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Title: Quantification of change in pelagic plankton network stability and topology based on empirical long-term data

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Highlights

- Plankton network stability increased while lake nutrient concentrations decreased
- *Dreissena polymorpha* larvae form a keystone group of the Müggelsee plankton network
- Pelagic plankton groups varied in their network centrality rank over time
- Stability and network centrality metrics may serve as ecosystem change indicators
- Long-term monitoring is crucial to assess anthropogenic impact on ecosystems

Abstract

Over the last 34 years, Lake Müggelsee has experienced concurrent warming and nutrient reduction. While the effects of environmental change on single taxonomic or physical-chemical variables have been relatively well researched in isolation, understanding how environmental change propagates through the ecological network remains a major challenge. Capitalizing on the long-term monitoring program of the German Long-Term Ecosystem Research Network site Lake Müggelsee (1979-ongoing), we identified three time periods (1979-1995; 1996-2005; 2006-2013) which differed significantly in phytoplankton biomass and relative plankton community composition. Using multivariate first order autoregressive (MAR1) modeling on 13 pelagic plankton groups and four abiotic variables, we quantified interaction networks and indicators of stability and centrality for each period. Our results suggested that the Müggelsee network was bottom-up regulated in all periods and that stability increased over time. Moreover, in all three networks, non-trophic and indirect interactions appeared to be as commonly present as trophic and direct interactions. Using network centrality measures of betweenness and closeness, we identified keystone plankton groups and groups particularly responsive to environmental change based on variation in centrality ranks over time. Given a more comprehensive understanding of the interaction network at hand, MAR1 model-derived stability and centrality measures may potentially be used as integrated ecological indicators to monitor changes in stability of lake ecosystems and to identify particularly vulnerable components of the network.

Keywords: community stability, interaction networks, long-term research, network centrality

1. Introduction

Lake ecosystems are considered important sentinels of environmental change as they integrate alterations in the catchment and atmosphere (Adrian et al., 2009; Williamson et al., 2009). Key response variables acting as sentinel variables include a wide range of physical, chemical and biological indicators that are sensitive to climate and land-use change (Adrian et al., 2009; Adrian et al., 2006). While the effects of anthropogenic pressure on key response variables are reasonably well understood in isolation, it remains a challenge to predict how global change affects the interactions among such variables and, thus, the ecological network of a lake and its stability. The lack of ground-truthed data on species interactions and community network response to stress has been identified as major gap in the bio-monitoring sciences (Gray et al., 2014). To better understand and predict how global change will affect community structure and stability and hence also associated ecosystem processes, it is necessary to assess how ecological networks change over time and under pressure.

Here, we make use of the long-term research program installed at the German Long-Term Ecosystem Research Network (LTER-D) site Müggelsee (Germany) to explore how changes in the phyto- and zooplankton biomass and community composition due to anthropogenic pressure affect the structure and stability of the pelagic interaction network utilizing multivariate first order autoregressive modelling (MAR1) and ecological network analysis. MAR1 modeling (Ives et al., 2003) allows the identification and quantification of network interactions and the derivation of stability metrics of ecological networks from long-term data (Hampton et al., 2013; Ives et al., 1999; Scheef et al., 2013). The resulting interaction matrix can also be used to inform ecological network analysis. MAR1 models have predominantly been used to elucidate trophic networks in both freshwater and marine systems, likely because short generation times of

plankton allow capturing hundreds of generations' worth of dynamics within few years. The method has been implemented to assess the food-web structure in deep lakes under changing climate and eutrophication (Hampton et al., 2008; Hampton et al., 2006) and the effect of predation on phytoplankton and ciliate population variability (Huber and Gaedke, 2006) as well as on disease transmission (Duffy, 2007), to appraise the response of pelagic networks to changes in fish predation pressure (Beisner et al., 2003; Ives et al., 1999) and to carbon and nutrient manipulations (Klug and Cottingham, 2001). As MAR1 models provide quantitative estimates of interaction strengths they allow the identification of direct and strong links but also of indirect "long and weak" links (Jordán, 2009).

Network stability indicators derived from MAR1 models are based on measurements of deviation from an "equilibrium" state, here the stationary distribution of a community under environmental noise. The stability indicators are expressed as variance of the stationary distribution in relation to the environmental variance (hereafter "variance"), return rate after perturbation ("resilience") and short term response to perturbation ("reactivity"), for a detailed derivation see Ives et al. (2003), for a short description see Table 1. These stability indicators are directly comparable across systems as they are not affected by the magnitude of fluctuations in system variables (Hampton et al., 2013) and hence also allow tracking stability of ecosystems over time. Most ecological networks in the literature describe networks aggregated over time or space and thus do not provide information about the variability in stability of networks in evolving natural systems (but see Francis et al., 2014). The application of MAR1 models and their derived indicators on sequential time periods can improve our assessment and predictive power on the response and stability of ecological networks under anthropogenic pressure. Tracking the variability in interaction strength among keystone groups in a network, or the

overall stability of the network over time may even serve as a leading indicator for ecosystem resilience and as advance warning for regime shifts (Francis et al., 2014; Kuiper et al., 2015).

The quantitative interaction matrix resulting from MAR1 models can be passed on to classic ecological network analysis to assess network properties such as closeness- and betweenness centrality. The centrality indicators can identify vertices (species, or groups of species) that are either well connected or connect otherwise disconnected compartments of the network and therefore take a keystone position in the network (Jordán et al., 2008). Changes in the position and dynamics of keystone species or groups are likely to cascade through the network as these groups are linked directly with many other groups in the network (Vasas and Jordán, 2006). Comparison of successive time period networks also allows tracking changes in the centrality scores and therefore the identification of groups that are particularly sensitive to environmental changes over time (Jordán and Osváth, 2009).

The aim of this study is to explore how changes in lake nutrient status and a warming trend affected the internal trophic (bottom up or top down) and non-trophic (competition, facilitation or indirect effects) interactions of the pelagic plankton as well as overall network stability and topology. We identified three periods between 1979 and 2013 which differed in phytoplankton biomass (period 1 versus periods 2 and 3) and plankton community composition (periods 2 and 3). These periods were analyzed for their interaction networks properties, including stability indicators and measures of network centrality. Our study is of exploratory nature, making use of the Müggelsee long-term dataset to assess interactions among pelagic plankton groups based on their temporal autocorrelation and is geared towards revealing

potentially overlooked keystone groups and key interactions in the plankton network as well as changes in network stability and centrality measures over time.

2. Methods

2.1. Study site

Lake Müggelsee is a shallow (mean depth 4.9 m, max depth 8 m), eutrophic lake situated southeast of the city of Berlin (Germany, 52° 26’ N, 13° 39’ E). The lake is polymictic and usually fully mixed due to the wind fetch of its relatively large surface area of ~750 ha (Driescher et al., 1993). The River Spree enters the lake from south-east and the outflow is situated in the north-west of the lake. This results in an average retention time of about 6-8 weeks (Köhler et al., 2005). Due to its location in a transition zone from a maritime to a more continentally characterized climate, the lake experiences large annual and inter-annual variability in local weather conditions.

Observed long-term changes: Over the past three decades, the lake has experienced an increase in seasonal warming by 2.4 and 2.3 K in spring and summer (Adrian et al., 2006; Wagner and Adrian, 2009) and a reduction in external nutrient loading by 50 % between 1990 and 2005 (Köhler et al., 2005). Driven by the reduction in nutrient load, phytoplankton biomass declined due to phosphorus limitation in spring and nitrogen limitation in summer (Köhler et al., 2005). This led to an increase in water transparency and a reappearance of macrophytes (Hilt et al., 2013). However, climate warming-induced increase in summer stratification (Wagner and Adrian, 2011) has been suggested to drive nutrient remobilization from the sediment (Wilhelm and Adrian, 2008). Buoyant cyanobacteria genera (*Microcystis* and *Anabaena*) benefitted from

elevated internal phosphorus release during stratified periods and genera capable of nitrogen fixation (*Anabaena* and *Aphanizomenon*) became prominent during nitrogen-limited prolonged stratification periods in summer (Wagner and Adrian, 2011). Thus, extensive algal summer blooms have remained common in the lake and blooming period extends into fall. Climate-change induced shifts in phenology (Adrian et al., 2006) affected the timing of diatom spring-bloom onset (earlier ice break-up promoted earlier bloom onset) and *Daphnia* population peaks (higher spring water temperature promoted earlier population peaks). Responses to warming in summer depended on species-specific thermal requirements and timing of warming with specific developmental stages, such as emergence from diapause (copepods), or spawning (*Dreissena*). Zooplankton species with high thermal tolerances (i.e. *Thermocyclops oithonoides*, *Thermocyclops crassus*) and/or taxa known to grow faster at high temperatures (e.g. rotifers) have become more abundant (Wagner and Adrian, 2011).

2.1.1. Sampling and sample processing

Since 1979, an intensive monitoring program of physical-chemical and biological variables has been installed at Müggelsee. Samples for pelagic phyto- and zooplankton and concentrations of dissolved reactive phosphorus (SRP), total phosphorus (TP), dissolved inorganic nitrogen (DIN = nitrate + ammonium) and dissolved reactive silicate (RSi) have been taken at fortnightly (winter) and weekly intervals (summer). Secchi depth was measured with a Secchi disk on each sampling occasion. Water temperatures were measured weekly between 8:00 and 9:00 AM at 0.5 m depth at a landing jetty (January 1979 - September 2002) and later at an in-lake station (September 2002-ongoing) first with a handheld mercury thermometer (January 1979 - April 1994) and later with an automated probe (April 1994 - ongoing, AD590 temperature transducer (Analog Devices, Norwood, US)). Due to systematic differences between probes and locations,

157 temperature measurements were corrected by +0.258 K for the handheld probe and +0.112 K for
158 the automated probe (Schmidt et al., unpublished data.). Missing values (92 out of 1818, longest
159 gap=10 weeks) were imputed by linear interpolation as they occurred mainly during winter
160 months. A detailed description of sampling and sample processing is given in Driescher et al.
161 (1993) and Gerten and Adrian (2000).

162 *2.1.2. Phytoplankton counting and identification*

163 Phytoplankton samples were fixed with Lugol's solution and counted according to the Utermöhl
164 method (Utermöhl, 1958). From 1979 until 1994 samples were counted mainly at phylum level,
165 with the exception of a few conspicuous diatom and cyanobacteria species. Since 1995,
166 phytoplankton has been counted to species level where feasible; otherwise to higher taxonomic
167 levels, in some cases with additional separation into size classes (centric diatoms, *Cryptomonas*
168 spp, *Aulacoseira* spp, *Peridinium* spp, *Gymnodinium* spp), resulting in a total number of 182 taxa
169 (Table A.1). Biovolume and fresh weight were calculated from cell or filament measurements
170 using the approach described in Padisák and Adrian (1999) and Mischke and Behrend (2007).

171 *2.1.3. Zooplankton counting and identification*

172 Zooplankton samples were concentrated over a 30 µm plankton net (from 20 L to 100 mL) and
173 fixed with Formaldehyde (4 % final concentration). Zooplankton were identified and counted to
174 species-level where feasible, otherwise to higher taxonomic levels, resulting in a total of 105 taxa
175 (Table A.2). Zooplankton abundance data were converted into dry-weight biomass (mg L^{-1}) to
176 properly assess its interactions with phytoplankton biomasses. Because zooplankton body size
177 values required for abundance-biomass conversions were not measured in our study system, we
178 used information from a recently compiled zooplankton trait database (crustacean and copepod
179 data: Hébert et al., in review) and searched Web of Science and Google Scholar databases for

taxa-specific size estimates and dry mass conversion factors for the remaining species (i.e. rotifer species; Table A.3). Several dry mass values were based on taxon-specific length-mass allometric equations, (see Bottrell et al., 1976; Culver et al., 1985; Dumont et al., 1975; McCauley, 1984). For some rotifer species, dry mass estimates were derived from literature biovolume values, assuming a gravity value of 1.025 for biovolume-biomass conversion (Hall et al., 1970; Wetzel and Likens, 2000). For *Leptodora* meta nauplii, the dry mass value was based on stage IV nauplius data (Cummins et al., 1969). For the taxa for which specific information could not be found, body size values were generalized to the genus level. For taxa that reflected general groups (e.g. rotifers spp.), we made assumptions based on generalized information of similar taxonomic resolution (Hall et al., 1970; Hall et al., 1976; Lynch, 1980; Wetzel and Likens, 2000). For non-mature copepods, dry mass estimates were based on all stages of nauplius and copepodite (I-IV and I-V, respectively) of copepod taxa present in our dataset. Due to constraints of the taxonomic identification of daphniid juveniles over the observed period, juveniles were proportionally allocated to the species identified for adult daphniids. We also gathered information about the typical diet of species (i.e. trophic level) from the same data sources, again generalizing genus values when species-specific information was lacking.

2.2. Data analysis

2.2.1. General strategy of the analysis

Our analysis was organized in five consecutive steps. We first assessed changes in phyto- and zooplankton community composition across 34 years by chronological clustering based on yearly averages on genus level, identifying three significantly distinct periods. Second, we grouped phyto- and zooplankton based on taxonomic and trophic information into 13 groups

(representing three trophic levels) for MAR1 modeling. Third, we assessed means and standard errors for biotic and abiotic variables per time period to describe chemical-physical and biotic changes in the lake and differences between periods. Fourth, we used multivariate autoregressive modeling to assess trophic and non-trophic interaction network strengths and stability measures in each period. And fifth, the resulting interaction matrices were passed on to network analysis to assess changes in closeness- and betweenness centrality ranks across periods.

2.2.2. Chronological clustering

To assess community composition changes with chronologically-constrained clustering, we aggregated the phyto- and zooplankton data on genus level, resulting in 61 phytoplankton and 40 zooplankton genera. Clustering was performed on the Euclidian distance matrix of the yearly averages of phytoplankton and zooplankton genus-level datasets separately, using constrained incremental sum of squares (CONISS) clustering (Grimm, 1987; function chclust in R package "rioja"), an agglomerative method that combines adjacent samples (here chronological order of years) while minimizing the increase in total within-cluster sum of squares. To determine the minimum number of clusters, we used a Broken Stick approach as stopping rule (Jackson, 1993; function bstick in R package "vegan"). To assess whether these clusters were significantly different we used ANOSIM on the Euclidian distance matrix with 999 permutations (Oksanen et al., 2007; function anosim in R package "vegan"). The temporal change of all genus time series was visualized by the "traffic-light plot" (Möllmann et al., 2009): Each genus-level time series was transformed into quintiles and then sorted in descending order by the average of the first five years. Note that we used full years of the high taxonomic resolution dataset of phytoplankton (January 1995 – December 2012) and zooplankton (January 1979- December 2012) for the clustering analysis.

225 *2.2.3. Taxonomic and trophic grouping*

226 To adequately populate the MAR1 models for the network analysis of the three time periods, we
227 followed the data preparation steps suggested by Scheef (2013). We aggregated the phyto- and
228 zooplankton taxa into 13 groups to reduce the number of potential parameters estimated in the
229 models. Capitalizing on the full length of the Müggelsee time series (January 1979 – September
230 2013), phytoplankton taxa were aggregated into 6 groups based on phylum (Table A.1):
231 Bacillariophyceae (N taxa = 34), Cyanophyceae (N taxa = 31), Cryptophyceae (N taxa = 14),
232 Chrysophyceae (N taxa = 13), Dinophyceae (N taxa = 17) and Chlorophyceae (including
233 Euglenophyceae and Charophyceae; N taxa = 73). Zooplankton taxa were aggregated into seven
234 groups based on taxonomic and trophic categories: omnivore–herbivore Cladocera (N taxa = 25),
235 Copepoda (N taxa = 12), Rotifera (N taxa = 52) and *Dreissena polymorpha* larvae (N taxa = 1)
236 as grazer groups; and omnivore-carnivore Cladocera (N taxa = 4), Copepoda (N taxa = 9), and
237 Rotifera (N taxa = 2) as predator groups. The omnivore-herbivore groups included species
238 described as herbivores in the literature but also omnivore species feeding on seston (e.g. most
239 rotifers) and the juvenile stages (nauplii and copepodites) of all copepods including those of the
240 carnivorous species (for an overview see supplementary material Table A.2 and for references
241 see Hébert et al., in review and Table A.3). The omnivore-carnivore groups included both,
242 primarily and exclusively carnivore species. We assigned trophic levels to general or genus-
243 based taxa (e.g. *Daphnia* spp.) based on the mean trophic level of species included in this taxon.

244 *2.2.4. Differences between periods*

245 We calculated means and standard errors for abiotic variables, overall phyto- and zooplankton
246 biomass and all 13 phyto- and zooplankton MAR1 groups per period. Differences in means
247 between periods were tested for with a Welch two-sample t-test (Welch, 1947).

248 *2.2.5. Network analysis*

249 We used 13 phyto- and zooplankton groups as variates in the MAR1 models and added water
250 surface temperature, SRP and DIN as exogenous covariates to assess the effects of long-term
251 changes in warming and eutrophication. Additionally, "month" was added as an exogenous
252 covariate to account for seasonality in our models (sensu Ives et al., 1999). All data were
253 aggregated to monthly intervals as this has been shown to efficiently capture time-lagged
254 responses of biotic interactions in other lake networks (Hampton and Schindler, 2006). Missing
255 values were filled with the long-term means for the respective month (phytoplankton 1 out of
256 417 months; zooplankton 18 of 417; SRP 7 of 417, DIN 25 of 417, and temperature 31 of 417).
257 Zeroes were replaced with random values between zero and the lowest observed non-zero value
258 for the respective group. Each time series was log-transformed and then z-scored by subtracting
259 the mean of the group and dividing by the standard deviation of the group (Scheef, 2013;
260 function prepare.data in R package "MAR1"). Log-transformation was applied to linearize
261 trophic interactions among groups (Ives et al., 1999) and z-scoring allowed direct comparison of
262 the interaction coefficients among groups.

263 In MAR1 models, the biomass of each group is predicted by multiple regressions using
264 the values of all other groups and exogenous variables from the previous time step as predictors
265 (Ives et al., 2003; Ives et al., 1999). The matrix formulation of the model is

266 $$X_t = A + BX_{t-1} + CU_{t-1} + E \qquad (1)$$

267 for p interacting groups (variates) and q exogenous groups (covariates) X_t is a p x 1 vector of the
268 z-scored and log-transformed biomasses of each group at time t; A is a p x 1 vector of the
269 intrinsic productivity (here equal to 0 as all time series are z-scored); BX_{t-1} is a p x p matrix of

interaction coefficients b_{ij} that describe how the biomass of group *j* at time *t-1* affects the per unit growth rate of group *i* at time *t*; U_{t-1} is a *q* x 1 vector of covariate values at time *t-1*, and C is the *p* x *q* matrix of coefficients c_{ij} that describe the effect of covariate *j* on group *i*; E is a *p* x 1 vector of process errors assumed to be drawn from a multivariate normal distribution with a mean of 0 and covariance matrix S. Following Ives et al. (1999) and Scheef et al. (2013), 100 models were constructed for each MAR1 group by randomly including or excluding endogenous (B) and exogenous (C) coefficients with equal probability (Scheef, 2013; function run.mar in R package "MAR1"). The best-of-100 model with the lowest Akaike's Information Criterion (AIC) was retained. The process was then repeated 100 times so that finally a single best-fit model out of 10 000 random models was generated. All coefficients that were retained in less than 15% of the best-of-100 models were excluded, and the model selection process was repeated with the remaining coefficients until no further coefficients fell under the 15% exclusion cut-off in the refined best-fit model. Bootstrapping (n=500) provided 95% confidence intervals for the coefficients in the best-fit model. Coefficients which had confidence intervals including zero were eliminated (Hampton and Schindler, 2006). The calculation of stability measures, network visualization and analysis (see below) was based on this final, bootstrapped model.

We did not restrict the sign of the interaction between groups (positive and negative interactions allowed) and explicitly allowed all biologically meaningful interactions, both trophic (interactions of groups of adjacent trophic levels) and non trophic (interactions of groups at the same trophic level). To reduce the number of coefficients estimated in the models, we excluded all direct interactions between predatory zooplankton and phytoplankton producers as well as direct effects of SRP and DIN on all zooplankters. Nevertheless, trophic cascades (bottom up and top down) should be detected as interactions between adjacent trophic levels in the network.

We used network analysis to further evaluate the potential importance of taxonomic groups in the Müggelsee plankton network. Ecological network analysis has been shown to be a useful tool to better understand the structure and functioning of ecosystems because it allows the analysis of graph properties (i.e. topology) of networks and thus helps the interpretation of the importance of cascading effects and non-trophic interactions (Jordán et al., 2008; Vasas and Jordán, 2006). For the purpose of this study, we used two classical network centrality indices: betweenness centrality and closeness centrality (see Table 1). A major assumption in this approach is that well-connected groups in the network based on these two indicators (i.e. higher values imply higher importance) are major interactors with many (strong) links to other groups and therefore exert a more important influence on the network than others (Jordán, 2009). As such, we presume that key groups in the network may be more important than others in maintaining network stability (Jordán and Osváth, 2009). The interaction matrices derived from the MAR1 models were used as input for the network analysis. The network structure was visualized using the 'qgraph' command in the R package "qgraph" (Epskamp et al., 2012) with force-directed layout using the Fruchterman-Reingold algorithm (Fruchterman and Reingold, 1991). Both betweenness and closeness centrality were computed using the R package "sna" (Butts, 2010). We assessed changes in importance of MAR1 groups between time periods by ranking centrality indicators in ascending order (rank 1 being the highest scoring group) and then computed the cumulative sum of rank changes (absolute rank change) between periods.

3. Results

3.1. Chronologically constrained clustering

Based on the CONISS clustering, Broken Stick suggested a minimum of two clusters in the phytoplankton dataset: 1995-2005; 2006-2012 and three clusters in the zooplankton dataset: 1979-1995; 1996-2008; 2009-2012. The clusters differed significantly based on one-way ANOSIM: phytoplankton: R=0.77, p=0.001, zooplankton: R=0.80, p=0.001 (Fig. 1). Based on the clustering result we divided the data into three periods: P1: 1979-1995; P2: 1996-2005; and P3: 2006-2013.

3.2. Mean differences between periods

Period 1 was characterized by high TP and DIN concentrations in the lake and low transparency. Phytoplankton biomass was high, mainly consisting of Bacillariophyceae, Cyanophyceae and Cryptophyceae (see also Table 2). Cladoceran herbivores and *Dreissena* larvae contributed most biomass to the herbivores. In period 2, TP and DIN concentrations were reduced (-19 % and -57 %, respectively). Overall phytoplankton biomass declined (-57 %) as did Cyanophyceae (-66 %), Bacillariophyceae (-56 %) and Cryptophyceae (-43 %) biomass. Chrysophyceae increased (1230%), although they did not reach substantial biomass (period 2 mean = 0.24 mg L^{-1}). Overall water transparency improved by 120 %. While herbivore Cladocera biomass decreased (-38 %), *Dreissena* larvae increased in biomass (+330 %) and became the largest contributor to grazer biomass. Period 3 was characterized by a decrease in water transparency and an increase in herbivorous (+170 %) and carnivorous (+189 %) Rotifera biomasses. Although yearly average Cyanophyceae biomass did not change significantly, the dominant species switched from *Aphanizomenon flos aquae* to *Planktothrix agardhii* (Table A.2). In the predatory Cladocera,

334 *Leptodora kindtii* became dominant instead of *Bythotrephes spp* (Table A.2). For an overview of
335 the seasonal and long-term dynamics, we present time series and yearly dynamics of
336 temperature, SRP and DIN as well as MAR1 group biomasses in Fig. 2.

337 ***3.3. Network analysis***

338 We fitted MAR1 models to data of three consecutive periods using 13 biotic groups as variates
339 and four environmental variables as covariates. The AIC best fitting and bootstrapped model
340 conditional R^2 for P1 ranged from 0.32 to 0.54 (median = 0.42), for P2 from 0.19 to 0.67
341 (median = 0.47) and for P3 from 0.25 to 0.57 (median = 0.41) (see Table A.4). The number of
342 non-zero interaction coefficients decreased over time (Table 3).

343 The interaction coefficients of trophic (between trophic levels: e.g. bottom up or top
344 down) and non-trophic (within trophic levels, e.g. competition or facilitation) processes are
345 summarized in Fig. 3. The bottom-up processes of phytoplankton-grazer (N per period= 5; 4; 6)
346 and grazer-predator (N per period= 5; 5; 3) showed mostly positive interactions, generally
347 indicating that increases in prey preceded increases in consumers at the next time step.
348 Generally, the strength of positive bottom-up interactions also seemed to increase over time.
349 However, a consistent negative interaction of herbivorous Cladocera on carnivorous Copepoda
350 was found in all periods (Fig. 4). Top-down processes of grazer-phytoplankton (N per period=
351 13; 2; 6) and predator-grazer (N per period= 4; 4; 5) however, also showed mostly positive
352 coefficients, suggesting that increases in consumers often preceded increases in their prey.
353 Specifically, all *Dreissena*-phytoplankton interaction coefficients as well as all carnivorous
354 Copepoda and Rotifera interactions with grazers were positive (Fig 4). The effects of
355 herbivorous zooplankton groups on phytoplankton groups were variable, showing both positive

and negative interactions. Non-trophic interactions were summarized for each trophic level separately, excluding the interaction coefficients of MAR1 groups with themselves (i.e. density dependence). Non-trophic interactions for phytoplankton (N per period= 7; 4; 3), zooplankton grazers (N per period= 5; 5; 5) and zooplankton predators (N per period= 2; 5; 2) showed positive as well as negative coefficients and were not consistent in signs across periods (Fig 3). Only carnivorous Cladocera had a consistent and strong negative effect on carnivorous Rotifera (Fig 4). The effect of each MAR1 group on itself indicated the strength of density dependence (N per period = 13; 11; 8) and ranged from 0.14 (carnivorous Rotifera, P1) to 0.84 (carnivorous Copepoda, P3); however, in MAR1 models of P2 and 3, not all density dependence coefficients were retained in the final bootstrapped model (Table A.4), suggesting that density dependent control in these groups was weak or not consistent during these periods. The effect coefficients of the environmental covariates surface water temperature (N per period = 0;4;0), SRP (N per period = 2;0;2), and DIN (N per period = 1;0;0) again varied and were not consistent across periods. Month accounted for seasonality in our models (N per period = 8; 3; 8) and showed multiple and strong interactions with all trophic levels (Table A.4). Generally, the P1 interaction network appeared less stable than the P2 and P3 as measures for resilience (return rate to stationary distribution after a perturbation) and reactivity (short term response to a perturbation) decreased from P1 to P2 and 3 (Table 4).

The visualization of the three networks using the Fruchterman-Reingold layout is shown in Fig. 4. The analysis of these networks revealed that *Dreissena* generally ranked first for both, closeness and betweenness centrality, except for closeness centrality in P3 where it ranked second (Fig. 5). Cladocera herbivores showed also high closeness centrality values across all periods ranking second in P1 and 2 and first in P3. Most groups were variable in closeness- and

betweenness centrality and the absolute rank change (i.e. cumulative sum of rank changes) across all periods ranged from 0 (closeness centrality for Cyanophyceae) to 19 (betweenness centrality for Bacillariophyceae). Few groups displayed consistent rank changes across periods. For example, copepod predators increased rank for closeness centrality from 13 (P1) to 3 (P3). Similarly, Dinophyceae increased in betweenness centrality rank from 13 (P1) to 4 (P3). Cryptophyceae decreased in closeness centrality ranks from 4 (P1) to 13 in (P3). Analogously, betweenness centrality ranks for this group also decreased consistently from 6 (P1) to 12 (P3).

4. Discussion

Here we explored changes in the pelagic plankton network structure and stability in the shallow temperate lake Müggelsee, which has undergone changes in eutrophication status and experienced a significant increase in surface water temperature over the last 34 years (Köhler et al., 2005; Wagner and Adrian, 2009). Using multivariate first order autoregressive (MAR1) modelling and ecological network analysis on 16 biotic and abiotic variables, we were able to show that the planktonic interaction network is still primarily driven by bottom-up processes. Furthermore, indirect and non-trophic interactions were at least as important as direct and trophic interactions in determining the structure and stability of the Müggelsee network. Moreover, the larvae of the invasive freshwater mussel *Dreissena polymorpha* were identified as a keystone group as they occupied the highest ranks in both closeness and betweenness centrality in the pelagic network during all three periods. Thus, these larvae affect and are affected by most planktonic groups in the pelagic network, and are therefore likely to play a critical role in community structure and stability. Based on rank changes in centrality indicators, we could also identify groups that responded strongly to environmental change such as Bacillariophyceae and

401 Chlorophyceae in the phytoplankton, or rotifer and copepod predators in the zooplankton. Given
402 the observed complexity of direct and indirect interactions, we here emphasize the need for long-
403 term ecological observations combined with a holistic approach in data analysis to assess the
404 effects of environmental change such as climate change on ecological networks and their
405 functioning, which cannot be mimicked in short-term experiments.

406 ***4.1. Network dynamics***

407 *4.1.1. Multivariate first order autoregressive models*

408 The three interaction networks derived from the MAR1 models showed a decreasing number of
409 interactions over time, although some interactions were retained in all three networks. Broadly,
410 we observed consistent positive resource-consumer interactions across all three periods,
411 suggesting that the biomass of consumers was sensitive to the biomass of resources in the
412 preceding month. However, consumer-resource interactions were both negative (i.e. top-down
413 control) as well as positive. In particular, all interactions of *Dreissena* larvae and phytoplankton
414 groups as well as almost all interactions of predatory zooplankton with herbivorous zooplankton
415 were positive. Positive effects of consumers on their resources may be brought about by varous
416 mechanisms such as consumers increasing nutrient cycling (Kitchell et al., 1979), consumer-
417 resource indirect facilitation scenarios (Abrams, 1992), inference competition or intra-guild
418 predation scenarios including either mutual (i.e. both predators prey on each other) or
419 hierarchical (one top predator preys on the intermediate predator) intra-guild predation (Vance-
420 Chalcraft et al., 2007). The aggregation of the data to monthly intervals may also have
421 contributed to the increased detection of indirect effects as these take longer to take effect.
422 Although the observed positive consumer-resource interactions may also have resulted from a

shared third variable such as an environmental driver, they nevertheless suggest that consumers were not able to control resource biomasses efficiently nor consistently. Overall, these results support a previous study that characterized phytoplankton and zooplankton biomass in Müggelsee as bottom-up regulated (Köhler et al., 2005). As MAR1-based interaction coefficients represent sustained (across seasons and years) interactions among large (and in our case sometimes quite heterogeneous) groups, interactions that are important for a short period per year, or those that are not consistent among years, tend to be eliminated during the model searching process. Despite the suggested overall lack of top-down control in our models, we cannot discount the importance of grazing during shorter periods of time, such as periods preceding the clear-water phase (Gerten and Adrian, 2000).

The MAR1 model results showed numerous interaction outcomes that are usually less commonly quantified. Positive effects of consumers on resources (e.g. *Dreissena* larvae on phytoplankton groups in all three periods) and negative effects of resources on consumers (e.g. herbivore Cladocera on predatory Copepoda) occurred in all three periods. The latter may have been brought about by resource competition between herbivore cladocerans and herbivorous juvenile stages of predatory copepods. Likewise, the models also suggested a predominance of positive interactions among groups of the same trophic level (e.g. *Dreissena* larvae on herbivore rotifers in periods 1 and 2), which may indicate direct facilitation (Brooker et al., 2008) or reflect indirect interactions such as competitive mutualism (McCormick and Stevenson, 1991). However, negative interaction within the same trophic level also occurred (e.g. between carnivorous cladocerans and carnivorous rotifers) which may have resulted from either interference competition or from intra-guild predation on the juveniles of the competitor (Arndt et al., 1993). Such negative interactions between consumers may also explain some of the

446 seemingly positive effects of consumers on their prey. Interference competition may lead to
447 positive effects on prey groups of the inferior competitor as interference in prey searching may
448 lead to an overall reduction in predation pressure for that prey (Sih et al., 1998). Intra-guild
449 predation among predators has been found to release prey under either mutual or hierarchical
450 intra-guild predation constellations (Vance-Chalcraft et al., 2007). Although apparently direct
451 interactions may have been caused by a range of indirect mechanisms which we can not identify
452 without laboratory experiments, our model results suggested that trophic and non-trophic
453 interactions are equally present across time, suggesting that both types of interactions regulate
454 population dynamics. This calls for a more integrative approach when assessing the effects of
455 environmental changes on networks or, conversely, extrapolating individual responses of species
456 to environmental changes to communities and ecosystem levels.

457 Our choice of 13 pelagic plankton groups in the MAR1 models yielded relatively
458 complex networks which resulted in uncertain interpretation of some interactions. Despite this,
459 the overall network complexity was still moderate given that our analysis focused on the pelagic
460 plankton interaction network and thus omitted other potentially important pelagic, littoral or
461 benthic organisms such as bacteria, fish, parasites, benthic macrofauna, or macrophytes that may
462 be crucial for explaining the ecosystem response to environmental change (e.g. Jeppesen et al.,
463 1998). In our study, the number of groups was restricted to aggregated taxonomic groups to
464 reduce the risk of over-parameterizing the models, and to improve the power of the analysis. As
465 a result, some of the groups were rather heterogeneous comprising many taxa (see Table A.1 and
466 A.2). Such constraints also hindered the assessment of the role of intra-group interactions that
467 may have affected the overall correlation of the group with other groups. Interactions among taxa
468 within their respective groups may be particularly concealed in the overall interaction

coefficients of the MAR1 groups, such as the effects of intra-guild predation as observed in the dominance switch between two omnivore-carnivore copopods *Cyclops vicinus* and *Cyclops kolensis* due to a reduction of shared phytoplankton resources (Scharfenberger et al., 2013).

4.1.2. Network stability indicators

The MAR1 results for stability measures suggested that the period 1 network was less stable than the period 2 and 3 networks. The period 1 network was more reactive to perturbations (for example heat waves or storm events) and took longer to return to its 'equilibrium' state than period 2 and 3 networks. The stability measures are derived from eigenvalues of the interaction matrix (B) (Ives et al., 2003). The variance indicator takes all eigenvalues in the system into account and is therefore sensitive to small eigenvalues. In contrast, the resilience and reactivity indicators are both strongly influenced by the dominant eigenvalue in the system with large dominant eigenvalues corresponding to the 'slowest' dimension in the system (Ives et al., 2003). The maximum eigenvalue in the period 1 network was larger than those of periods 2 and 3, making return time to the 'equilibrium' state slow. Smaller maximum eigenvalues and overall smaller eigenvalues of the interaction matrices of periods 2 and 3 reduced return times as well as reactivity. This suggests that the networks of periods 2 and 3 responded less strongly to perturbations and returned faster to their 'equilibrium' state since the interactions in the networks did not greatly amplify the effect of environmental variability. This may reflect the different trophic states the lake has gone through, from hyper-eutrophic in the first period (less stable) to an intermediate trophic state in the 1990ties (highest stability) and a more eutrophic state in the last period (slightly less stable again). Such an increase in stability with a reduction of nutrient load has also been reported for the Lake Washington food web by Francis and coauthors (2014).

4.1.3. Network centrality indicator ranks

Rank changes in centrality indicators can be interpreted as a sign of the responsiveness to changes in the environment and reflect changes in the relative role of a group within the network through time (Jordán and Osváth, 2009). Particularly changes in well-connected groups (closeness centrality), or in groups that are key in connecting otherwise little-connected parts of the network (betweenness centrality), are likely to have cascading effects through the network (Solé and Montoya, 2001). Here, high values of closeness centrality were found for many herbivorous grazers and phytoplankton groups in all periods. Similar results were obtained for betweenness centrality for which mostly herbivorous grazers and phytoplankton groups showed high values. The topological importance of herbivores in the Müggelsee network is consistent with their functional importance in food-chain dynamics (Polis and Strong, 1996). It should be noted however, that the centrality of herbivores in our MAR1 models may be somewhat overestimated as we did not allow for direct interactions between carnivorous zooplankton and phytoplankton producers, and as such, the number of possible interactions was larger for herbivores than for other trophic levels.

Dreissena polymorpha larvae appeared to be the most influential group in the Müggelsee network based on closeness and betweenness centrality ranks, with herbivorous cladocerans as close second. *Dreissena* larvae ranked persistently high throughout all periods, despite changes in biomass between periods (i.e. significant increase between periods 1 and 2). This continuous increase in the abundance of *Dreissena* larvae in Müggelsee co-occurred with a phenological shift in the first spawning event advancing by about two weeks, and in turn, an extension of its pelagic life phase (Adrian et al., 2006; Wilhelm and Adrian, 2007). This phenological shift has likely given *Dreissena* larvae a competitive advantage over filter-feeding cladocerans during

514 spring (Adrian et al., 2006). While the individual filtration capacity of *Dreissena* larvae is lower
515 than that of cladocerans by a factor 10-30 (MacIsaac et al., 1992), the higher overall biomass of
516 *Dreissena* larvae as compared to the herbivorous cladocerans (see Fig. 2) may have resulted in
517 similar or even higher grazing pressure which in turn would explain their prominent role in the
518 Müggelsee pelagic network. Their central network position may furthermore be explained by
519 their susceptibility to predation by calanoid copepods as observed in the Great Lakes (Liebig and
520 Vanderploeg, 1995) and their ability to feed on a wide variety of potential food (albeit within a
521 narrow size range) including bacteria, cyanobacteria, chlorophytes, rotifers and detritus (Sprung,
522 1993). Given the centrality of the *Dreissena* larvae in the pelagic network of Müggelsee and their
523 significance in benthic littoral food webs observed in other lakes (Ozersky et al., 2012), the
524 implementation of long-term monitoring of all life stages is a prerequisite for fully understanding
525 the effect and success of this invader on ecosystem dynamics.

526 Groups that are particularly sensitive to changes in the environment were expected to
527 change centrality ranks quite dynamically. The predatory Rotifera provided one example of a
528 group shifting from rank 5 to 3 to 10 in closeness centrality and from rank 8 to 3 to 13 in
529 betweenness centrality. The dynamic position of this group in the Müggelsee network may
530 partially be explained by a dominance change in the group of its cladoceran predator. The
531 dominant species in the group of predatory rotifers was *Asplanchna* sp. which was negatively
532 affected by predatory cladocerans in all three periods. While the predatory cladoceran *Leptodora*
533 *kindtii* was dominant in periods 1 and 3, *Bythotrephes* dominated in period 2. As these
534 cladoceran predators differ in their feeding and phenological traits (Branstrator, 2005) such a
535 dominance shift may affect the dynamics of the groups they interact with.

Groups that are apparently less sensitive to environmental change were expected to maintain a constant rank over time. Despite its high biomass throughout most of spring - and summer periods, Cyanophyceae provided an example for a group that was neither central nor shifted ranks over time. Cyanobacteria are well-known for their comparably low edibility and most species within this group have anti-grazer defenses by colony formation or toxin synthesis and secretion. These mechanisms may result in a decoupling of Cyanobacteria dynamics from herbivore dynamics and therefore in a less central position within the network, and may also explain why Cyanobacteria can develop such high biomasses while neither being a central group nor changing their network position. While *Aphanizomenon flos-aqua*e was the dominant species during period 2, *Planktothrix agardhii* dominated in period 3. Such a species shift without changes in group biomass or centrality rank may hint at a compensatory effect.

4.2. Implications and outlook

Aquatic ecosystem networks are undoubtedly and ubiquitously complex. Our results suggest that both, trophic and non-trophic interactions are commonly present and of similar interaction strengths, and hence important for structuring the topology as well as determining the stability of pelagic interaction networks. This integrative view of different types of interactions in communities is supported by findings from aquatic mesocosm experiments (Hammill et al., 2015) and terrestrial plant food webs (Ohgushi, 2008). However, the network analysis of the pelagic network of Müggelsee over a 34 year period leaves us with a long standing conclusion of studying such systems: namely to “realize that everything connects to everything else” - Leonardo da Vinci (1452-1519). Although we can not identify the mechanisms underlying many of the observed network interactions, the analysis nonetheless documented intricate relationships among ecosystem components with regards to the importance of indirect interactions in

559 structuring aquatic networks and the central role of an invasive species in the case of the
560 Müggelsee pelagic plankton network.

561 These insights into ecosystem-level behavior and dynamics were enabled through the use
562 of long-term observational data, which have provided ecologists with a valuable tool to
563 understand ecosystem-level responses to anthropogenic pressures over prolonged time scales.
564 This is an important point to reiterate (cf. Lindenmayer et al., 2012; Magnuson, 1990), because
565 often only mechanisms of single system-level dynamics in, for example, climate impact research
566 are well explored for individual case studies. Nevertheless, understanding major drivers of
567 networks remains difficult as it is shown in this study. There also appears to be a trade-off with
568 regards to the level of interpretability of the mechanisms that can indeed be reached in specific
569 system-level studies compared to the level of understanding that can be obtained through a more
570 holistic approach. Despite these limitations, our results show that network stability and centrality
571 rank positions do change over time and may serve as potential "sentinel" variables for climate
572 impact monitoring (Adrian et al., 2009). Future endeavors may address the current limits to
573 interpretation by utilizing a combined approach of experimental, modelling and observational
574 studies to identify the mechanisms underlying some of the less easily explained interactions
575 identified in the MAR1 models (e.g. positive interactions between zooplankton predator groups)
576 and to assess the validity of the observed interactions to improve their interpretation and
577 predictability under climate change scenarios. Moreover, experiments may be used to assess how
578 network centrality measures are linked to numerical or functional importance of organisms.
579 Based on the quality of such relationships, centrality measures may serve as indicators of
580 reconfigurations in networks under pressure.

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Figure captions:

Fig. 1: Quintile plots and CONISS clustering dendrograms of the chronological clustering for phytoplankton and zooplankton. Quintile plots are based on time series of annual genus level averages transformed into quintiles and sorted by descending averages of the first five years. Light greys indicate low value, dark greys indicate higher values. Clustering was based on the Euclidian distance matrix of genus level yearly averages using constrained incremental sum of squares (CONISS) clustering. Number of clusters was assessed using Broken Stick. Clusters are denoted by horizontal dashed lines.

Fig. 2: Time series graphs of MAR1 analysis variates and covariates. Line plots present time series of monthly mean values of the respective variable and the corresponding box plots present median values of the monthly data across all years. For the box plots, data were log10 transformed and then scaled between 0 and 1 to emphasize the seasonal dynamics. Horizontal lines in the box plots denote the medians; boxes denote the 25th and 75th percentile; the whiskers denote non outlier range, circles are outliers.

Fig. 3: Boxplots of interaction coefficients for each period (P1, P2, P3) categorized by interaction type: trophic: bottom up or top down; non trophic: competition: among groups of the same trophic level; abiotic: environment-group effects; season: season-group effects. Horizontal lines in the box plots denote the medians; boxes denote the 25th and 75th percentile; the whiskers denote non-outlier range, circles are outliers.

Fig. 4: Interaction networks based on the best-fit MAR1 model for each period using a Fruchterman-Reingold layout. Line thickness quantifies interaction strength (see table A.4). Arrows point towards the response group. Dashed lines are negative; solid lines are positive

effects. Zooplankton groups are represented by illustrations; *Dreissena* are represented in their adult form. Phytoplankton groups are represented in gray boxes with abbreviations: Bacillariophyceae (Dia), Cyanophyceae (Cyn), Cryptophyceae (Cry), Chrysophyceae (Chr), Dinophyceae (Din) and Chlorophyceae (Chl). Environmental covariates are encircled and the abbreviations “P”, ”N” and “Tmp” correspond to SRP, DIN and water surface temperature, respectively. Month was included for the calculation of interaction strengths and network layout but subsequently removed from the graph for clarity along with environmental covariates not retained in the best-fit MAR1 models (for values: table A.4).

Fig. 5: Slope graphs of rank lists for closeness centrality and betweenness centrality of all MAR1 groups per time period. Highest values for both indicators have the lowest rank and are considered important organisms in the interaction network. Grey lines are constant ranks and positive rank changes and black lines are negative rank changes between periods. Absolute rank change is the cumulative sum of rank changes over the whole time period.

820 # Tables

821 Table 1: Ecological indicators used in this study to describe changes in the Müggelsee network stability
822 and topology

Ecological indicator	Description	Ecological significance	Key references
Variance	The lower the stationary distribution variance in relation to the environmental variance, the more stable the system. The determinant of the interaction matrix ('DetB') shows how much group (or species) interactions increase the variance of the stationary distribution relative to that of the environmental noise (i.e. stability increases with decreasing DetB).	Unstable systems with low resilience (slow return to its stationary distribution) and low resistance (high reactivity) tend to fluctuate more strongly as species interactions amplify the system response to environmental variation.	(Ives et al., 2003)
Resilience	The dominant eigenvalue of the Kronecker product B⊗B ('maxeigen KrB') limits the return rate of the community to its stationary distribution after a perturbation. Resilience increases as return rate increases (i.e.'maxeigen KrB' decreases)	More stable systems return to their 'equilibrium' state more quickly after a perturbation (e.g. heat waves, storms etc) than unstable ones.	(Ives et al., 2003)
Reactivity	The maximum eigenvalue of the interaction matrix B ('maxeigen BxB') represents the potential maximal reaction strength of a system to a perturbation. Resistance increases as reactivity decreases.	Unstable systems show larger deviations form the stationary distribution after perturbations.	(Ives et al., 2003)
Closeness centrality	This indicator emphasizes the distance from each vertex to every other vertex in the network. A vertex with the direct connection to every other vertex in the network will have a high closeness value, whereas a vertex which is connected to other vertices through many intermediaries will have a low closeness value.	Closeness centrality focuses on the strength of influence over the entire network, changes in organisms with high closeness centrality values influence the network dynamics more than changes in organisms with lower values.	(Jordán et al., 2008; Vasas and Jordán, 2006)
Betweenness centrality	This indicator is derived from the number of shortest paths passing through a given vertex (intermediary). To calculate betweenness centrality, all the shortest paths between any two vertices in the network are found and then the number of these shortest paths that go through each vertex is counted.	Groups with high betweenness centrality are not necessarily connected directly to all other vertices. High betweenness groups are considered important because they provide (the only) link between otherwise unconnected network vertices.	(Jordán et al., 2008; Vasas and Jordán, 2006)

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Table 2: Summary statistics of Müggelsee variables for periods P1: 1979-1995, P2: 1996-2005 and P3: 2006-2012: Mean (Mean) and standard error (SE) are reported for all environmental and biotic variables per period. Difference in the means between P1 and P2 as well as P2 and P3 were tested with a Welch two sample t-test (p<0.05)

	Period 1		**Period 2**		**Period 3**					
	1979-1995		**1996-2005**		**2006-2012**		**Period 1 vs Period 2**		**Period 2 vs Period 3**	
Variable	**Mean**	**SE**	**Mean**	**SE**	**MEAN**	**SE**	**t-test**	***p-value***	**t-test**	***p-value***
Temperature (°C)	10.88	0.48	10.94	0.67	11.34	0.82	t(24.3) = -0.27	0.79	t(14.6) = -1.95	0.07
SRP (µg L^{-1})	66.87	6.00	65.14	5.37	74.54	9.35	t(23.6) = 0.16	0.88	t (9.8) = -0.85	0.41
TP (µg L^{-1})	154.78	7.76	125.31	6.94	132.75	11.08	t(24) = 2.08	**0.047**	t(10.3)= -0.53	0.61
DIN (mg L^{-1})	1.24	0.09	0.531	0.04	0.439	0.05	t(20.1) = 3.86	< **0.001**	t(14.3) = 0.98	0.34
RSi (mg L^{-1})	4.02	0.16	4.27	0.19	4.81	0.28	t(20.3) = -0.91	0.37	t(7.8) = -0.91	0.39
Secchi (m)	1.85	0.06	2.21	0.08	1.82	0.09	t(24.1) = -4.21	< **0.001**	t(7.9)= 3.26	**0.01**
Phytoplankton biomass (mg L^{-1})	10.42	0.76	4.40	0.34	4.84	0.57	t(21.7) = 7.14	< **0.001**	t(10.2) = -0.6584	0.52
Zooplankton biomass (mg L^{-1})	0.21	0.01	0.22	0.01	0.24	0.02	t(24.9) = -0.47	0.64	t(8.2) = -0.9522	0.36
Cyanophyceae (mg L^{-1})	3.50	0.38	1.18	0.24	1.14	0.23	t(23.1) = 3.89	< **0.001**	t(14.7) = 0.12	0.91
Bacillariophyceae (mg L^{-1})	5.30	0.42	2.33	0.23	2.74	0.38	t(23.1) = 7.31	< **0.001**	t(10.4) = -0.75	0.47
Chrysophyceae (mg L^{-1})	0.018	0.01	0.240	0.07	0.088	0.01	t(9.3) = -2.45	**0.035**	t(9.2) = 1.69	0.13
Dinophyceae (mg L^{-1})	0.084	0.01	0.091	0.02	0.189	0.06	t(12.4) = -0.2	0.84	t(7.4) = -1.07	0.32
Cryptophyceae (mg L^{-1})	0.840	0.05	0.478	0.04	0.430	0.04	t(22.9) = 4.28	< **0.001**	t(13.8) = 0.54	0.60
Chlorophyceae (mg L^{-1})	0.141	0.01	0.168	0.03	0.214	0.06	t(11.7) = -0.56	0.58	t(11.3) = -0.58	0.57
Cladocera herbivore (mg L^{-1})	0.016	0.001	0.010	0.001	0.012	0.002	t(23.3) = 3.08	**0.005**	t(9.3) = -0.87	0.41
Copepod herbivore (mg L^{-1})	0.005	0.0003	0.006	0.0004	0.0051	0.0004	t(16.4) = -2.75	**0.01**	t(8.6) = 0.95	0.37
Rotifer herbivore (mg L^{-1})	0.003	0.0003	0.003	0.0003	0.0051	0.0006	t(23.5) = 1.14	0.27	t(7.8) = -3.01	**0.02**
Dreissena larvae (mg L^{-1})	0.011	0.0019	0.047	0.0099	0.037	0.0093	t(10) = -4.67	< **0.001**	t(14.9) = 1.04	0.31
Cladocera predator (mg L^{-1})	0.003	0.0005	0.001	0.0002	0.0024	0.0004	t(25) = 0.12	0.91	t(6.7) = -1.67	0.14
Copepod predator (mg L^{-1})	0.010	0.0009	0.013	0.0015	0.012	0.0019	t(19.6) = -1.39	0.18	t(11.9) = 0.36	0.73
Rotifer predator (mg L^{-1})	0.0009	0.0002	0.0009	0.0002	0.0017	0.0005	t(22.2) = 3.86	< **0.001**	t(11.6) = -2.60	**0.02**

Table 3: Summary of total number of possible interactions in the B matrix (i.e. interaction matrix) and C matrix (i.e. covariate effects matrix) per period: number of interaction coefficients equal zero (no interaction retained in the bootstrapped model), number of positive interaction coefficients (increases in the predictor at *t-1* are related to increases in the respondent at *t*), and number of negative interaction coefficients (increases in the predictor at *t-1* are related to decreases in the respondent at *t*).

	Period 1 (1979-1995)			**Period 2 (1996-2005)**			**Period 3 (2006-2012)**		
	Total	**B**	**C**	**Total**	**B**	**C**	**Total**	**B**	**C**
Total no. of coefficients	221	169	52	221	169	52	221	169	52
No. coefficients = 0	156	115	41	174	129	45	173	131	42
No. coefficients > 0	46	40	6	37	34	3	35	31	4
No. coefficients < 0	19	14	5	10	6	4	13	7	6

867 Table 4: Summary of stability measures derived from MAR1 models for each period. Decreasing values
868 for the variance, resilience and reactivity indicators suggest increasing stability of the network.

Stability measure	Indicator	Period 1 (1979-1995)	Period 2 (1996-2005)	Period 3 (2006-2013)
Variance	DetB	0.14	0.13	0.12
Resilience	maxeigen KrB	0.89	0.42	0.52
Reactivity	maxeigen BxB	0.83	0.53	0.58

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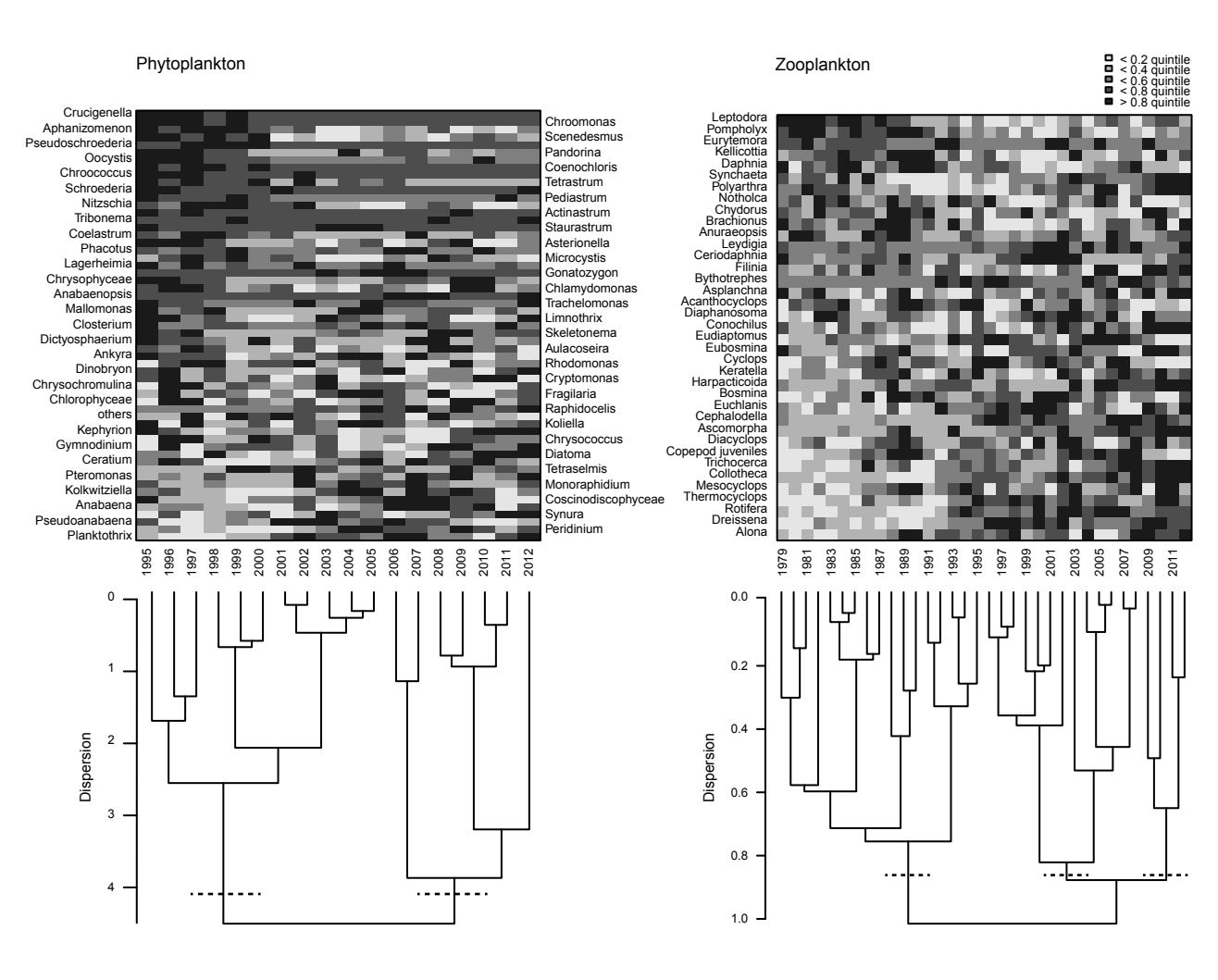
Phytoplankton
Crucigenella
Aphanizomenon
Pseudoschroederia
Oocystis
Chroococcus
Schroederia
Nitzschia
Tribonema
Coelastrum
Phacotus
Lagerheimia
Chrysophyceae
Anabaenopsis
Mallomonas
Closterium
Dictyosphaerium
Ankyra
Dinobryon
Chrysochromulina
Chlorophyceae
others
Kephyrion
Gymnodinium
Ceratium
Pteromonas
Kolkwitziella
Anabaena
Pseudoanabaena
Planktothrix
Chroomonas
Scenedesmus
Pandorina
Coenochloris
Tetrastrum
Pediastrum
Actinastrum
Staurastrum
Asterionella
Microcystis
Gonatozygon
Chlamydomonas
Trachelomonas
Limnothrix
Skeletonema
Aulacoseira
Rhodomonas
Cryptomonas
Fragilaria
Raphidocelis
Koliella
Chrysococcus
Diatoma
Tetraselmis
Monoraphidium
Coscinodiscophyceae
Synura
Peridinium
1995
1996
1997
1998
1999
2000
2001
2002
2003
2004
2005
2006
2007
2008
2009
2010
2011
2012
Dispersion
0
1
2
3
4
Zooplankton
< 0.2 quintile
< 0.4 quintile
< 0.6 quintile
< 0.8 quintile
> 0.8 quintile
Leptodora
Pompholyx
Eurytemora
Kellicottia
Daphnia
Synchaeta
Polyarthra
Notholca
Chydorus
Brachionus
Anuraeopsis
Leydigia
Ceriodaphnia
Filinia
Bythotrephes
Asplanchna
Acanthocyclops
Diaphanosoma
Conochilus
Eudiaptomus
Eubosmina
Cyclops
Keratella
Harpacticoida
Bosmina
Euchlanis
Cephalodella
Ascomorpha
Diacyclops
Copepod juveniles
Trichocerca
Collotheca
Mesocyclops
Thermocyclops
Rotifera
Dreissena
Alona
1979
1981
1983
1985
1987
1989
1991
1993
1995
1997
1999
2001
2003
2005
2007
2009
2011
Dispersion
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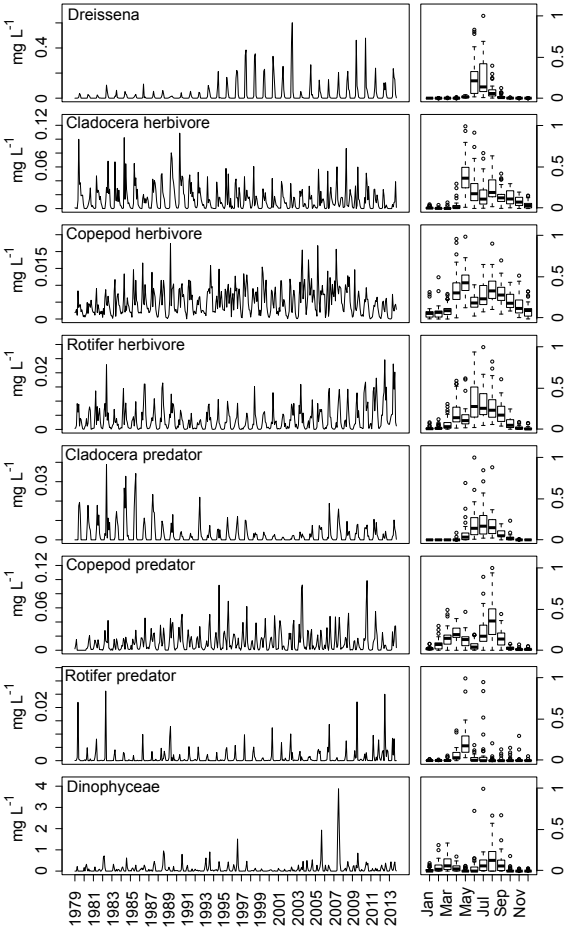
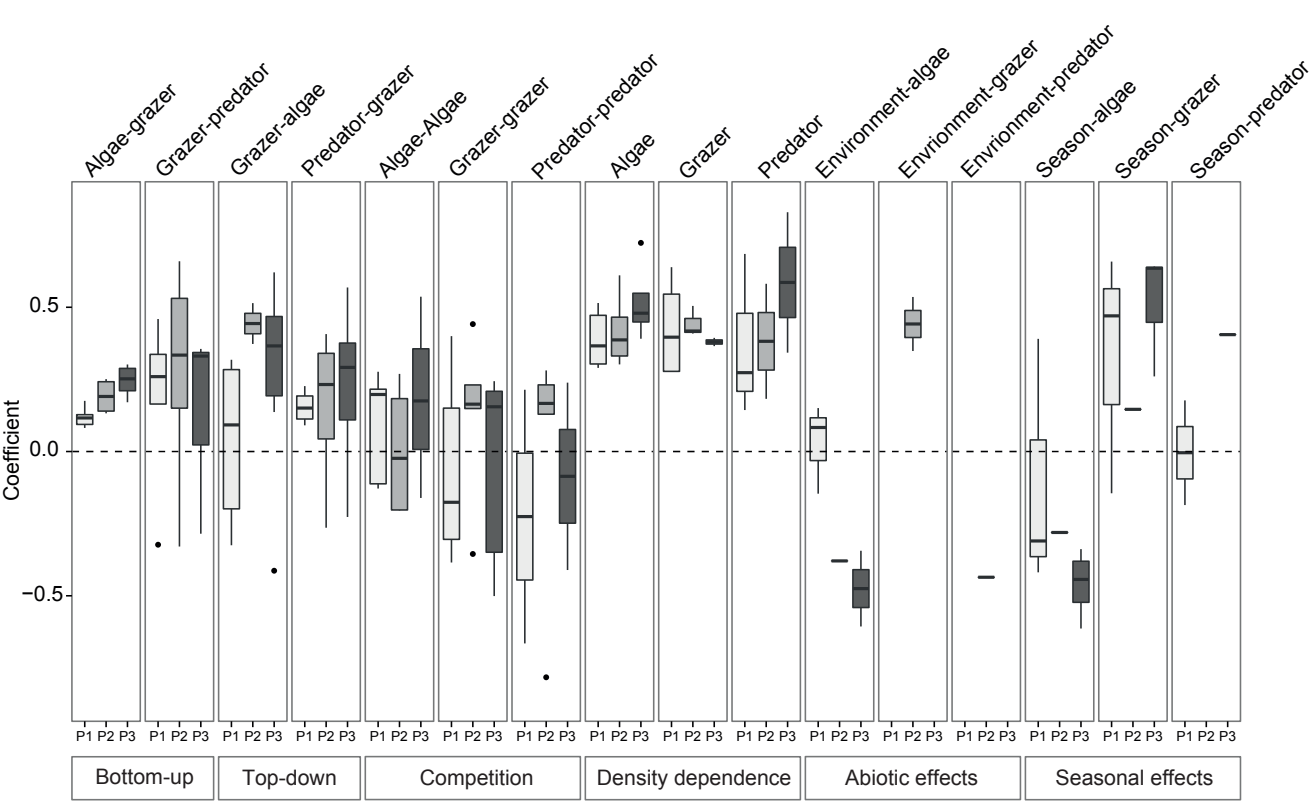
1.0


Dreissena
Cladocera herbivore
Copepod herbivore
Rotifer herbivore
Cladocera predator
Copepod predator
Rotifer predator
Dinophyceae
Cryptophyceae
Chlorophyceae
Cyanophyceae
Bacillariophyceae
Chrysophyceae
Temperature
SRP
DIN
mg L-1
µg L-1
°C
1979 1981 1983 1985 1987 1989 1991 1993 1995 1997 1999 2001 2003 2005 2007 2009 2011 2013
Jan Mar May Jul Sep Nov


Algae-grazer
Grazer-predator
Grazer-algae
Predator-grazer
Algae-Algae
Grazer-grazer
Predator-predator
Algae
Grazer
Predator
Environment-algae
Envrionment-grazer
Envrionment-predator
Season-algae
Season-grazer
Season-predator
Coefficient
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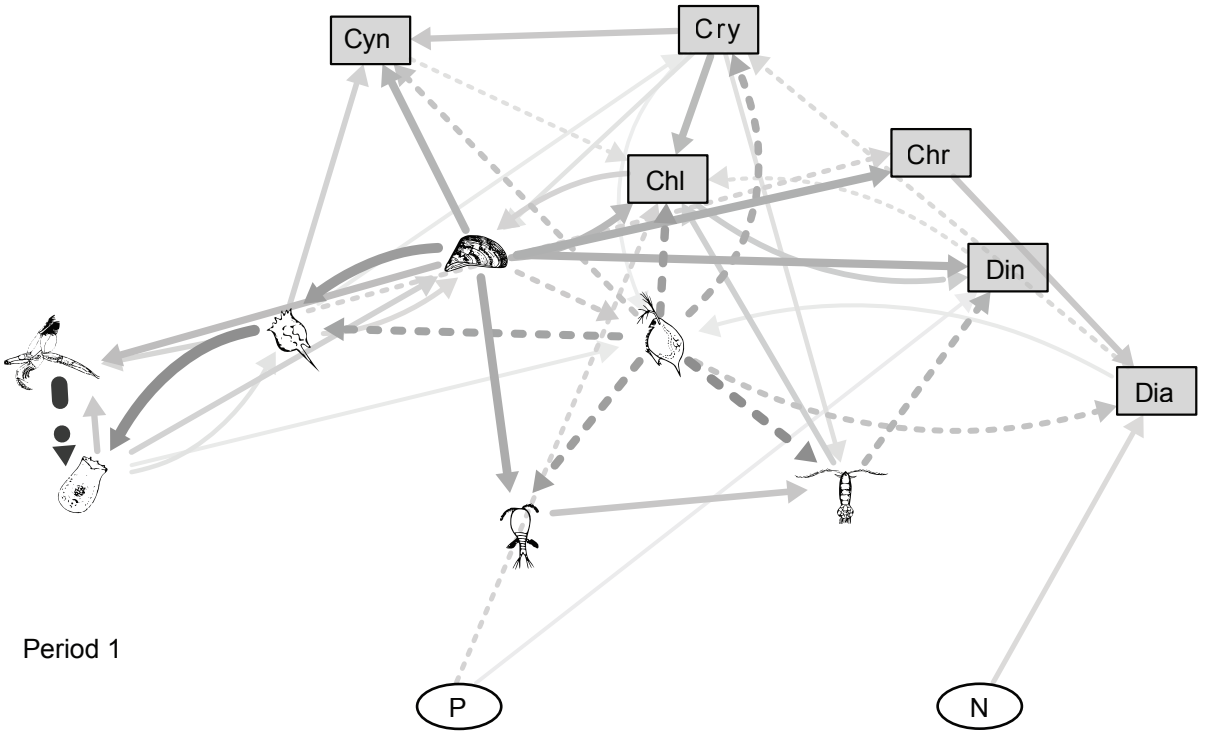
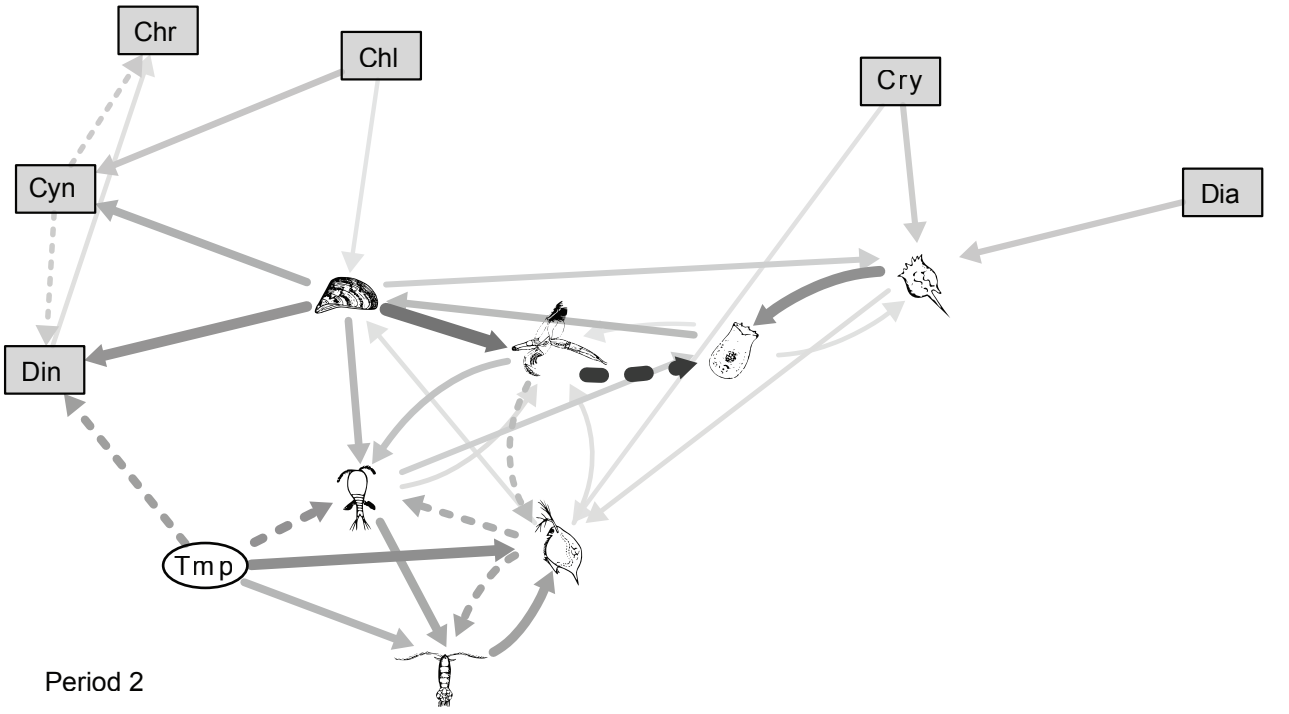
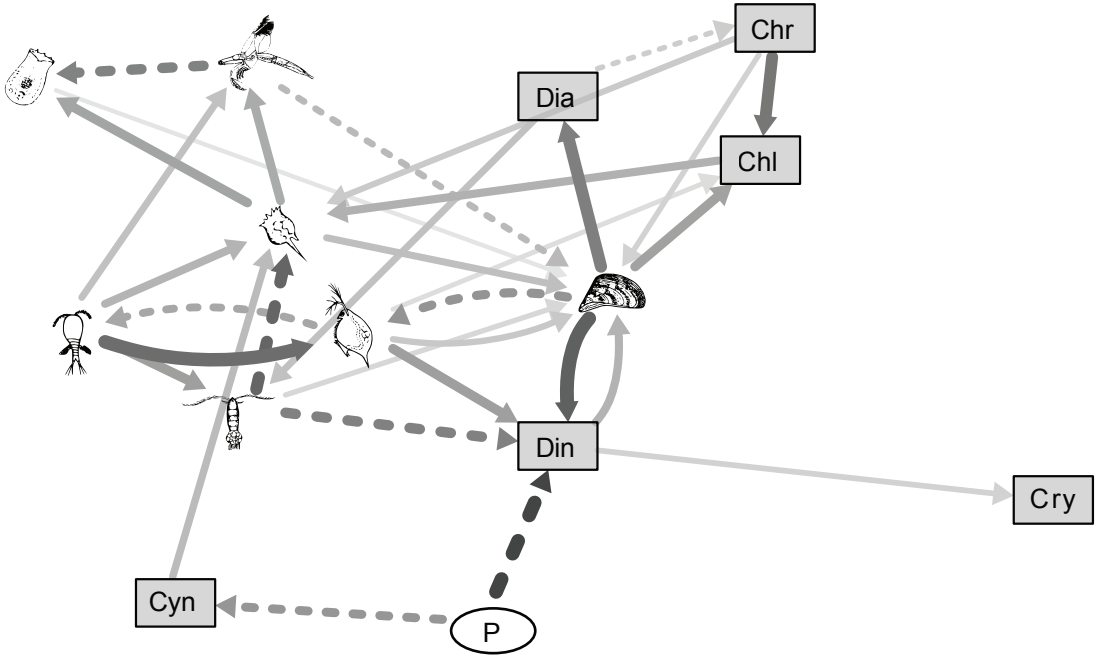
0.0
−0.5
P1 P2 P3
Bottom-up
Top-down
Competition
Density dependence
Abiotic effects
Seasonal effects



Period 1



Period 2



Period 3

Positive effect

Negative effect

Environmental covariate

Phytoplankton

Zooplankton herbivores

Rotifer

Copepod

Cladocera

Dreissena

Zooplankton predators

Rotifer

Copepod

Cladocera