used by planktonic herbivores, while a greater proportion is indirectly consumed (after cell death) by bacterivores. This may accelerate the rate of nutrient regeneration (Ejsmont-Karabin 1983) and thus increase the pool of nutrients and the trophic state of lake waters in general. Experiments carried out by Bossard & Uehlinger (1993) have shown that the exclusion of crustaceans increases the residence time of total P by a reduced P loss through sedimentation. Thus, the presence of crustaceans should reduce the pool of phosphorus and, consequently, the trophic state of waters. This role of large cladocerans in the phytoplankton biomass control has been confirmed by Ejsmont-Karabin et al. (2004), although in some cases other zooplankton organisms may play the same role.

Fish can affect the processes of eutrophication as demonstrated by Stenson (1982). His manipulation of fish at the top of the ecosystem resulted in changes in nutrient cycles and the development of a new rotifer community. A significant structuring role of fish was also shown in 8-year studies on three eutrophic lakes (Jeppesen et al. 2000). Reduction in planktivores in the lakes resulted in an increase in cladoceran size and a decrease in chlorophyll a concentrations.

Zebra mussels *Dreissena polymorpha* may also play a quantitatively important role in the nutrient budget of lakes (Goedkoop et al. 2011). They are expected to efficiently trap nutrients, thus it has been proposed that they can be used to increase water quality. In large-scale studies carried out in Saginaw Bay (Fahnenstiel et al. 1995), *D. polymorpha* caused a strong decrease in the trophic state of Saginaw Bay (Lake Huron), i.e. a 59% and 43% decrease in chlorophyll a and total phosphorus, respectively, and a 60% increase in Secchi disk transparency. However, *D. polymorpha* produces large quantities of detritus, which may also improve the trophic state of lake waters (Ejsmont-Karabin et al. 2004).

In experiments on the impact of Crustacea, bivalves (*Dreissena polymorpha*), fish and nutrients, the pool of nutrients was increased both directly by the input of phosphorus and nitrogen or indirectly by the introduction of animal biomass. As a result, these inputs are expected to change the trophic state of water in our experiment. However, the functioning of invertebrate populations may also have an impact on the trophic state. The nature of changes, especially in variants with *D. polymorpha*, fish and crustaceans, is difficult to predict. The main objective of this study was to assess the effect of adding different animal populations on the trophic state of lake waters. We used rotifers as indicators of trophic changes in the experiment as this group of zooplankton responds concurrently to environmental changes due to their high intrinsic growth rates (Beaver & Crisman 1990; Ejsmont-Karabin 2012; Pociecha et al. 2018).

Materials and methods

The experiment was carried out from 31 July to 1 September 2012 and consisted of 12 treatments, each of which was replicated in triplicate mesocosms (36 mesocosms in total). The outdoor mesocosms were originally filled with 270 l of unfiltered water from the pelagic zone of eutrophic Lake Mikołajskie (498 ha; Masurian Lake District, northeastern Poland) and kept on the shore of that lake during the experiment. Mesocosms (plastic containers with internal dimensions of 0.94 m × 0.50 m, a height of 0.64 m and a capacity of 300 l) were devoid of sediments.

Three mesocosms were filled with unfiltered water that contained the natural abundance of zooplankton and served as the control (C). One-liter samples collected from each mesocosm were examined for zooplankton on the day the experiment started. According to the results of the observations, the control consisted of small cladocerans *Chydorus sphaericus* (O.F. Müller), *Bosmina coregoni* Baird, *B. longirostris* (O.F. Müller), *Ceriodaphnia pulchella* Sars and copepods *Eudiaptomus gracilis* Sars, *E. graciloides* (Lilljeborg), *Mesocyclops leuckarti* (Claus), *Thermocyclops oithonoides* (Sars) and a community of rotifers strongly dominated by species of the genus *Polyarthra* (*P. remata* Skorikov and *P. vulgaris* Carlin) and two species indicative of high trophic status: *Anuraeopsis fissa* (Gosse) and *Trichocerca pusilla* (Lauterborn). The remaining treatments consisted of unfiltered lake water plus: crustacean species (Cr), *Dreissena polymorpha* (Dp), nutrients (N), fish (F), Crustacea & *D. polymorpha* (CrDp), fish & Crustacea (FCr), nutrients & Crustacea (NCr), nutrients & *D. polymorpha* (NDp), nutrients & fish (NF), nutrients & *D. polymorpha* & Crustacea (NDpCr), nutrients & fish & Crustacea (NFCr).

The Crustacea (Cr) treatment was a mixture of crustaceans: 500 ind. of *Daphnia pulex* (Leydig), 140 ind. of *Simocephalus vetulus* (O. F.Müller), and a few individuals of *Macrocyclops albidus* (Jurine) collected from neighboring waterbodies and added to the unfiltered lake water. These species were not present in water from Lake Mikołajskie.

Dreissena polymorpha individuals, which were scarce in Lake Mikołajskie, were transported in coolers from Lake Boczne to the field station. They were added to the mesocosms within 24 h of their collection. About 200 individuals of *D. polymorpha* (i.e. 250 g m^{-2}) were selected randomly from the common pool and introduced into each of the 12 mesocosms.

We enriched the water in six treatments (18 mesocosms) with 1.728 mg l^{-1} N-NO_3, 0.192 mg l^{-1} N-NH_4 and 0.120 mg l^{-1} P-PO_4, in order to obtain concentrations of nutrients typical of highly eutrophicated waters.

The fish treatment consisted in placing one individual of the ruffe (*Gymnocephalus cernua*) into slotted boxes that were suspended in the mesocosms. The total fish length (*longitudo totalis*) in one mesocosm was 8.3 ± 0.4 cm and the mean fish size did not change over the course of the experiment. The species was selected for its ability to live in different trophic conditions and because ruffe of this size may feed on large zooplankton, e.g. cladocerans, large copepods (Gutsch & Hoffman 2016).

One-liter samples were collected from each mesocosm on days 1, 12, 22 and 32 of the experiment to examine rotifers. The samples were fixed with Lugol's solution, condensed on a plankton net with a mesh size of 30 µm, and again fixed in 2% formalin. Individual rotifer biomass was determined based on relationships between body length and body weight for each species (Ejsmont-Karabin 1998).

Rotifer trophic state indices (TSI_{mean}) were used to assess changes in the trophic state of the mesocosms based on: (1) the number of rotifers (N, ind. l^{-1}): $TSI_N = 5.38 \ Ln(N) + 19.28$; (2) total biomass of the rotifer community (B, mg w.wt. l^{-1}): $TSI_B = 5.63 \ Ln(B) + 64.47$; (3) percentage of bacterivores in the total number of rotifers (BAC, %): $TSI_{BAC} = 0.23 \ BAC + 44.30$; (4) ratio of the biomass of rotifers to their count (B:N, mg w.wt. $ind.^{-1}$): $TSI_{B:N} = 3.85 \ (B:N)^{-0.318}$; (5) percentage of the form *tecta* in the population of *Keratella cochlearis* (TECTA, %): $TSI_{TECTA} = 0.198 \ TECTA + 48.8$; (6) contribution of species indicative of high trophic status to the size of the indicator group (IHT, %): $TSI_{IHT} = 0.203 \ IHT + 40.0$ (Ejsmont-Karabin 2012).

The rotifer trophic state index (TSI_{mean}) was defined as a mean of the particular zooplankton indices. Although eutrophication is a continuous process, for practical reasons it has been assumed that mesocosms with a TSI_{mean} under 45 were mesotrophic, 45–55 were meso-eutrophic, 55–65 were eutrophic, and greater than 65 were hypertrophic (Ejsmont-Karabin 2012).

We used a generalized linear model (GLM) for statistical analysis, which was designed to assess the effect of one or more treatment factors on one or more dependent variables. Our analyses involved three factors, including treatment (A), the number of mesocosms (B), and time, i.e. date (C). We analyzed TSI_{mean} (averaged over all six TSI indices), TSI_N based on the number of rotifers, TSI_B based on rotifer biomass, TSI_{BAC} based on the percentage of bacterivores in the total number of rotifers, $TSI_{B/N}$ based on the ratio of biomass to count. Two TSI indices (percentage of the *tecta* form in the population of *Keratella cochlearis* and contribution of species which indicate a high trophic status in the size of the indicator group) were not included in the statistical analysis, because data for some dates were missing. However, they could be used in support of or against statistical model results.

We constructed mixed GLMs to compare the treatments (A), interactions between treatment (A) and time (C), and the effects of mesocosm (B). The treatment and time (repeated measure) were specified as fixed factors, while the mesocosm was randomly nested within the treatments (B(A)). Data were $\log_e$ transformed as necessary to help meet the assumptions of normality of residuals. When significant treatment effects were detected with ANOVA, Fisher's LSD post hoc test ($p < 0.05$) was used to determine which factor significantly affected the TSI. A comparison between the control and each of the treatments was performed using Dunnett's procedure. All statistical analyses and the construction of plots were performed in R 3.3 (R Core Team 2017) and in the integrated software Biosystem office (Petrosyan 2014).

Results

Rotifers increased their abundance in 61% of the 36 mesocosms by the end of the experiment (Table 1). However, the addition of nutrients together with crustaceans and/or fish often resulted in a reduced number of rotifers, as rotifer density increased in only 25% of the treatments. The highest increase in rotifer density was observed in the control (2.5-fold), the Crustacea treatment (3.9-fold), the *D. polymorpha* treatment (3.4-fold), and the fish and Crustacea treatment (2.6-fold).

Rotifer biomass in the control did not change throughout the experiment. It increased in four treatments and the mean increase was 1.9-fold (SD = 0.4). The highest increase in rotifer biomass was

Table 1

Quantitative and qualitative features of rotifer communities used to calculate rotifer trophic state indices: N – the number of rotifers (ind. l^{-1}), B – rotifer biomass (mg l^{-1}), BAC – bacterivores in the total number of rotifers (%), TECTA – percentage of the tecta form in the *Keratella cochlearis* population; B/N – ratio of rotifer biomass to rotifer count (µg ind.$^{-1}$), IHT – contribution of species indicating the high trophic status in the indicator group (%); N/a – not applicable

Treatment	No.	After 1 day of the experiment						After 32 days of the experiment					
		N	B	BAC	TECTA	B/N	IHT	N	B	BAC	TECTA	B/N	IHT
Control	1	714	0.160	5.3	50.0	0.236	96.4	1516	0.117	78.8	0.0	0.077	100.0
	2	923	0.201	12.7	24.0	0.218	97.9	2498	0.187	89.4	7.2	0.075	N/a
	3	1346	0.363	15.3	15.8	0.269	95.5	3405	0.444	74.4	0.8	0.130	100.0
C + Crustacea (Cr)	1	700	0.185	5.1	15.4	0.264	89.0	2185	0.190	81.5	0.5	0.087	100.0
	2	567	0.138	8.1	22.2	0.243	96.1	3993	0.424	83.4	3.9	0.106	100.0
	3	512	0.098	17.4	0.0	0.192	86.0	709	0.082	51.6	N/a	0.116	100.0
C + Dreissena (Dp)	1	380	0.088	1.3	0.0	0.290	87.5	520	0.076	51.7	0.0	0.147	100.0
	2	78	0.017	1.3	N/a	0.217	50.0	818	0.119	43.6	N/a	0.145	N/a
	3	123	0.027	4.1	0.0	0.216	100.0	637	0.104	57.5	N/a	0.163	N/a
C + Nutrients (N)	1	217	0.040	2.3	100.0	0.184	100.0	1996	0.276	53.7	N/a	0.138	N/a
	2	7338	1.652	5.2	33.7	0.225	92.2	2253	0.282	59.2	2.1	0.125	93.6
	3	3181	0.736	7.4	18.8	0.231	95.7	1065	0.135	80.2	2.2	0.127	98.6
C + Fish (F)	1	1147	0.270	7.1	8.0	0.236	95.2	1428	0.141	60.3	0.0	0.099	100.0
	2	1616	0.358	14.0	39.1	0.222	95.9	2615	0.440	76.1	6.8	0.168	100.0
	3	1465	0.351	2.6	23.5	0.240	90.4	3044	0.285	66.0	0.0	0.094	100.0
C + Crustacea + Dreissena (CrDp)	1	408	0.090	2.2	N/a	0.220	90.5	186	0.045	50.0	N/a	0.241	N/a
	2	136	0.030	0.7	N/a	0.224	33.3	135	0.021	28.1	N/a	0.151	N/a
	3	308	0.072	4.5	0.0	0.234	68.4	290	0.044	52.4	N/a	0.151	N/a
C + Fish + Crustacea (FCr)	1	726	0.189	0.8	0.0	0.260	85.6	1162	0.090	70.6	0.0	0.078	100.0
	2	1907	0.479	7.0	41.9	0.251	92.0	3004	0.369	71.7	0.5	0.123	100.0
	3	1663	0.418	7.3	5.9	0.251	93.6	6988	0.469	85.3	3.5	0.067	100.0
C + Nutrients + Crustacea (NCr)	1	3190	0.682	6.5	26.9	0.214	96.4	4698	0.407	87.3	1.2	0.087	99.6
	2	5304	1.072	5.5	19.8	0.202	93.6	2008	0.170	69.2	1.2	0.085	99.6
	3	4054	0.881	3.8	25.7	0.217	92.7	3442	0.465	48.8	N/a	0.135	100.0
C + Nutrients + Dreissena (NDp)	1	451	0.104	1.8	N/a	0.231	55.5	285	0.039	58.6	N/a	0.136	100.0
	2	328	0.071	0.6	N/a	0.217	0.0	1886	0.316	65.9	N/a	0.168	N/a
	3	504	0.132	0.2	0.0	0.262	12.9	2132	0.311	63.9	N/a	0.146	N/a
C + Nutrients + Fish (NF)	1	3331	0.730	3.4	38.9	0.219	97.6	1153	0.112	79.9	0.5	0.097	99.6
	2	2076	0.424	16.2	27.8	0.204	97.9	4719	0.573	47.8	N/a	0.121	N/a
	3	5327	1.285	1.3	35.4	0.241	84.8	772	0.062	35.6	0.0	0.081	99.2
C + Nutrients + Dreissena + Crustacea (NDpCr)	1	1003	0.217	0.0	0.0	0.217	0.0	844	0.133	73.2	N/a	0.158	N/a
	2	584	0.132	0.9	N/a	0.227	54.5	1555	0.298	48.8	N/a	0.192	N/a
	3	251	0.065	0.0	N/a	0.258	14.3	647	0.144	49.6	N/a	0.223	N/a
C + Nutrients + Fish + Crustacea (NFCr)	1	1674	0.389	3.5	46.2	0.233	92.5	714	0.740	75.8	1.5	0.103	100.0
	2	7374	1.619	6.5	40.5	0.221	94.5	2652	0.336	58.9	0.0	0.127	97.5
	3	3888	0.880	7.8	28.9	0.226	94.4	2990	0.342	60.6	N/a	0.114	100.0

observed in the treatments with added *D. polymorpha* and nutrients. We observed a decrease in rotifer biomass in the remaining seven treatments. The 2.2-fold (mean with SD = 1.0) decrease was observed mainly in the treatments with added Crustacea. Thus, the increase was observed less frequently than in the case of the number of rotifers, which means that rotifer species occurring at the end of the experiment were smaller. This finding is confirmed by the very high increase in the abundance of small bacterivorous rotifers during the experiment, which was observed in all treatments (Table 1). Their contribution to the total number of rotifers increased 7 to 191 times, with the highest increase recorded in the treatments with added nutrients and *D. polymorpha*. This resulted in an almost twofold decrease in the biomass to count ratio. The decrease was the lowest (1.2-fold) in the treatments with *D. polymorpha* plus Crustacea, and the highest in the control and the mesocosms with fish plus Crustacea (Table 1).

The *tecta* form of *Keratella cochlearis*, which proved to be an excellent index of trophic changes (Ejsmont-Karabin 2012), was present at the beginning of the experiment in all mesocosms, except those with *D. polymorpha* (Table 1). The proportion of the form in the populations of *K. cochlearis* was about 30% in the control and from ca 30% to 50% in the mesocosms with added nutrients. At the end of the experiment, this form was absent or scarce in all treatments.

At the beginning of the experiment, rotifer communities were dominated by pelagic species, however, in the middle of the experiment they were significantly replaced by littoral species from the genera *Lecane*, *Lepadella* and *Colurella*. At the end of the experiment, the contribution of species indicating a high trophic status could not be assessed in 36% of the mesocosms, because all indicator species disappeared from the rotifer communities. In 44% of the mesocosms, only species indicative of high trophic status remained, whereas both indicator groups were present in 20% of the mesocosms. At the beginning of the experiment, rotifers indicating both low and high trophic status occurred in 89% of the mesocosms (Table 1). The contribution of species that indicated high trophic status was relatively high both at the beginning and the end of the experiment.

R-squared statistics for mixed GLMs showed that the fitted models explained 89.9% of the variability in Log$_e$[TSI$_{mean}$], 88.4% in Log$_e$[TSI$_N$], 86.3% in Log$_e$[TSI$_B$], 91.3% in Log$_e$[TSI$_{BAC}$], and 74.5% in Log$_e$[TSI$_{B/N}$], respectively. The Durbin-Watson (DWs) test indicated that there was no serial autocorrelation in the residuals for any of the mixed GLMs: Log$_e$[TSI$_{mean}$] – DWs = 2.4, p^{**} = 0.98; Log$_e$[TSI$_N$] – DWs = 2.2 , p^{**} = 0.88, Log$_e$[TSI$_B$] – DWs = 2.2, p^{**} = 0.86; Log$_e$[TSI$_{BAC}$] – DWs = 2.6, p^{**} = 0.99; Log$_e$[TSI$_{B/N}$] – DWs = 2.63, p^{**} = 0.99 (see *R*-squared statistic R^2 of GLM, Durbin-Watson statistic DWs, p^{**} value in Table 2). All the study factors significantly affected each of the TSI indices (Table 2), indicating that the trophic level varied depending on the treatment and mesocosm associated with the treatment and time.

The mean values of the trophic state index in all treatments were markedly higher at the end of the experiment than at its beginning (Fig. 1). However, during the first decade of the experiment with added *D. polymorpha* (both with nutrients and Crustacea), the TSI values decreased, and then increased again. Usually, but not in the control, the input of nutrients resulted in a slight increase in the TSI values.

Discussion

In most of our experiments, all treatment factors led to an increase in the rotifer trophic state indices. In all cases without the addition of nutrients, the rotifer trophic state indices indicated a high mesoeutrophic status at the beginning of the experiment and low eutrophic status at the end. In a few cases with the addition of nutrients, the trophic status was eutrophic. The increase resulted mostly from an increasing role of small detritophages in the rotifer communities. This observation is in accordance with Pejler's (1983) suggestion that most species indicating eutrophic conditions feed on very small particles (i.e. bacteria), whereas species indicating oligotrophic conditions are usually filtrators consuming coarser particles.

Dreissena polymorpha can reduce algal biomass, even toxic strains of cyanoprokaryotes (Sarnelle et al. 2012) and counteract the effects of eutrophication (Dzialowski & Jessie 2009). In response to *D. polymorpha*, a strong reduction in the number and biomass of rotifers, and consequently a decrease in mean values of the rotifer trophic state index was recorded in our experiment. However, the phenomenon was observed at the beginning of the experiment, while at the end of the experiment the values of rotifer indices of high trophic status increased again as a result of the increased role of small littoral species. Gannon & Stemberger (1978) observed that littoral species of rotifers (e.g. *Lecane*, *Euchlanis*, *Colurella*, *Lepadella* and others) became abundant in the pelagial of eutrophic waters. The causes of this phenomenon are unknown. In this study, the occurrence of small littoral rotifers from the genera *Lecane*, *Lepadella* and *Colurella* may be explained by mesocosm walls acting as a littoral substrate and the lack of competitors and predators among open water species.

The impact of fish on the trophic state is more complicated. Experiments conducted in mesocosms (Drenner et al. 1996) have shown that filter-feeding omnivorous fish interacted synergistically with the trophic state and usually increased the abundance of nanophytoplankton. Reinertsen & Langeland (1982) concluded that fish reduced the biomass of large cladocerans, resulting in an increase in the phytoplankton turnover. Our study seems to confirm the above conclusion, as the addition of nutrients to the mesocosms with fish and crustaceans did not change the trophic state. Experiments in enclosures (Hessen & Nilssen 1985) with added fertilizers, fish and competitors (Cladocera) provided evidence that the influence from potential competitors was less important than other factors.

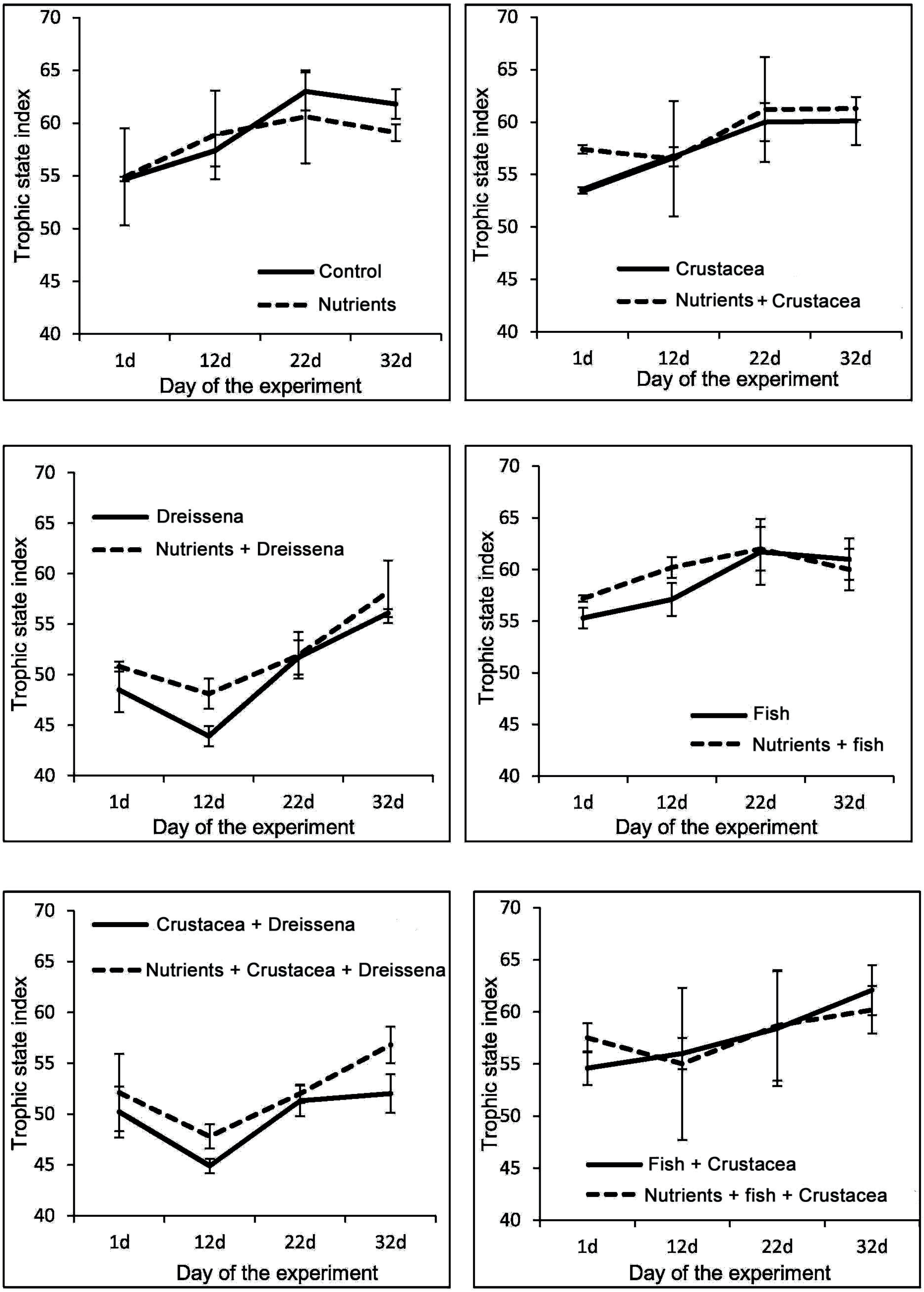

Figure 1

Changes in the mean values of the trophic state index in six experimental treatments without nutrients and six treatments with added nutrients

Table 2

Results of the effects of treatment, interaction of treatments × time and mesocosms on trophic state indices measured 4 times during the experiment. Mesocosms were treated as a random-nested factor in GLM. *F* is Fisher's test; DF$_1$, DF$_2$ – degrees of freedom of the *F*-ratio; $p*$– *p* value for the factor effects. Significant values are given in bold.

Source	DF$_1$/DF$_2$	*F*-ratio	$p*$-value	R-Squared statistic (R^2) of GLM, Durbin-Watson statistic (DWs; $p**$ value)
		Log$_e$[TSI$_N$]		
A – Treatments	11/72	17.33	**<< 0.0001**	**R^2 = 88.4 (%);** DWs = 2.2 ($p**$ = 0.88)
B(A) – Mesocosms (Treatments)	24/72	1.94	**0.017**	
C – Time	3/ 72	11.79	**<< 0.001**	
A × C – Treatments × Time	33/72	2.98	**0.0001**	
		Log$_e$[TSI$_B$]		
A – Treatments	11/72	17.24	**<< 0.0001**	**R^2 = 86.3 (%)** DWs = 2.2 $p**$ = 0.86)
B(A) – Mesocosms (Treatments)	24/72	1.65	**0.05**	
C – Time	3/ 72	9.48	**<< 0.0001**	
A × C – Treatments × Time	33/72	2.96	**0.0001**	
		Log$_e$[TSI$_{BAC}$]		
A – Treatments	11/68	2.08	**0.03**	**R^2 = 91.3 (%)** DWs = 2.6; ($p**$ = 0.99)
B(A) – Mesocosms (Treatments)	24/68	2.57	**0.001**	
C – Time	3/ 68	168.7	**<< 0.0001**	
A × C – Treatments × Time	33/68	1.28	**0.19**	
		Log$_e$[TSI$_{B/N}$]		
A – Treatments	11/72	1.98	**0.043**	**R^2 = 74.5 (%)** DWs = 2.63; ($p**$= 0.99)
B(A) – Mesocosms (Treatments)	24/72	1.36	0.16	
C – Time	3/ 72	33.90	**<< 0.0001**	
A × C – Treatments × Time	33/72	1.41	0.11	
		Log$_e$[TSI$_{mean}$]		
A – Treatments	11/72	16.04	**<< 0.0001**	**R^2** = 89.9; DWs = 2.4 ($p**$ = 0.98)
B(A) – Mesocosms (Treatments)	24/72	2.05	**0.01**	
C – Time	3/ 72	49.4	**<< 0.0001**	
A × C – Treatments × Time	33/72	2.37	**0.0012**	

A decrease in the trophic state may be expected in the experiments with *Daphnia*, similarly to enclosure experiments by Paterson et al. (2002), where total P in the water column decreased due to an increase in P sedimentation. This effect of Cladocera was also observed in our experiments, but the presence of *Daphnia* and *Simocephalus* only slightly decreased the trophic state in the mesocosms. The addition of nutrients in the treatments with Crustacea did not result in an increase in the trophic state, although it increased the trophic state in the remaining treatments. Similarly, a five-year study by Reinertsen and Langeland (1982) showed that the addition of nutrients, when zooplankton were dominated by cladocerans, did not increase the phytoplankton biomass. The role of large cladocerans in clearing lake water was explained by their "efficient" filtering as well as suppression of ciliates and rotifers through predation, interference and resource competition (Ejsmont-Karabin et al. 2004). The suppression of rotifers by large cladocerans can cause high mortality rates among sensitive species, even at very low *Daphnia* densities (Gilbert 1988). This may affect the rotifer community structure if some of the species coexist with *Daphnia*. However, the inhibitory effect was not observed in our experiment. On the contrary, the highest densities of rotifers were found in treatments with added crustaceans. It is possible that the shift of the pelagic rotifers into the littoral community during the experiment allowed rotifers to avoid suppression by Cladocera. The role of small cladocerans was apparently different, as rotifers are less inhibited by small cladocerans and may reach high densities in their presence (Gilbert 1988).

It is difficult to assess to what extent our results describe the conditions of the lake, i.e. changes in the trophic state of the lake due to the presence of crustaceans, *D. polymorpha* or fish. Experiments with large limnocorrals showed some significant differences in lake conditions (Bloesch et al. 1988). However, even if these experiments did not allow a very detailed quantitative interpretation of the results, they indicated a significant impact of zooplankton on the seston.

Our results clearly demonstrated that *D. polymorpha* is able to reduce the trophic state, as indicated by changes in rotifer trophic state indices at the beginning of the experiment, regardless of *D. polymorpha* interactions with other factors such as added nutrients, the introduction of Crustacea or fish. Contrary to our expectations, neither the nutrients added once at the beginning of the experiment nor the introduction of fish or crustaceans affected the trophic levels. Thus, *D. polymorpha* can serve as a powerful factor controlling rotifers and the trophic state. The effects of *D. polymorpha* can be related to its effective filtration ability. Due to their mass development and filtration activity, these organisms can filter the whole volume of a lake over a relatively short time. For example, the entire water volume of Lake Naroch in Belarus (V = 710.4 million liters) was filtered during 123 days (Karatayev et al. 2005). In our experiment, however, this effect became weaker on the 32nd day compared to the 12th and 22nd day. This was caused by the excretion of nutrients by zebra mussels, which can reduce the effects of its purification rate. Therefore, such *D. polymorpha* effects could be short term.

In general, rotifer communities respond very quickly to changes in their environment, sometimes through complete reconstruction. Therefore, changes in the structure of their communities can be used to indicate changes in ambient conditions.

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