RESEARCH ARTICLE

Declining calcium concentration drives shifts toward smaller and less nutritious zooplankton in northern lakes

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Abstract

Zooplankton community composition of northern lakes is changing due to the inter-active effects of climate change and recovery from acidification, yet limited data are available to assess these changes combined. Here, we built a database using archives of temperature, water chemistry and zooplankton data from 60 Scandinavian lakes that represent broad spatial and temporal gradients in key parameters: temperature, calcium (Ca), total phosphorus (TP), total organic carbon (TOC), and pH. Using machine learning techniques, we found that Ca was the most important determinant of the relative abundance of all zooplankton groups studied, while pH was second, and TOC third in importance. Further, we found that Ca is declining in almost all lakes, and we detected a critical Ca threshold in lake water of $1.3\,mg\,L^{-1}$, below which the relative abundance of zooplankton shifts toward dominance of *Holopedium gibberum* and small cladocerans at the expense of *Daphnia* and copepods. Our findings suggest that low Ca concentrations may shape zooplankton communities, and that current trajectories of Ca decline could promote widespread changes in pelagic food webs as zooplankton are important trophic links from phytoplankton to fish and different zooplankton species play different roles in this context.

KEYWORDS
calanoids, calcium, cladocerans, cyclopoids, *Daphnia*, *Holopedium*, lakes, phosphorus, temperature, zooplankton community composition

1 | INTRODUCTION

Successful efforts to combat acidification combined with climate change are causing some unforeseen changes in northern lakes. Lake pH is increasing due to reduced atmospheric sulfate deposition, which is accompanied by widespread declines in calcium (Ca) and total phosphorus (TP) concentrations and increased concentrations of colored total organic carbon (TOC, e.g., "browning") (Eimers et al., 2009; Finstad et al., 2016; Garmo et al., 2014; Isles et al., 2018, 2021; Jeziorski et al., 2008; Monteith et al., 2007; Weyhenmeyer et al., 2019). Adding to this, most lakes are also experiencing a pronounced warming (O'Reilly et al., 2015). Drivers for decreasing Ca in boreal regions include reduced acid rain, increased uptake by forests and timber harvesting and in some cases also dilution by increased rainfall (Finstad et al., 2016; Watmough et al., 2003; Zetterberg et al., 2016). In certain regions, there may also be increases in Ca owing to liming or increased forest root weathering (de Wit et al., 2023). Similarly, changes in TP and TOC are globally widespread and can be attributed to similar drivers as for Ca (de Wit et al., 2016; Huser et al., 2018; Isles et al., 2023; Monteith et al., 2007; Toming et al., 2020). Ca and P are widely recognized as potentially limiting elements for freshwater zooplankton, yet with species-specific responses, and thus changes in the concentrations of these elements will likely impact zooplankton community structure and production (Andersen & Hessen, 1991; Jeziorski et al., 2015). Still, our understanding of how the cumulative effects of the widespread changes in both Ca and TP concentrations in lakes affect the abundance and diversity of zooplankton and the food webs of lakes is incomplete (Palmer & Yan, 2013; Yan et al., 2008). Since zooplankton plays a key role in pelagic food webs (Jansson et al., 2007), improved understanding of the potential zooplankton community changes caused by Ca and TP declines is demanded. Significant changes in zooplankton community composition may impact transfer efficiency of energy and essential elements to fish, but also impact other processes such as nutrient recycling and grazing impacts on phytoplankton.

The ecological impacts of declining Ca on zooplankton community composition are widely recognized for North American temperate soft water lakes (Arnott et al., 2017; Jeziorski et al., 2015; Ross & Arnott, 2021), but hardly assessed for northern European lakes, although they may experience similar Ca declines (Weyhenmeyer et al., 2019). Calcium is crucial for the fitness of zooplankton with calcified exoskeletons and is mostly taken up directly from the lake water over respiratory surfaces (Hessen et al., 2000). For daphnids, it was found that direct uptake from water accounted for 97%–100% of Ca-acquisition (Tan & Wang, 2009). Low lake Ca concentrations (e.g., <1.5–$2\,\mathrm{mg\,L^{-1}}$; Jeziorski et al., 2015; Jeziorski & Yan, 2006) can therefore negatively, or even lethally, affect zooplankton taxa with high Ca demands if the Ca levels fall below critical thresholds needed for growth, reproduction, and survival (Hessen et al., 2000; Jeziorski et al., 2015; Jeziorski & Yan, 2006; Wærvågen et al., 2002). Suboptimal Ca levels lead to reduced calcification and fitness (Hessen et al., 2000), and can cause shifts in size distribution of especially cladocerans, typically from large sized

Daphnia toward smaller individuals, thereby lowering zooplankton biomass and food resources for fish (Hessen et al., 2000; Jeziorski & Yan, 2006). Declining Ca, and lower pH, can also promote extensive lake "jellification", that is, a replacement of high Ca demanding zooplankton like *Daphnia* by the jelly coated and less Ca demanding *Holopedium* (Hessen et al., 1995a; Jeziorski et al., 2015), where lake pH now seems to have a secondary role in this replacement compared to declining Ca (Couture et al., 2021; Ross & Arnott, 2021). Recent studies have also demonstrated that *Daphnia* spp. can diverge in their tolerance to low Ca (Ashforth & Yan, 2008; Azan & Arnott, 2018; Ross & Arnott, 2021; Tan & Wang, 2010). But few studies have addressed the possible impacts of low or declining lake Ca concentrations on copepods. Previous lake studies have inferred low Ca sensitivity among copepods based on their spatial distribution and relatively low body Ca content (Wærvågen et al., 2002). Yet, recent experimental studies suggest that the growth of both copepods and cladocerans (e.g., *Daphnia* spp.) can be impacted by Ca (Azan & Arnott, 2018; Ross & Arnott, 2021; Tan & Wang, 2010), suggesting that other factors than bodily Ca content are relevant for species-specific Ca demands (Azan & Arnott, 2018). For instance, growth and thus molting rate constitute a drain of Ca bound in the carapace, and thus are also relevant for the demands (Tan & Wang, 2010). However, while spatial data and experimental studies suggest zooplankton community effect of Ca concentrations, temporal impact on zooplankton community composition in response to gradual changes in key elements such as Ca remains largely unknown.

In parallel to declining Ca, temporal changes in TP may interact with Ca as determinants of zooplankton community composition (Hessen et al., 1995b; Prater et al., 2016). Unlike Ca, P is obtained primarily via food, for example, by grazing on phytoplankton (Hessen, 1992). Downward trends in lake TP concentrations can therefore affect not only phytoplankton food quantity (Hessen et al., 2003; Isles et al., 2021; Yan et al., 2008) but also stoichiometric quality (Bergström et al., 2022; Hessen, 1992; Hessen et al., 2003). Since zooplankton species vary in their demand for ribosomal P needed for growth (e.g., high P demand in large cladocerans especially *Daphnia* and in cyclopoids copepods; lower P demand in small cladocerans, *Holopedium gibberum* and calanoid copepods; Andersen & Hessen, 1991; Bergström et al., 2022; Hessen et al., 2013; Jeziorski et al., 2015), nutrient depletion may affect zooplankton community composition (Bergström et al., 2022).

Lake TP concentrations, and the dietary P obtained by zooplankton, are however indirectly impacted by TOC concentrations, since TOC impacts light and nutrient availability, and phytoplankton biomass depending on the TOC:TP stoichiometry (Isles et al., 2021). Increased TOC concentrations may also favor unselective filtering feeding cladocerans capable of feeding directly on detritus and bacteria over copepods that selectively feed primarily on phytoplankton (Berggren et al., 2015; Bergström et al., 2021; Hessen, 1985), although both bacteria and detritus are low quality food that do not sustain rapid growth or reproduction of *Daphnia* (Wenzel et al., 2021). Increased TOC further induces additional warming of the epilimnion (Puts et al., 2023), where higher water temperatures

combined with reduced phytoplankton biomass may enhance the susceptibility of *Daphnia* to low Ca (Ashforth & Yan, 2008). Thus, the impact of temporal changes in water quality on zooplankton community composition is also related to climate (Ashforth & Yan, 2008; Couture et al., 2021).

While there may be other limiting elements, we here focus on Ca and TP since their widespread importance is well documented, their physiological impact is mechanistically well understood (Hessen et al., 2013), and because these elements are shown to vary substantially in space and time (Isles et al., 2023; Weyhenmeyer et al., 2019). The cumulative effects of reduction of these essential elements (Ca and P) and warming of northern lakes potentially promote smaller zooplankton taxa, negatively affecting grazing rates and energy and essential element transfer to fish. Spatial analysis of lakes suggests that zooplankton community is affected by both Ca and TP (Palmer & Yan, 2013; Wærvågen et al., 2002), but a temporal analysis of the zooplankton community across broad climate and chemical gradients has not yet been performed.

In this study, we compiled temporal data of air temperature, water chemistry (Ca, TP, pH, and TOC), and zooplankton from 60 Scandinavian lakes that represent broad climate and chemical gradients. We (1) analyzed if air temperature and lake water chemistry (Ca, TP, pH, and TOC) were correlated with the relative abundance of five major zooplankton groups (cyclopoids, calanoids, small cladocerans, *Daphnia* spp., and *Holopedium gibberum*), (2) used machine learning to identify critical thresholds in air temperature and lake water chemistry above/below of which shifts in zooplankton assemblages occur, and (3) conducted trend analysis to determine whether the lakes were moving toward or away from these critical thresholds.

We predicted the following:

1. Lake Ca concentration is the main determinant of the spatial distribution in zooplankton community composition.
2. Below a critical lake water Ca threshold of $1.5 \, mg \, L^{-1}$, zooplankton community composition shift from dominance of larger species of high Ca demand (large *Daphnia* spp., and copepods) toward species of smaller body size and/or low Ca demand (small cladocerans and *Holopedium gibberum*).
3. Both lake Ca and TP concentrations are declining to reach or fall below critical thresholds where major shifts in zooplankton community composition may occur.

2 | MATERIALS AND METHODS

2.1 | Air temperature, water chemistry, and zooplankton data

We gathered long-term data on air temperature, water chemistry and on zooplankton for lakes in Norway and Sweden (see lake coordinates in Table S1). The lake water chemistry and zooplankton data were obtained from ongoing Norwegian and Swedish lake monitoring programs. The data from the Norwegian lake monitoring program includes mainland lakes sampled annually from 1986 (Schartau et al., 2020; Figure 1; Table S1). The Swedish lake monitoring program includes lakes sampled from 1991, where the zooplankton monitoring has mainly been focused on the southern parts of Sweden (Figure 1; Table S1). Some of the Swedish lakes ($n = 18$) are limed (Table S1). In total, this yielded annual zooplankton data from 60 lakes with a common sampling window from 1991 to 2020. Mean (median) lake surface area of the Norwegian and Swedish lakes are 0.71 (0.29) km^2 and 0.64 (0.29) km^2, respectively (Table S1). Hence, these monitoring lakes are typically small and shallow (Table S1) with median concentrations (and Lower Quartile Q1 and Upper Quartile Q3) of: Ca = 3.2 [Q1 = 0.9, Q3 = 6.5] mg L^{-1}, TP = 5.7 [Q1 = 3.6, Q3 = 7.8] μg L^{-1}, pH = 6.4 [Q1 = 5.8, Q3 = 7.0], and TOC = 8.7 [Q1 = 3.4, Q3 = 12.4] mg L^{-1}, respectively (Table S2).

Lake water chemistry from the surface water (0.2–1 m depth) were gathered over the open water season (June to early October) and were sampled on 3–4 occasions yearly. In Norway, lake water chemistry analysis was performed at the accredited laboratory at Norwegian Institute for Water Research (NIVA). For detailed description of the analytical procedures for the Norwegian lakes, see Hindar et al. (2020). In Sweden, lake water chemistry was analyzed at the accredited laboratory at the Department of Aquatic Sciences and Assessment, Swedish University of Agricultural Sciences. The analytical procedures for the Swedish lakes water chemistry followed international (ISO) or European (EN) standards (SS-EN ISO 5667-1:2007). From these data, we estimated mean water chemistry for the open water season for each year and each chemical parameter (Ca, TP, pH, and TOC). For pH, we transformed values to H^+ concentrations before estimating mean values and transformed them back to pH for the figure illustrations. Total nitrogen (TN) was not included since TN in many boreal lakes is highly related with TOC due to large parts of TN is organic N (i.e., humic substances; see Bergström, 2010). We instead use lake TP as an indicator of lake trophic status.

Zooplankton samples were taken on 2–3 occasions between June to late September, that is, on fewer occasions compared to the frequency of the water chemistry sampling but taken on the same dates as water chemistry. In Norway, zooplankton was collected by hauling the water column using a plankton net (mesh size 90 μm) according to Norwegian standard (NS-EN-15110). Samples were preserved with alkaline Lugol's solution. Organisms were identified and counted to the finest possible taxonomic level by microscopy by taxonomists at the accredited laboratory at NIVA (Schartau et al., 2020). In Sweden, zooplankton was collected with a 4.2 L Limnos water sampler at one centrally located station per lake, with sampling at 2-m depth intervals. The collected water was mixed in a bucket, separately for epilimnetic and hypolimnetic samples, and animals were collected on a 40 μm net and preserved with alkaline Lugol's iodine solution. Pelagic zooplankton was estimated as the sum of individuals in the epilimnion and hypolimnion.
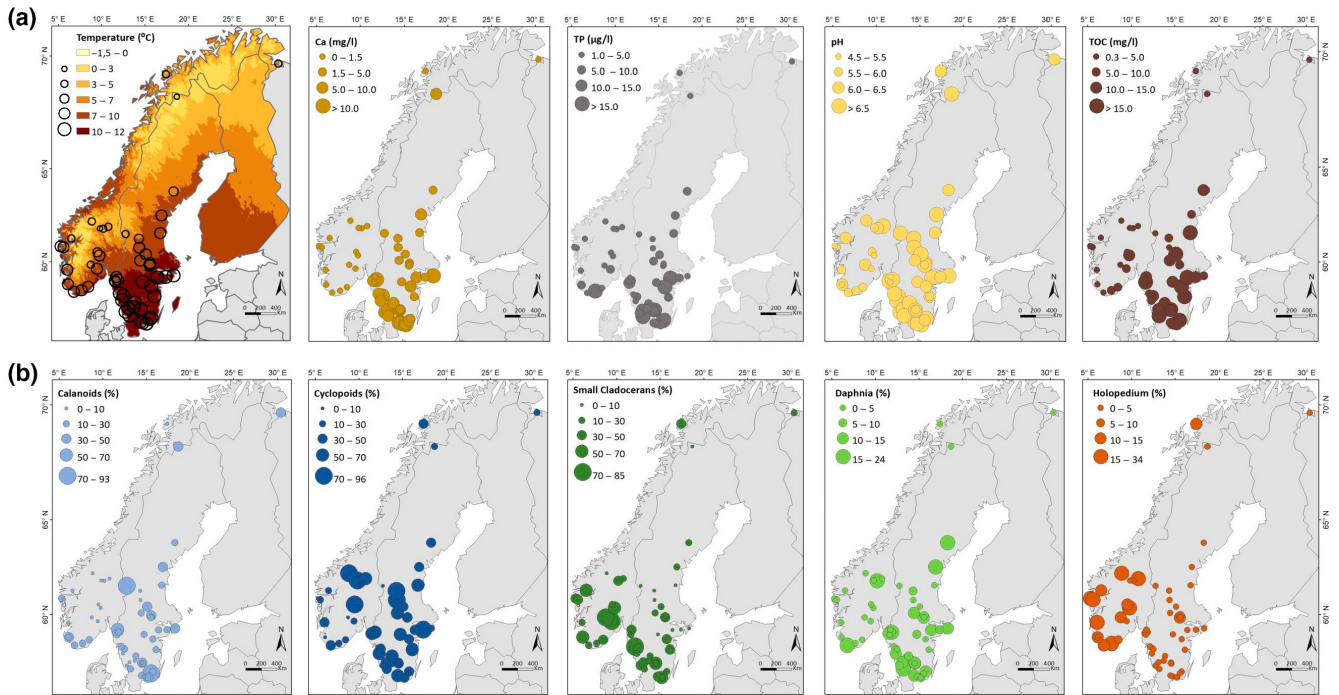
FIGURE 1 (a) Annual mean maximum air temperature and spatial distribution of water chemistry parameters (mean concentrations), and (b) spatial distribution of five zooplankton groups (mean relative abundance in %) in 60 study lakes in Fennoscandia for the 2006–2010 period.

Organisms were identified and counted to the finest possible taxonomic level by microscopy by taxonomists at the accredited laboratory at the Department of Aquatic Sciences and assessment, Swedish University of Agricultural Sciences.

For zooplankton, we used individual counts of each pelagic zooplankton taxa, and grouped them into groups of calanoids and cyclopoids (adults plus copepodites), small cladocerans (dominated by *Bosmina* and *Eubosmina* but also small numbers [≤4%] of *Ceriodaphnia*, *Chydorus*, *Diaphanasoma*, and *Alona*), large cladocerans (mainly *Daphnia* [98%] but also small numbers of *Sida* and *Limnosida* [≤2%]), and *Holopedium gibberum* (i.e., in total five groups of zooplankton). Predatory zooplankton was not included. Calanoids and cyclopoids were grouped separately since they in recent experimental studies have been shown to have high Ca demand (Azan & Arnott, 2018). For cladocerans, the division was based on the knowledge that small cladocerans (*Bosmina*, *Eubosmina*, *Ceriodaphnia*, *Chydorus*, *Diaphanasoma*, and *Alona*) and *Holopedium gibberum* have low Ca demand and are less sensitive to critical low lake water Ca concentrations, compared to large sized cladocerans (e.g., *Daphnia*) with typical high Ca demand required for moulting (Azan & Arnott, 2018; Couture et al., 2021; Jeziorski et al., 2015; Jeziorski & Yan, 2006). We further grouped all *Daphnia* species to one group (i.e., as spp.), although it is recognized that Ca demand among different species of *Daphnia* can vary (e.g., *D. catawaba and D. ambigua* have been shown in experimental studies to have low Ca demand and thrive at low Ca; Ross & Arnott, 2021). However, these low Ca demanding species of *Daphnia* are not present in Scandinavian lakes. In our dataset, 88% of the *Daphnia* counts in Norway was *Daphnia longispina* and 70% of

the *Daphnia* counts in Sweden was *Daphnia cristata*, both relatively large sized species of *Daphnia*. Moreover, 21% of the *Daphnia* counts in Sweden was not identified to species (i.e., set to *Daphnia* spp.). Based on this knowledge on the species distribution and counting precision, we therefore grouped all *Daphnia* species together into one single group. We then summed up the individual counts over summer for all zooplankton, and for each group, and then estimated their relative abundance (in %) of each group of total zooplankton for each year.

Water temperature data were not available from the Norwegian lakes. Point measurements of lake water temperature may, however, not be representative for temperature over season and at different depths. To have a full dataset, we instead used mean air temperature as a proxy for lake water temperature (Livingstone & Lotter, 1998; McCombie, 1959) like other studies (see Couture et al., 2021; Hessen et al., 2007). Mean annual maximum air temperature (hereafter: temperature) was determined for each lake over the 2000–2019 period. Annual temperatures were chosen from June to September temperatures to account for all year-around temperature gradients and different reproductive cycles of zooplankton (e.g., hatching). Maximum instead of mean annual temperature was used, as maximum temperature is more likely to affect organismal performance and survival (Buckley & Huey, 2016; Kingsolver & Buckley, 2017), and has been shown to be a better predictor than mean temperature for spatial distribution of plankton in northern lakes (Hallstan et al., 2013). Temperatures were extracted from the "TerraClimate dataset" (high-spatial resolution; 1/24°, ~4km) monthly air temperature grids (see Abatzoglou et al., 2018).

2.2 | Relationships between environmental factors and zooplankton—spatial analyses

Since not all study lakes were sampled in all years and to maximize the number of years with overlapping data, we selected a 5-year period (2006–2010), which had overlapping data for all 60 lakes which resulted in 291 common sampling points. This common 5-year period for our 60 study lakes were used for spatial analyses, for example, for correlation and critical threshold analyses and tests of prediction 1 and 2.

For the spatial analyses, we used the Spearman's correlation test to determine relationships among chemical parameters (Ca, TP, TOC, and pH), temperature and all zooplankton groups (small cladocerans, *Daphnia* spp., *Holopedium gibberum*, cyclopoids, and calanoids) for the 60 study lakes. This was performed on mean data for each lake from the selected 5-year period (2006–2010). We also performed a redundancy analysis (RDA) using the same data as for the Spearman's correlation to evaluate the direction of change (positive vs. negative) in the zooplankton groups across the gradients in water chemistry and temperature (prediction 1).

To identify critical thresholds of water chemistry parameters and temperature, across which changes in the relative abundance of zooplankton species occurred, we used machine learning and performed regression tree and random forest analyses. The analyses were performed on mean data for the common five-year period (2006–2010) for the 60 lakes. Since there was strong collinearity between Ca and pH, and between TP and TOC (i.e., these parameters had large Spearman's rho, see Figure 2) that could potentially result in overperformance of the regression trees and a decrease in

the accuracy of random forest models (Strobl et al., 2009; Tolosi & Lengauer, 2011), we performed two separate models (runs). The first model included Ca, TP, and temperature, while the second model included pH, TOC, and temperature (prediction 1–2).

A critical threshold of a variable occurs when the variable value falls below the threshold needed for bodily demands, which promotes shifts in zooplankton community composition toward species with lower demands. A critical threshold could apply to lake water Ca concentration taken up by zooplankton directly from lake water (Hessen et al., 2000), or to lake water TP concentration taken up by zooplankton indirectly from food (Prater et al., 2016; Yan et al., 2008). Critical thresholds can also apply to lake water TOC indirectly since TOC concentrations impact food quantity (phytoplankton biomass; Isles et al., 2021) and quality (Hessen et al., 2003, 2013). Lake water pH impacts zooplankton directly, where different species are known to be of different tolerance to acid- and non-acid conditions (Hessen et al., 1995a; Yan et al., 2008). Therefore, critical thresholds also apply for lake water pH. Temperature directly affects zooplankton metabolic rates and has been shown to influence zooplankton community composition together with water quality parameters (Couture et al., 2021). A critical threshold for temperature is not strictly applicable, but we defined temperature thresholds as the turning point where shifts from negative to positive relative abundance was observed for the major taxa.

We used temperature, Ca, and TP as explanatory variables in run one (model one), and temperature, pH, and TOC as explanatory variables in run two (model 2), while zooplankton groups (cyclopoids, calanoids, *Holopedium gibberum*, *Daphnia* spp, and small cladocerans) were used as a response variable in each run. The regression trees
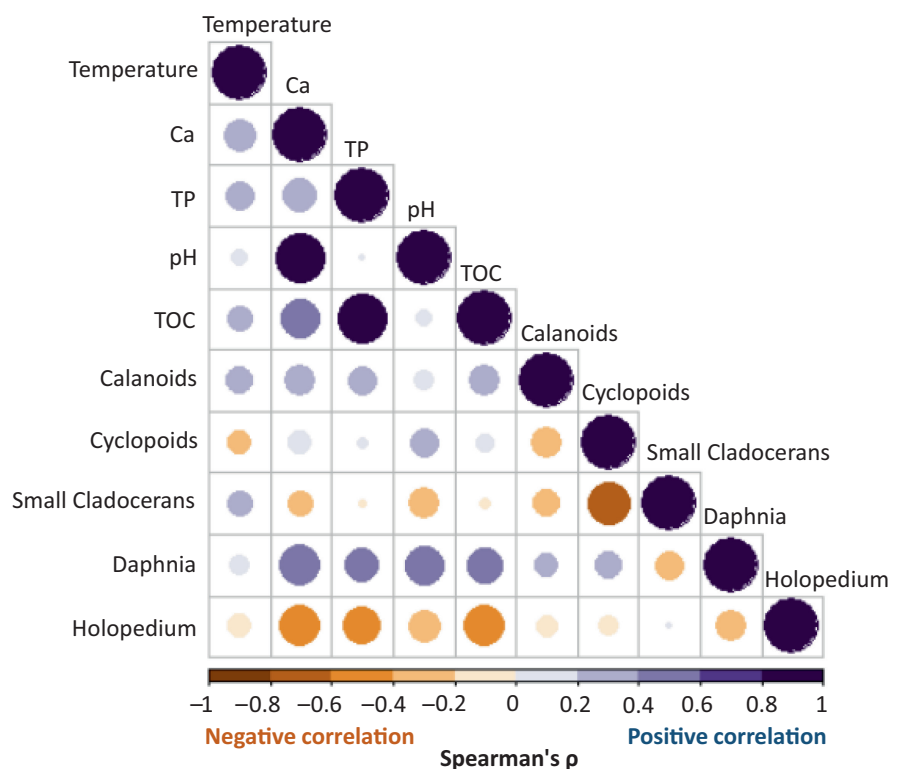

FIGURE 2 Spearman correlation matrix performed on temperature, water chemistry parameters (mean concentrations) and zooplankton (mean relative abundance in %) for the 2006–2010 period for each lake (number of lakes = 60).

were pruned at the branch where the cross-validation error was minimized by the complexity parameter (De'ath & Fabricius, 2000). We considered a value at the first regression tree node (i.e., a point where the tree splits into two branches for the first time) as a critical threshold. We then aggregated the results of all regression trees on a single plot showing the critical thresholds at the first regression node for each zooplankton group and explanatory variable (prediction 1–2).

To assess the stability of our regression tree models, we then performed the random forest analysis. In the random forests, 10,000 regression trees (instead of a single tree) were produced, and then results were aggregated into a single random forest model and presented as the "variable importance plot". "Importance" was measured by the percent increase in mean squared error (*%IncMSE*); the higher the value of the *%IncMSE* score, the higher the importance of the variable in the model (Strobl et al., 2009). Yet, to reassure the order of importance for each parameter, we also performed a random forest model that included all parameters regardless of the collinearity (i.e., "combined" model). We then compared *%IncMSE* scores of this "combined" model with the scores of the separate models (prediction 1–2).

2.3 | Trends in chemistry, climate, and zooplankton

We also selected 20 lakes that had the longest overlapping time series—20 years (from 2000 to 2019) which resulted in 340 common sampling points. These 18 lakes, of which three were limed in the past (Table S1), were used for trend analyses and testing of prediction 3. The nonparametric Mann–Kendall test (Kendall, 1975) was performed on temperature, chemical parameters, and zooplankton groups, and Kendall statistics (Kendall *tau* and *p* values) were analyzed to detect trends over the 20-year period. We also calculated Sen's slopes (1968; unit/year) to estimate the rates of change for temperature, chemical parameters, and zooplankton groups in each lake.

Since our spatial analyses reveal that Ca was the most or second most important determinant for the spatial distribution and relative abundance of all zooplankton groups (i.e., 2.2 and Results), and since Ca concentrations were declining in almost all lakes (i.e., 2.3 and Results), we assessed (i.e., prediction 3) if lakes were moving toward or away from critical lake water Ca thresholds that could promote shift in zooplankton community composition.

Thus, we averaged Ca concentrations over the 20-year period (2000–2019) in each of the 18 lakes. We then divided the lakes into two bins according to the most common thresholds found for Ca concentrations in zooplankton (i.e., 1.3 mg L^{-1}; see Figure 3). The first bin included lakes with Ca concentrations between 0.3 and 1.3 mg L^{-1} (i.e., from the minimum to the value equal to the most common Ca threshold found for zooplankton in our study). The second bin included lakes from 1.4 mg L^{-1} to the maximum Ca concentration (5.4 mg L^{-1}). We then selected the corresponding relative abundance of each zooplankton group (%) for each lake and each year, and then

calculated mean Ca concentrations and mean relative zooplankton abundance in each of the two bins. The nonparametric Mann–Kendall test was then performed on these Ca and zooplankton means from the two bins to determine if the Ca and zooplankton have significant ($p < .1$) monotonic trends over the 20-year period. Finally, to determine whether a relationship existed between the time series of Ca and zooplankton, we performed the cross-correlation test on the Ca and zooplankton means from each bin using the cross-correlation function (CCF; Derrick & Joshua, 2004) (for prediction 3).

2.4 | Statistical analyses

The statistical analyses were performed in R (v. 4.2.2; R Core Team, 2022). The correlation, regression tree and random forest analyses were performed using R packages *hmisc*, *rpart*, and *randomForest*. The RDA analysis was performed with the *vegan* and *factoextra* packages, while the Mann–Kendall tests and trend analyses were performed with the *trend* package. For the correlation and trend analyses, the significance level was set to $p \le .1$. We choose 0.1 instead of 0.05 because a $p < .1$ is a reasonable probability of error to accept given the source (two different monitoring programs) and type (ecological time series) of data, as well as the type of analysis performed (time series analysis; Wasserstein & Lazar, 2016; Wasserstein et al., 2019, but also Oliver et al., 2018; Paltsev & Creed, 2022). The use of $p < .1$ allowed the inclusion of more lakes in the averaging, which helped reduce the potential confounding influence of other lake-specific factors on the regression relationship, which outweighed the downside of accepting a less significant trend. Further, the conclusions are not drawn on individual lakes (where high *p*-values would be more problematic). All *p*-values are presented in Tables S4–S6, where 16%, 25%, 14%, and 25% of the *p*-values used, respectively, were between 0.1 and 0.05, the rest below 0.05.

3 | RESULTS

3.1 | Environmental factors and zooplankton community structure

Data from the 60 Scandinavian lakes revealed a longitudinal (west to east) increase in mean temperature and mean concentrations of Ca, TP, pH, and TOC for the common five-year period from 2006 to 2010 (Figure 1a). The composition of the zooplankton community varied across these temperatures and chemical gradients (Figure 1b). A Spearman correlation analysis revealed that temperature showed weak relationships to water chemistry (Ca, TP, pH, TOC) and to zooplankton (Figure 2). However, strong positive and negative relationships existed between water chemistry and the relative abundance of zooplankton (Figure 2; Figure S1, Tables S3 and S4). The relative abundance of *Daphnia* spp. was positively correlated to Ca, TP, pH, TOC, while for *Holopedium gibberum* (hereafter: *Holopedium*) this
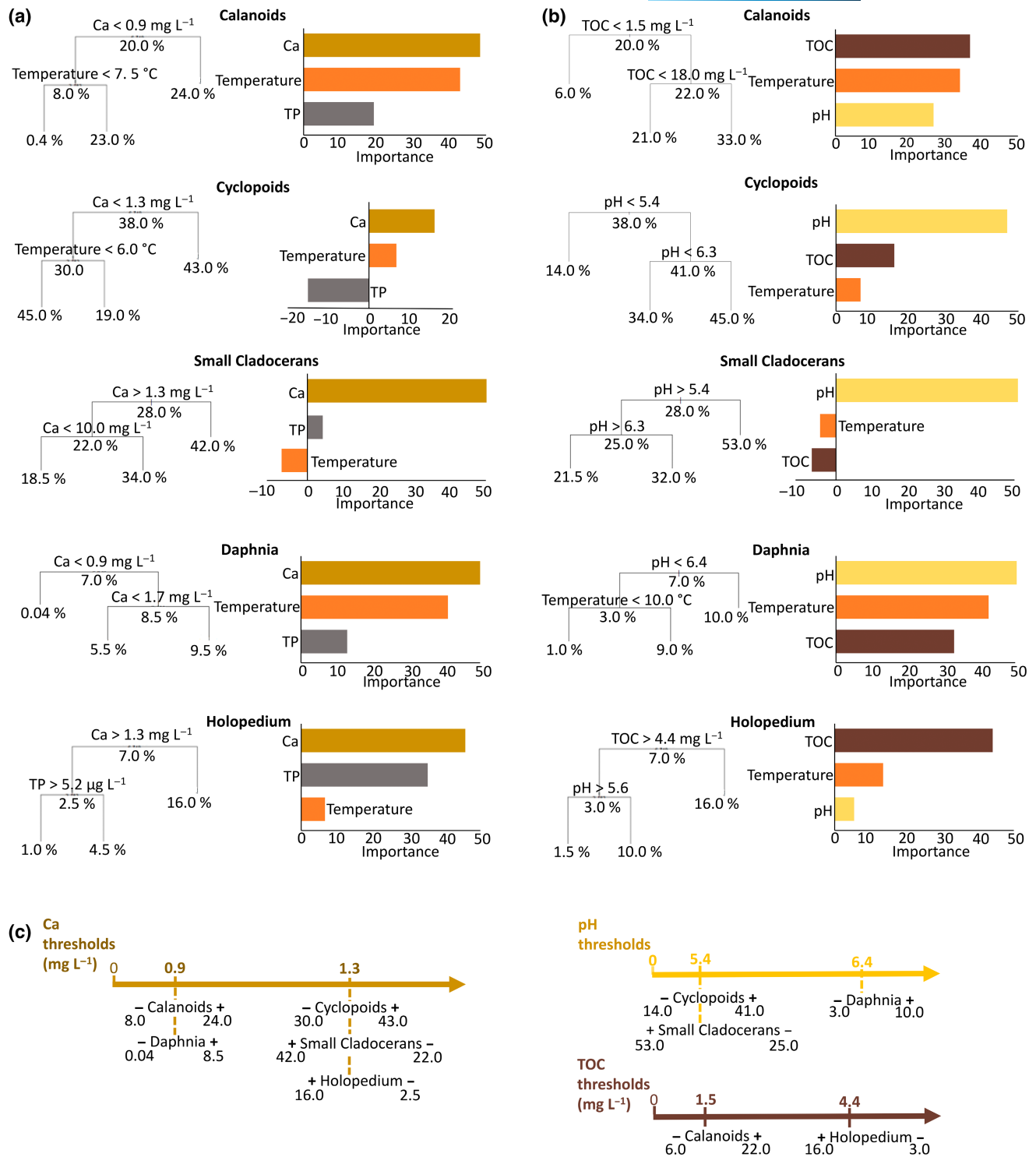
FIGURE 3 Results of the regression tree and random forest ("importance plots" on the right panel) analyses for (a) Ca, TP, and temperature, and (b) pH, TOC, and temperature performed on 60 study lakes. "Importance" was measured by the percent increase in mean squared error (*%IncMSE*); the higher the value of %IncMSE score, the higher the importance of the variable in the model; (c) Critical thresholds in Ca, pH and TOC identified for each zooplankton group using the regression tree analysis. "+" and "−" indicate if relative zooplankton abundance increases ("+") or decreases ("−") below and above the threshold in Ca, pH, and TOC; numbers below the "+" and "−" indicate mean abundance (in %) of a zooplankton group below and above the threshold.

was negatively related to these variables. For calanoids, the relative abundance was positively (but weaker than *Daphnia* and *Holopedium*) correlated to Ca, TP, and TOC, and temperature, whereas relative

abundance of cyclopoids was weakly positively and negatively correlated to pH and temperature, respectively. The relative abundance of small cladocerans showed overall weak relationships with water

chemistry (Figure 2). Our RDA analysis supports these directions (positive vs. negative) of the relationships between water chemistry parameters, temperature, and all zooplankton groups (see Figure S1).

3.2 | Critical thresholds in environmental factors based on spatial analysis

Since there was strong collinearity between Ca and pH, and between TP and TOC (see Figure 2), we ran two separate regression tree analyses followed by two separate random forest analyses for each pair (Ca, TP, and temperature vs. pH, TOC, and temperature) to determine if critical thresholds existed beyond which significant changes in the relative abundance of zooplankton species occurred (see Section 2).

The first regression tree and random forest analyses with Ca, TP, and temperature (Figure 3a) revealed that Ca was the most important determinant for the relative abundance of *each* zooplankton group assessed (e.g., situated at the first regression node and with highest importance in the random forest analyses), while temperatures and TP were second and third important determinants, and then only for selected zooplankton groups (Figure 3a). *Daphnia* spp. (i.e., large cladocerans) and calanoids had the lowest estimated critical threshold for Ca ($0.9\,mg\,L^{-1}$), whereas cyclopoids, small cladocerans, and *Holopedium* all gathered around the Ca critical threshold of $1.3\,mg\,L^{-1}$. Thus, the spatial analysis revealed that Ca levels below the respective critical thresholds of 0.9 and $1.3\,mg\,L^{-1}$ are causing shifts in zooplankton communities from species that have high Ca demands such as *Daphnia* spp. (decreased from 7% to <0.1%) toward species that have lower Ca demands such as *Holopedium* (increased from 7% to 16%) and small cladocerans (increased from 28% to 42%; Figure 3c). Cyclopoid and calanoid copepods declined in relative abundance below their Ca critical thresholds of 1.3 and $0.9\,mg\,L^{-1}$ from 43% to 30% and from 24% to 8%, respectively (Figure 3c).

For the second regression tree and random forest analyses with pH, TOC, and temperature (Figure 3b), the importance varied among the determinants, with pH found to be the most important determinant for the relative abundance of cyclopoids, small cladocerans, and *Daphnia* spp., whereas TOC was the most important determinant for the relative abundance of *Holopedium* and calanoids (Figure 3b). Identified thresholds in pH were 5.4 for cyclopoids and small cladocerans, with an increase in relative abundance of small cladocerans below, and an increase in relative abundance of cyclopoids above pH of 5.4 (Figure 3c). The pH threshold for the relative abundance of *Daphnia* spp. was 6.4 (decreasing in abundance below and increasing in abundance above; Figure 3c). The identified TOC threshold for the relative abundance of calanoids was $1.5\,mg\,L^{-1}$ (declining below and increasing above the threshold), whereas for *Holopedium* the TOC threshold was at a higher concentration ($4.4\,mg\,L^{-1}$) with increasing relative abundances below and decreasing relative abundances above this threshold (Figure 3c).

To reassure the order of importance of the determinants assessed separately in the regression tree and random forest analysis

(Figure 3a,b), we re-ran the random forest with all determinants included in one model (see Figure S2). Again, Ca was the most important determinant for the relative abundance of calanoids, *Daphnia* spp. and small cladocerans, and came out second in order of importance as a determinant for *Holopedium* slightly after TOC. Ca was also the second most important determinant for the relative abundance of cyclopoids after pH (Figure S2), but Ca showed much lower importance for the spatial distribution than did pH. Hence, according to our analyses Ca concentrations are clearly important for the spatial distribution of all zooplankton groups (identified first or second in order of importance), although this distribution may be modified spatially by variation in pH and TOC (Figure 3). This order of importance of the parameters assessed for spatial distribution in relative abundance for zooplankton was also identified in the RDA (Figure S1), although this analysis cannot be used for identifying thresholds (see Section 2).

3.3 | Temporal Ca-driven shifts in zooplankton community structure

Trends in temperature, Ca, TP, pH, TOC, and the relative abundance of the zooplankton groups were assessed in 18 of the 60 Scandinavian lakes for a 20-year period (2000–2019) using Mann–Kendall test. During this time, these lakes showed trends of increasing temperatures and in TOC, and generally negative trends in Ca, and either negative or positive trends in TP and pH (Figure 4). During
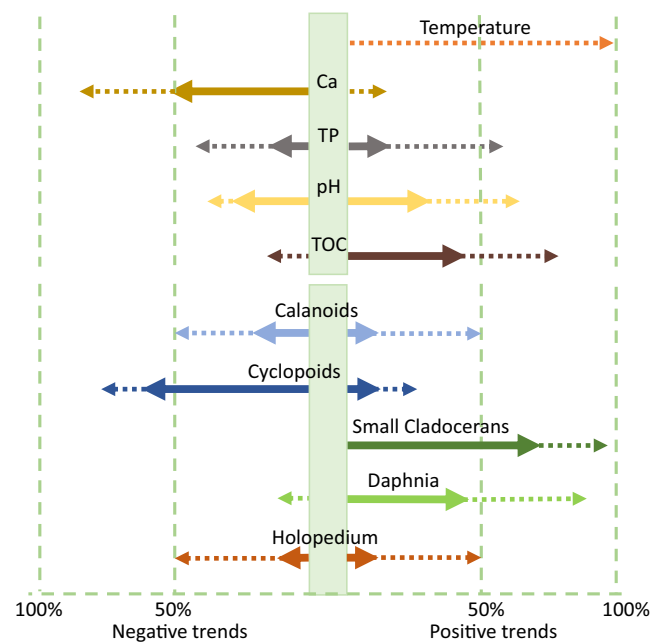

FIGURE 4 Monotonic trends in temperature, water chemistry parameters and zooplankton in 18 study lakes shown as Kendall *tau*. The lengths of dotted arrows represent the proportion of lakes with positive or negative *tau*, and the length of solid arrows represents the proportion of lakes with significant trends at level $p < .1$.

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the same time, zooplankton in these lakes generally showed negative trends in the relative abundance of cyclopoids, positive trends in relative abundances of small cladocerans and in *Daphnia* spp., and mixed trends (negative and positive) in relative abundance of calanoids and *Holopedium* (Figure 4; Tables S5 and S6).

Since Ca was identified as the most (Figure 3a), or first to second (Figure S2), important determinant for the relative abundance of all zooplankton groups assessed, and since Ca thresholds were identified for all of these zooplankton groups (Figure 3c), we assessed the relative abundances of the zooplankton groups in lakes above and below the common critical Ca threshold of 1.3 mg L⁻¹ (see Section 2), and then we assessed if the Ca declines for the 20-year period in these lakes (Figure 4) were associated with changes in the relative abundance of the zooplankton groups using cross-correlation tests between time series (Figure 5; Figure S3, Tables S5–S7).

We found that the relative abundance of the different zooplankton groups varied above and below the identified common Ca critical threshold (1.3 mg L⁻¹) for the 20-year period (Figure 5). *Above* the Ca critical threshold, copepods had the largest relative

abundance (20%–37% for calanoids; 31.3%–61.4% for cyclopoids), followed by cladocerans (4%–26.6% for small cladocerans; 3.8%–15% for *Daphnia*), and *Holopedium* (0.7%–5.4%). *Below* the Ca critical threshold, the relative abundance of the different zooplankton groups changed, ranging between 14%–31% for calanoids, 6%–31% for cyclopoids, 26.7%–67.3% for small cladocerans, 0.4%–9.3% for *Daphnia* and 1.8%–19.2% for *Holopedium* (Figure 5).

Moreover, we found that the Ca declines over this 20-year period were associated with major trends and shifts in the relative abundance of zooplankton groups (Figure 5; Tables S6 and S7). *Above* the critical Ca threshold, Ca declines were associated with declines in relative abundances of calanoid and cyclopoid copepods but increases in relative abundances of small and large cladocerans (*Daphnia*) and *Holopedium*. Most trends were significant (at level of $p < .1$), except that for calanoids. *Below* the critical Ca threshold, the relative abundance of calanoids was stable, while the relative abundance of cyclopoids continued to decline with declining Ca but not significantly ($p > .1$). The relative abundance of small cladocerans continued to rise ($p < .1$), whereas *Daphnia* dropped to very
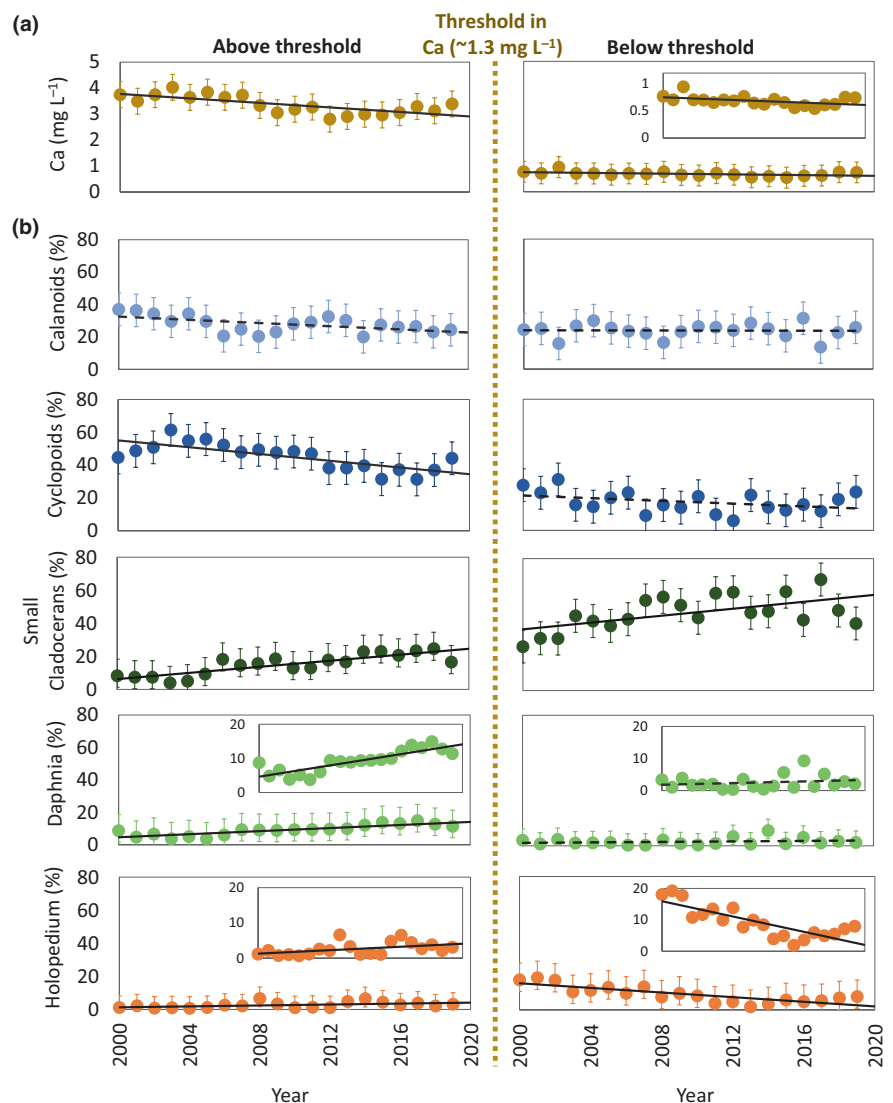

FIGURE 5 Lakes below and above the common critical threshold in Ca showing trends in (a) Ca, and (b) zooplankton toward or away from the common critical Ca threshold for the period of 20 years (2000–2019). Orange dotted line represents the common Ca critical threshold. Black lines represent significant ($p < .1$) trends (an increase or decrease in relative zooplankton abundance), while black dotted lines represent a non-significant increase or decrease.

low relative abundances below the Ca threshold and remained low in relative abundance over the 20-year period. *Holopedium* showed significant declining trends ($p < .1$) with declining Ca below the Ca threshold.

4 | DISCUSSION

Lake Ca concentration was the most evident and important variable in determining zooplankton community composition spatially in this set of northern lakes. To our knowledge, this is the first study illustrating that lake Ca concentrations are indeed most strongly associated with differences in the relative abundance of *all* the zooplankton groups that we assessed. We identified lake water Ca thresholds of 0.9–1.3 mg L^{-1}, below which shifts occurred in the relative abundance of zooplankton toward dominance of *Holopedium gibberum* and small cladocerans at the expense of *Daphnia* spp. and copepods. We also found that Ca is declining in almost all lakes and that the current trajectories of Ca declines suggest community shifts toward dominance of smaller and less nutritious zooplankton.

4.1 | Critical thresholds shaping zooplankton community composition

The critical Ca thresholds for cladocerans (0.9–1.3 mg L^{-1}) did not align completely with previously reported demands based on their Ca content for these zooplankton species (i.e., lower demand for small than large cladocerans [*Daphnia*]; Wærvågen et al., 2002). Yet, as Ca concentration shifted from above to below the critical threshold, the relative abundance of large *Daphnia* decreased while small cladocerans and *Holopedium* increased. These findings are supported by previous studies reporting that lake water Ca concentration is critical in determining the relative distribution and shifts in abundance of different cladoceran species (Jeziorski et al., 2015; Jeziorski & Yan, 2006; Wærvågen et al., 2002). The critical Ca thresholds for copepods were like those for cladocerans (calanoids 0.9 mg L^{-1}, cyclopoids 1.3 mg L^{-1}). Despite anticipated low demands inferred from their Ca contents (Wærvågen et al., 2002), the relative abundance of cyclopoid and calanoid copepods declined below their critical Ca thresholds. Our results therefore disagree with previous lake studies, which suggested that copepod distributions are not influenced by Ca concentrations (Wærvågen et al., 2002). Recent in situ lake experiments also confirm that the actual growth rates of many zooplankton species, including that of copepods, are reduced at low Ca (Azan & Arnott, 2018), and that body Ca content, especially in copepods, may be a poor proxy for the species-specific Ca demand (Azan & Arnott, 2018). Thus, our common Ca threshold of 1.3 mg L^{-1} aligns well with previous recognized Ca thresholds for large cladocerans e.g., *Daphnia* (<1.5 mg L^{-1}; Ashforth & Yan, 2008; Azan & Arnott, 2018; Jeziorski et al., 2015; Jeziorski & Yan, 2006), but importantly that this Ca threshold also seems to be valid for copepods. With the relative abundance of large cladocerans (*Daphnia*)

and copepods declining and small cladocerans and *Holopedium* increasing below the common critical Ca threshold of 1.3 mg L^{-1}, lake Ca concentrations are indeed important for determining zooplankton community composition in northern lakes.

We also found that lake pH was an important determinant for the spatial distribution of especially cyclopoids (ranked first) but also for *Daphnia* and small cladocerans (ranked secondly; Figure 3), where cyclopoids and *Daphnia* declined in relative abundance below their respective thresholds and with increased acidity (pH of 5.4 and 6.4, respectively). Relative abundance of cyclopoids has been shown to decline with declining pH in Canadian shield lakes (Palmer & Yan, 2013), likely attributed to the acid-sensitivity of common cyclopoids like *Cyclops scutifer* (Holt et al., 2003), a cyclopoid species also common in Scandinavian lakes. Acid sensitivity among *Daphnia* is well known (Hessen et al., 1995a; Holt et al., 2003) and interacts with lake Ca concentrations in shaping abundances of large sized *Daphnia* (Ross & Arnott, 2021). The close coupling between Ca and pH makes it hard for a strict separation of the effects of these two parameters on relative *Daphnia* abundances. Our random forest analysis for the combined model also showed almost identical importance of lake Ca concentrations as for pH, suggesting almost equal importance of Ca and pH for *Daphnia*. Yet, for cyclopoids pH was clearly more important compared to Ca. For *Holopedium*, TOC was slightly higher in importance in the combined model before Ca. *Holopedium* is recognized as a species indicator of lakes of low TOC, but also of low Ca and low pH (Derry & Arnott, 2011; Hessen et al., 1995a), which is supported by our results (e.g., *Holopedium* increased in relative abundance below TOC- and Ca thresholds of 4.4 and 1.3 mg L^{-1}, respectively).

Although TP concentration was found to weakly impact community composition, it should, however, be important for overall zooplankton biomass (McCauley & Kalff, 1981). Declining trends in TP should also impact phytoplankton biomass. Phytoplankton use not only dissolved inorganic P, but also P derived from enzymatic hydrolysis of dissolved organic P and stored P (Wetzel, 2001), and from humic-Fe-P complexes (Jansson et al., 2001), wherefore bioavailable pools of P in oligotrophic lakes correspond to TP (Bergström, 2010; Lewis & Wurtsbaugh, 2008). Since TP was correlated with TOC, and due to the unimodal impact of TOC on phytoplankton biomass depending on TOC: nutrient stoichiometry (Isles et al., 2021), it might be hard to identify thresholds in TP for different zooplankton groups. Spatially though (see Figure 1), P demanding *Daphnia* (Andersen & Hessen, 1991) was low in relative abundance in P-poor lakes to the west where the less P demanding *Holopedium* (Jeziorski et al., 2015) was high in relative abundance. Temperature was not identified as one of the important factors in determining thresholds (Figure 3) but came out second in order after Ca for calanoids and third for *Daphnia* after Ca and pH in the combined model. Warming combined with reduced algal biomass has been shown to enhance *Daphnia* susceptibility to low Ca (Ashforth & Yan, 2008). Temperature might have had a similar impact in our study lakes.

Overall, our data suggest that lake Ca concentration is the first to second most important determinant for the spatial distribution of all

the assessed zooplankton groups. While there are good mechanistic explanations for taxon-specific Ca-responses linked with their Ca demands (see above), the distribution and the relative abundance of different zooplankton responses are undoubtedly impacted and influenced by other physico-chemical factors (see above) and biological factors, some of which may be confounded with Ca. For example, inter-specific differences in feeding strategy and competitive edge for food resources may influence the relative abundance of zooplankton groups (Gliwicz, 1990; Hessen, 1985). Further, predator–prey interactions among zooplankton taxa (Brandl, 2005; Gliwicz & Umana, 1994), or between fish and zooplankton, (Andersen et al., 2020; Brooks & Dodson, 1965; Langeland & Nøst, 1995), may also be influential here. It is known that cyprinid fish abundances, which are efficient planktivores, increase from west to east in the landscape (Andersen et al., 2020; Langeland & Nøst, 1995), like the observed spatial gradient in Ca (Figure 1); however, no conclusive data on fish abundance were available in all study lakes, so we were unable to test fish influence on the relative distribution of different zooplankton. The cumulative effects of these factors may contribute to the observed spatial patterns in the relative abundance of zooplankton groups.

4.2 | Are we moving closer to or away from the critical Ca thresholds?

The temporal assessment revealed that most lakes are experiencing Ca declines (Figure 4), and that these declines are either moving toward or falling below the critical Ca thresholds for individual zooplankton groups, and so substantial shifts in zooplankton community composition are likely to occur. While pH likely contributes to structuring zooplankton communities (see above), the monotonic trends of Ca show that Ca is changing in a more consistent and significant rate, so it is likely the most important variable in shaping the communities. Ca declines probably will continue, as they are associated with ongoing global change drivers including recovery from acidification and climate change (de Wit et al., 2016, 2023; Futter et al., 2014), as well as forest harvesting that removes base cations from the catchment (Joki-Heiskala et al., 2003; Moldan et al., 2017; Watmough et al., 2003), and adjusted or halted lake liming.

Above the critical Ca threshold (Figure 5), the declining Ca trend was associated with increasing trends of small cladocerans and *Holopedium* of low Ca demand (Azan & Arnott, 2018; Jeziorski et al., 2015) and a declining trend of calanoids and especially cyclopoids of high Ca demand (Azan & Arnott, 2018), as expected. However, this declining Ca trend was also associated with an increasing trend of large cladocerans of high Ca demand, for example, *Daphnia* (Jeziorski et al., 2015), which was not expected. It could be that above the Ca threshold even though Ca was declining the lake Ca concentrations was still high enough ($3\,mg\,L^{-1}$) so that *Daphnia* may have benefitted from the slight improvement in lake pH following acidification recovery (see Figure 4; Yan et al., 2008) and/or from the significant TOC increase promoting increased phytoplankton

biomass (see Figure 4; Isles et al., 2021). It also seems unlikely that fish predation could be the main driver for the temporal trends in zooplankton community composition above the critical Ca threshold, unless fish abundances also changed consistently with time. Our data suggest Ca is the main driver of the temporal trends in zooplankton community composition above the critical Ca threshold, although these trends are affected to some extent by other factors such as pH and TOC.

Below the critical Ca threshold, the relative abundances in zooplankton followed their unidirectional trends above their thresholds (Figure 5), with the notable exceptions for *Daphnia* and *Holopedium*. *Daphnia* dropped immediately below the Ca threshold of $1.3\,mg\,L^{-1}$ and remained low in relative abundance over time, whereas *Holopedium* relative abundances dropped more gradually with time from around 20% in 2000 to only a few percent in 2019, which was associated with the declining Ca trend. These losses in relative abundances of *Daphnia* and *Holopedium* coincided with extremely low Ca levels of 0.5–$1\,mg\,L^{-1}$ (see Figure 5) and cannot be attributed to intensified fish predation, because these low Ca western lakes (Figure 1) are characterized by the absence of cyprinids and overall lower fish predation (Andersen et al., 2020). Our study therefore illustrates that waters with extremely dilute Ca levels of $\leq 0.5\,mg\,L^{-1}$ seem detrimental or even lethal for *Daphnia* and *Holopedium*. Such low levels are neither uncommon in Nordic waters (Hessen et al., 2017), nor globally (Weyhenmeyer et al., 2019), meaning that reductions and fluctuations in Ca levels may have far-reaching consequences, not only for zooplankton, but for a wide range of calcifying or bony organisms, for example, mollusca. Moreover, the significant increase in relative abundance of small cladocerans below the critical Ca threshold may, in addition to low Ca, be an effect of released competition for available food resources (Gliwicz, 1990; Hessen, 1985). That is, when the generalist, filter-feeding *Daphnia* abundance is low, and when the relative abundances of *Holopedium* and cyclopoids declines, small cladocerans will have increasingly higher access to both small and large food particles not being utilized by *Daphnia*, *Holopedium* or cyclopoids (Brandl, 2005; Gliwicz, 1990; Hessen, 1985).

4.3 | Lake Ca declines promotes small bodied and less nutritious zooplankton

We found that lake Ca concentrations were strongly associated with differences in the relative abundance of both cladocerans and copepods, and that community shifts are likely to occur when Ca concentrations falls below 0.9–$1.3\,mg\,L^{-1}$; for example, from groups with high Ca demand (*Daphnia* spp. and copepods) toward groups of low Ca demand (*Holopedium gibberum* and small cladocerans). Consistent declining Ca trends in lakes are further associated with significant trends in relative abundance of zooplankton toward increasing dominance by especially small cladocerans. The trajectories in zooplankton community composition that are linked to declining Ca therefore suggest large impacts on the nutritional content of zooplankton for fish. In addition of

impacting Ca content (Hessen et al., 2000), TP content in zooplankton is likely to be reduced making them less nutritious for fish (i.e., P-limitation is relevant also for fish; Benstead et al., 2014; Sterner et al., 2002) since small cladocerans and *Holopedium* have lower P content compared to *Daphnia* and cyclopoids (Andersen & Hessen, 1991; Bergström et al., 2022; Jeziorski et al., 2015). It is also likely that these community shifts will impact the overall polyunsaturated fatty acid (PUFA) composition of zooplankton, since copepods are rich in docosahexaenoic acid (DHA) but cladocerans are rich in eicosapentaenoic acid (EPA) instead (Lau et al., 2021; Persson & Vrede, 2006). Decreases in dominance by copepods in zooplankton with Ca declines may suggest reduced dietary supply of DHA to fish that is strongly needed for neural development and physiological functions (Sargent et al., 2003). However, freshwater fish are often efficient to regulate their PUFA content by conversion, indicated in relatively stable EPA and DHA content across lake productivity gradient (Keva et al., 2021). Additionally, shifts in zooplankton community and diversity driven by declines in Ca to dominance of small cladocerans may have negative impact on visually feeding and size selective fish predators (Brooks & Dodson, 1965; Lazarro, 1987), especially if there is no increase in zooplankton abundance and if the Ca declines is accompanied with lake browning (increased TOC; Hayden et al., 2017). Although our data point to a dominance of especially small cladocerans at the expense of larger individuals (*Daphnia and* cyclopoids) with declining Ca trends, to what extent these community shifts mean in terms of trends in zooplankton biomass is not possible for us to evaluate since our data did not allow for biomass estimates. Future research should therefore include biomass estimates to evaluate how declining Ca trends might impact and be associated with trends in zooplankton biomass.

4.4 | Global change and zooplankton

The large-scale impacts of global change [e.g., warming, hydrological intensification, acidification recovery, extension of vegetation cover, and forest harvesting (Creed et al., 2018; Finstad et al., 2016; Moldan et al., 2017; Watmough et al., 2003)], are likely to promote continued declines in catchment delivery and lake concentrations of Ca that might hamper plankton and consumer development in northern lakes. Yet, other lake environmental variables, and changes in these, such as pH (Futter et al., 2014), TP (Isles et al., 2023), TOC (de Wit et al., 2023) and warming (Puts et al., 2023), may modify the responses in zooplankton to declining Ca. While Ca-poor lakes are prevalent globally (Weyhenmeyer et al., 2019), and the decrease in Ca poses challenges to the fauna in these lakes, lake at low latitudes—typically more buffered and nutrient rich—show little tendency of Ca decline, even when previously exposed to acid deposition (cf., Weyhenmeyer et al., 2019). This indicates that for temperate low latitude lakes other environmental variables such as pH, TP, and TOC, and warming, might have a greater impact compared to Ca in affecting the composition of the zooplankton communities. Moreover, because of the different richness and community composition of zooplankton in high-versus low-latitude lakes, the conclusions on species replacement are not directly transferable to low-latitude lakes. In addition, a large part of northern lakes is situated in boreal catchment, that impact water properties, for example, by high concentrations of TOC. Clearly more comparative studies in lakes across different latitudes are required to assess responses of zooplankton communities and zooplankton biomass changes to global change. As northern high-latitude lakes by number are the most common lake type globally (Verpoorter et al., 2014), and are vulnerable to environmental changes for example, because of their oligotrophic and soft water nature (Isles et al., 2023; Weyhenmeyer et al., 2019), as well as changes in catchment inputs of TOC and nutrients (Isles et al., 2021), it is essential to assess these impacts that may have severe consequences on the productivity and ecosystem services provided by northern lakes.

DATA AVAILABILITY STATEMENT

Raw data on water chemistry and zooplankton from Swedish lakes is publicly available at the Department of Aquatic Sciences and Assessment, Swedish University of Agricultural Sciences at https://miljodata.slu.se or https://miljodata.slu.se/MVM/Search. Raw data on water chemistry and zooplankton from Norwegian lakes is hosted at Norwegian Institute for Water Research (NIVA) and is openly available in Zenodo at https://doi.org/10.5281/zenodo.10694066. The data that support the findings of this study are openly available in Zenodo at https://zenodo.org/records/10694066. Air temperatures for each lake were extracted from the TerraClimate datasets (https://www.nature.com/articles/sdata2017191).

CODE AVAILABILITY STATEMENT

The code used to analyse the data is available at: https://github.com/AleseyPA/regression-trees.git.

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REFERENCES

Abatzoglou, J. T., Dobrowski, S. Z., Parks, S. A., & Hegewisch, K. C. (2018). Terraclimate, a high-resolution global dataset of monthly climate and climatic water balance from 1958–2015. *Scientific Data*, 5, 170191.

Andersen, T., & Hessen, D. O. (1991). Carbon, nitrogen, and phosphorus content of freshwater zooplankton. *Limnology and Oceanography*, 36, 807–814.

Andersen, T., Hessen, D. O., Håll, J. P., Khomich, M., Kyle, M., Lindholm, M., Rasconi, S., Skjelbred, B., Thrane, J.-E., & Walseng, B. (2020). Congruence, but no cascade—Pelagic biodiversity across three trophic levels in Nordic lakes. *Ecology and Evolution*, 10, 8153–8165.

Arnott, S. E., Azan, S. S. E., & Ross, A. J. (2017). Calcium decline reduces population growth rates of zooplankton in field mesocosms. *Canadian Journal of Zoology*, 95, 323–333.

Ashforth, D., & Yan, N. D. (2008). The interactive effects of calcium concentration and temperature on the survival and reproduction of Daphnia pulex at high and low food concentrations. *Limnology and Oceanography*, 53(2), 420–432.

Azan, S. S., & Arnott, S. E. (2018). The impact of calcium decline on population growth rates of crustacean zooplankton in Canadian Shield lakes. *Limnology and Oceanography*, 63, 602–616.

Benstead, J. P., Hood, J. M., Whelan, N. V., Kendrick, M. R., Nelson, D., Hanninen, A. F., & Demi, L. M. (2014). Coupling of dietary phosphorus and growth across diverse fish taxa: A meta-analysis of experimental aquaculture studies. *Ecology*, 95, 2768–2777.

Berggren, M., Bergström, A.-K., & Karlsson, J. (2015). Intraspecific autochthonous and allochthonous resource use by zooplankton in a humic lake during the transitions between winter, summer, and fall. *PLoS One*, 10, 1–14.

Bergström, A.-K. (2010). The use of use of TN:TP and DIN:TP ratios as indicators for phytoplankton nutrient limitation in oligotrophic lakes affected by N deposition. *Aquatic Sciences*, 72, 277–281.

Bergström, A.-K., Deininger, A., Jonsson, A., Karlsson, J., & Vrede, T. (2021). Effects of nitrogen enrichment on zooplankton biomass and N:P recycling ratios across a DOC gradient in northern-latitude lakes. *Hydrobiologia*, 848, 4991–5010.

Bergström, A.-K., Lau, D. C. P., Isles, P. D. F., Jonsson, A., & Creed, I. F. (2022). Biomass, community composition and N:P recycling ratios of zooplankton in northern high-latitude lakes with contrasting levels of N deposition and dissolved organic carbon. *Freshwater Biology*, 67, 1508–1520.

Brandl, Z. (2005). Freshwater copepods and rotifers: Predators and their prey. *Hydrobiologia*, 546, 475–489.

Brooks, J. L., & Dodson, S. I. (1965). Predation, body size, and composition of plankton. *Science*, 150, 28–35.

Buckley, L. B., & Huey, R. B. (2016). How extreme temperatures impact organisms and the evolution of their thermal tolerance. *Integrative and Comparative Biology*, 56, 98–109.

Couture, S., Hudon, C., Gagnon, P., Taranu, Z. E., Pinel-Alloul, B., Houle, D., Aldamman, L., Beauvais, C., & Lachapelle, M. (2021). Zooplankton communities in Precambrian Shield lakes (Quebec, Canada): Responses to spatial and temporal gradients in water chemistry and climate. Canadian journal of fisheries and aquatic sciences. *Canadian Journal of Aquatic Sciences*, 78, 567–579.

Creed, I. F., Bergström, A.-K., Trick, C. G., Grimm, N. B., Hessen, D. O., Karlsson, J., Kidd, K. A., Kritzberg, E., McKnight, D. M., Freeman, E. C., Senar, O. E., Andersson, A., Ask, J., Berggren, M., Cherif, M., Giesler, R., Hotchkiss, E. R., Kortelainen, P., Palta, M. M., … Weyhenmeyer, G. A. (2018). Global change-driven effects on dissolved organic matter composition: Implications for food webs of northern lakes. *Global Change Biology*, 24, 3692–3714.

de Wit, H. A., Garmo, Ø. A., Jackson-Blake, L. A., Clayer, F., Vogt, R. D., Austnes, K., Kaste, Ø., Gundersen, C. B., Guerrerro, J. L., & Atle Hindar, A. (2023). Changing water chemistry in one thousand Norwegian lakes during three decades of cleaner air and climate change. *Global Biogeochemical Cycles*, 37, e2022GB007509.

de Wit, H. A., Valinia, S., Weyhenmeyer, G. A., Futter, M. N., Kortelainen, P., Austnes, K., Hessen, D. O., Räike, A., Laudon, H., & Vuorenmaa,

J. (2016). Current browning of surface waters will be further promoted by wetter climate. *Environmental Science and Technology Letters*, 3, 430–435.

De'ath, G., & Fabricius, K. E. (2000). Classification and regression trees: A powerful yet simple technique for ecological data. *Ecology*, 81, 3178–3192.

Derrick, T. R., & Joshua, M. T. (2004). Time series analysis: The cross-correlation function. In N. Stergiou (Ed.), *Innovative analyses of human movement* (pp. 189–205). Human Kinetic publishers.

Derry, A. M., & Arnott, S. E. (2011). Zooplankton community response to experimental acidification in shield lakes with different ecological histories. *Canadian Journal of Aquatic Sciences*, 78, 567–579.

Eimers, M. C., Watmough, S. A., Paterson, A. M., Dillon, P. J., & Yao, H. (2009). Long-term declines in phosphorus export from forested catchments in south-central Ontario. *Canadian Journal of Fisheries and Aquatic Sciences*, 66, 1682–1692.

Finstad, A. G., Andersen, T., Larsen, S., Tominaga, K., Blumentrath, S., de Wit, H. A., Tømmervik, H., & Hessen, D. O. (2016). From greening to browning: Catchment vegetation development and reduced S-deposition promote organic carbon load on decadal time scales in Nordic lakes. *Scientific Reports*, 6, 1–8.

Futter, M. N., Valinia, S., Löfgren, S., Köhler, S. J., & Fölster, J. (2014). Long-term trends in water chemistry of acid-sensitive Swedish lakes show slow recovery from historic acidification. *Ambio*, 43, 77–90.

Garmo, Ø. A., Skjelkvåle, B. L., de Wit, H. A., Colombo, L., Curtis, C., Fölster, J., Hoffmann, A., Hruška, J., Høgåsen, T., Jeffries, D. S., Keller, W. B., Krám, P., Majer, V., Monteith, D. T., Paterson, A. M., Rogora, M., Rzychon, D., Steingruber, S., John, L., ... Worsztynowicz, A. (2014). Trends in surface water chemistry in acidified areas in Europe and North America from 1990 to 2008. *Water, Air, & Soil Pollution*, 225, 1880.

Gliwicz, Z. M. (1990). Food thresholds and body size in Cladocerans. *Nature*, 343, 638–640.

Gliwicz, Z. M., & Umana, G. (1994). Cladoceran body size and vulnerability to copepod predation. *Limnology and Oceanography*, 39, 419–442.

Hallstan, S., Trigal, C., Johansson, K. S. L., & Johnson, R. K. (2013). The impact of climate on the geographical distribution of phytoplankton species in boreal lakes. *Oecologia*, 173, 1625–1638.

Hayden, B., Myllykangas, J.-P., Rolls, R. J., & Kahilainen, K. K. (2017). Climate and productivity shape fish and invertebrate community structure in subarctic lakes. *Freshwater Biology*, 62, 990–1003.

Hessen, D. O. (1985). Filtering structures and particle size selection in coexisting Cladocera. *Oecologia*, 66, 368–372.

Hessen, D. O. (1992). Nutrient element limitation of zooplankton production. *The American Naturalist*, 140, 799–814.

Hessen, D. O., Alstad, N. E. W., & Skardal, L. (2000). Calcium limitation in Daphnia magna. *Journal of Plankton Research*, 22, 553–568.

Hessen, D. O., Andersen, T., Brettum, P., & Faafeng, B. A. (2003). Phytoplankton contribution to sestonic mass and elemental ratios in lakes: Implications for zooplankton nutrition. *Limnology and Oceanography*, 48, 1289–1296.

Hessen, D. O., Andersen, T., Tominaga, K., & Finstad, A. G. (2017). When soft waters becomes softer; drivers of critically low levels of Ca in Norwegian lakes. *Limnology and Oceanography*, 62, 289–298.

Hessen, D. O., Bakkestuen, V., & Walseng, B. (2007). Energy input and zooplankton species richness. *Ecography*, 30, 749–758.

Hessen, D. O., Elser, J. J., Sterner, R. W., & Urabe, J. (2013). Ecological stoichiometry: An elementary approach using basic principles. *Limnology and Oceanography*, 58, 2219–2236.

Hessen, D. O., Faafeng, B. A., & Andersen, T. (1995a). Competition or niche segregation between *Holopedium* and *Daphnia*; empirical light on abiotic key parameters. *Hydrobiologia*, 307, 253–261.

Hessen, D. O., Faafeng, B. A., & Andersen, T. (1995b). Replacement of herbivore zooplankton species along gradients of ecosystem productivity and fish predation pressure. *Canadian Journal of Aquatic Sciences*, 52, 433–742.

Hindar, A., Garmo, Ø. A., Austnes, K., & Sample, J. E. (2020). Nasjonal innsjøundersøkelse 2019. NIVA-rapport 7530-2020.

Holt, C. A., Yan, N. D., & Somers, K. M. (2003). pH 6 as the threshold to use in critical load modeling for zooplankton community change with acidification in lakes of south-central Ontario: Accounting for morphometry and geography. *Canadian Journal of Aquatic Sciences*, 60, 151–158.

Huser, B. J., Futter, M. N., Wang, R., & Fölster, J. (2018). Persistent and widespread long-term phosphorus declines in boreal lakes in Sweden. *Science of the Total Environment*, 613-614, 240–249.

Isles, P. D. F., Creed, I. F., & Bergström, A. K. (2018). Recent synchronous declines in DIN:TP in Swedish lakes. *Global Biogeochemical Cycles*, 32, 208–225.

Isles, P. D. F., Creed, I. F., Hessen, D. O., Kortelainen, P., Paterson, M., Pomati, F., Rusak, J. A., Vuorenmaa, J., & Bergström, A.-K. (2023). Widespread synchrony in phosphorus concentrations in northern lakes linked to winter temperature and summer precipitation. *Limnology and Oceanography Letters*, 8, 639–648.

Isles, P. D. F., Creed, I. F., Jonsson, A., & Bergström, A.-K. (2021). Tradeoffs between light and nutrient availability across gradients of dissolved organic carbon lead to spatially and temporally variable responses of lake phytoplankton biomass to browning. *Ecosystems*, 24, 1837–1852.

Jansson, M., Bergström, A.-K., Drakare, S., & Blomqvist, P. (2001). Nutrient limitation of bacterioplankton and phytoplankton in humic lakes in northern Sweden. *Freshwater Biology*, 46, 653–666.

Jansson, M., Persson, L., DeRoos, A., Jones, R. I., & Tranvik, L. J. (2007). Terrestrial carbon and intraspecific size-variation shape lake ecosystems. *Trends in Ecology and Evolution*, 22, 316–322.

Jeziorski, A., Tanentzap, A. J., Yan, N. D., Paterson, A. M., Palmer, M. E., Korosi, J. B., Rusak, J. A., Arts, M. T., Keller, W., Ingram, R., Cairns, A., & Smol, J. P. (2015). The jellification of north temperate lakes. *Proceedings of the Royal Society B: Biological Sciences*, 282, 20142449.

Jeziorski, A., & Yan, N. D. (2006). Species identity and aqueous calcium concentrations as determinants of calcium concentrations of freshwater crustacean zooplankton. *Canadian Journal of Fisheries and Aquatic Sciences*, 63, 1007–1013.

Jeziorski, A., Yan, N. D., Paterson, A. M., DeSellas, A. M., Turner, M. A., Jeffries, D. S., Keller, B., Weeber, R. C., McNicol, D. K., Palmer, M. E., McIver, K., Arseneau, K., Ginn, B. K., Cumming, B. F., & Smol, J. P. (2008). The widespread threat of calcium decline in freshwaters. *Science*, 322, 1374–1377.

Joki-Heiskala, P., Johansson, M., Holmberg, M., Mattsson, T., Forsius, M., & Kortelainen, P. (2003). Long-term base cation balances of forest mineral soils in Finland. *Water, Air and Soil Pollution*, 150, 255–273.

Kendall, M. G. (1975). *Rank correlation methods* (4th ed.). Charles Griffin (Oxford University Press). ISBN-13: 978-0195208375.

Keva, O., Taipale, S. J., Hayden, B., Thomas, S. M., Vesterinen, J., Kankaala, P., & Kahilainen, K. K. (2021). Increasing temperature and productivity change biomass, trophic pyramids and community-level omega-3 fatty acid content in subarctic lake food webs. *Global Change Biology*, 27, 282–296.

Kingsolver, J. G., & Buckley, L. B. (2017). Quantifying thermal extremes and biological variation to predict evolutionary responses to changing climate. *Philosophical Transactions of the Royal Society B: Biological Sciences*, 372, 20160147.

Langeland, A., & Nøst, T. (1995). Gill raker structure and selective predation on zooplankton by particulate feeding fish. *Journal of Fish Biology*, 47, 719–732.

Lau, D. C. P., Jonsson, A., Isles, P. D., Creed, I. F., & Bergström, A. K. (2021). Lowered nutritional quality of plankton caused by global environmental changes. *Global Change Biology*, 27, 6294–6306.

Lazarro, X. (1987). A review of planktivorous fishes: A review of planktivorous fishes: Their evolution, feeding behaviours, selectivities, and impacts. *Hydrobiologia*, 146, 97–167.

Lewis, W. M., & Wurtsbaugh, W. A. (2008). Control of lacustrine phytoplankton by nutrients: Erosion of the phosphorus paradigm. *International Review of Hydrobiology*, 93, 446–465.

Livingstone, D. M., & Lotter, A. F. (1998). The relationship between air and water temperatures in lakes of the Swiss Plateau: A case study with palæolimnological implications. *Journal of Paleolimnology*, 19, 181–198.

McCauley, E., & Kalff, J. (1981). Empirical relationships between phytoplankton and zooplankton biomass in lakes. *Canadian Journal of Aquatic Sciences*, 38, 458–463.

McCombie, A. M. (1959). Some relations between air temperatures and the surface water temperatures of lakes. *Limnology and Oceanography*, 4, 252–258.

Moldan, M., Stadmark, J., Fölster, J., Jutterström, S., Futter, M. N., Cosby, B. J., & Wright, R. F. (2017). Consequences of intensive forest harvesting on the recovery of Swedish lakes from acidification and on critical load exceedances. *Science of the Total Environment*, 603–604, 562–569.

Monteith, D. T., Stoddard, J. L., Evans, C. D., de Wit, H. A., Forsius, M., Høgåsen, T., Wilander, A., Skjelkvåle, B. L., Jeffries, D. S., Vuorenmaa, J., Keller, B., Kopacek, J., & Vesely, J. (2007). Dissolved organic carbon trends resulting from changes in atmospheric deposition chemistry. *Nature*, 450, 537–541.

Oliver, E. C. J., Donat, M. G., Burrows, M. T., Moore, P. J., Smale, D. A., Alexander, L. V., Benthuysen, J. A., Feng, M., Sen Gupta, A., Hobday, A. J., Holbrook, N. J., Perkins-Kirkpatrick, S. E., Scannell, H. A., Starub, S. C., & Wernberg, T. (2018). Longer and more frequent marine heatwaves over the past century. *Nature Communications*, 9, 1324.

O'Reilly, C. M., Rowley, R. J., Schneider, P., Lenters, J. D., Mcintyre, P. B., Kraemer, B. M., Weyhenmeyer, G. A., Straile, D., Dong, B., Adrian, R., Allan, M. G., Anneville, O., Arvola, L., Austin, J., Bailey, J. L., Baron, J. S., Brookes, J. D., de Eyto, E., Dokulil, M. T., … Zhang, G. (2015). Rapid and highly variable warming of lake surface waters around the globe. *Geophysical Research Letters*, 42, 10773–10781.

Palmer, M., & Yan, N. D. (2013). Decadal-scale regional changes in Canadian freshwater zooplankton: The likely consequence of complex interactions among multiple anthropogenic stressors. *Freshwater Biology*, 58, 1366–1378.

Paltsev, A., & Creed, I. F. (2022). Are northern lakes in relatively intact temperate forests showing signs of increasing phytoplankton biomass? *Ecosystems*, 25, 727–755.

Persson, J., & Vrede, T. (2006). Polyunsaturated fatty acids in zooplankton: Variation due to taxonomy and trophic position. *Freshwater Biology*, 51, 887–900.

Prater, C., Wagner, N. D., & Frost, P. C. (2016). Effects of calcium and phosphorus limitation on the nutritional ecophysiology of *Daphnia*. *Limnology and Oceanography*, 61, 268–278.

Puts, I. C., Ask, J., Deininger, A., Jonsson, A., Karlsson, J., & Bergström, A.-K. (2023). Browning affects pelagic productivity in northern lakes by surface water warming and carbon fertilization. *Global Change Biology*, 29, 375–390.

R Core Team. (2022). *R: A language and environment for statistical computing*. R Foundation for Statistical Computing.

Ross, A. J., & Arnott, S. E. (2021). Similar zooplankton responses to low pH and calcium may impair long-term recovery from acidification. *Ecological Applications*, 32, e2512.

Sargent, J. R., Tocher, D. R., & Bell, J. G. (2003). The lipids. In J. E. Halver & R. W. Hardy (Eds.), *Fish nutrition* (3rd ed., pp. 181–257). Elsevier (Academic Press).

Schartau, A. K., Birkeland, I. B., Bodin, C. L., Garmo, Ø., Lie, E. F., Saksgård, R., Skancke, L. B., Velle, G., & Walseng, B. (2020). Forsuringstilstand og trender i norske innsjøer og elver med biologisk overvåking. Effects of long-range transported pollutants in Norwegian lakes and rivers with biological monitoring – Acidification status and trends. Overvåkningsrapport M-1823 – 2020. Norwegian Environment Agency.

Sen, P. K. (1968). Estimates of the regression coefficient based on Kendall's tau. *Journal of the American Statistical Association*, 63, 1379–1389.

Sterner, R. W., Clasen, J., Lampert, W., & Weisse, T. (2002). Carbon:Phosphorus stoichiometry and food chain production. *Ecology Letters*, 1, 146–150.

Strobl, C., Malley, J., & Tutz, G. (2009). An introduction to recursive partitioning: Rationale, application, and characteristics of classification and regression trees, bagging, and random forests. *Psychological Methods*, 14, 323–348.

Tan, Q.-G., & Wang, W.-X. (2009). The regulation of calcium in Daphnia magna reared in different calcium environments. *Limnology and Oceanography*, 54, 746–756.

Tan, Q.-G., & Wang, W.-X. (2010). Interspecies differences in calcium content and requirement in four freshwater cladocerans explained by biokinetic parameters. *Limnology and Oceanography*, 55, 1426–1434.

Tolosi, L., & Lengauer, T. (2011). Classification with correlated features: Unreliability of feature ranking and solutions. *Bioinformatics*, 27, 1986–1994.

Toming, K., Kotta, J., Uuemaa, E., Sobek, S., Kutser, T., & Tranvik, L. J. (2020). Predicting lake dissolved organic carbon at a global scale. *Scientific Reports*, 18, 8471.

Verpoorter, C., Tiit Kutser, T., Seekell, D. A., & Tranvik, L. J. (2014). A global inventory of lakes based on high-resolution satellite imagery. *Geophysical Research Letters*, 41, 6396–6402.

Wærvågen, S. B., Rukke, N. A., & Hessen, D. O. (2002). Calcium content of crustacean zooplankton and its potential role in species distribution. *Freshwater Biology*, 47, 1866–1878.

Wasserstein, R. L., & Lazar, N. A. (2016). The ASA Statement on *p*-values: Context, process, and purpose. *American Statistician*, 70, 129–131.

Wasserstein, R. L., Schirm, A. L., & Lazar, N. A. (2019). Moving to a world beyond "*p* < 0.05". *American Statistician*, 73, 1–19.

Watmough, S. A., Aherne, J., & Dillon, P. J. (2003). Potential impact of forest harvesting on lake chemistry in south-central Ontario at current levels of acid deposition. *Canadian Journal of Aquatic Sciences*, 60, 1095–1103.

Wenzel, A., Vrede, T., Jansson, M., & Bergström, A.-K. (2021). Daphnia performance on diets containing different combinations of high-quality algae, heterotrophic bacteria, and allochthonous particulate organic matter. *Freshwater Biology*, 66, 157–168.

Wetzel, R. G. (2001). *Limnology: Lake and river ecosystem* (p. 1006). Academic Press.

Weyhenmeyer, G. A., Hartmann, J., Hessen, D. O., Kopáček, J., Hejzlar, J., Jacquet, S., Hamilton, S. K., Verburg, P., Leach, T. H., Schmid, M., Flaim, G., Nõges, T., Nõges, P., Wentzky, V. C., Rogora, M., Rusak, J. A., Kosten, S., Paterson, A. M., Teubner, K., … Zechmeister, T. (2019). Widespread diminishing anthropogenic effects on calcium in freshwaters. *Scientific Reports*, 9, 10450.

Yan, N. D., Somers, K. M., Girard, R. E., Paterson, A. M., Keller, W., Ramcharan, C. W., Rusak, J. A., Ingram, R., Morgan, G. E., & Gunn, J. M. (2008). Long-term trends in zooplankton of Dorset, Ontario, lakes: The probable interactive effects of changes in pH, total

phosphorus, dissolved organic carbon, and predators. *Canadian Journal of Fisheries and Aquatic Sciences, 65,* 862–877.

Zetterberg, T., Olsson, B. A., Löfgren, S., Hyvönen, R., & Brandtberg, O. (2016). Long-term soil calcium depletion after conventional and whole-tree harvest. *Forest Ecology and Management, 369,* 102–115.

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.