Aquat. Sci. (2010) 72:393–402
DOI 10.1007/s00027-010-0143-x

Aquatic Sciences

RESEARCH ARTICLE

# Temporal discontinuity of nutrient limitation in plankton communities

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Received: 22 October 2009 / Accepted: 27 April 2010 / Published online: 21 May 2010


**Abstract** Ideas on how various measures of nutrient limitation relate to plankton biomass and species composition are re-examined. While long-term and multi-lake studies typically focus on determining overall biomass, seasonal studies are more focused toward understanding species composition. We use physiological assays to assess short-term nutrient deficiency of nitrogen and phosphorus in two moderately fertile lakes. While biomass in the lakes was considered to ultimately be limited by total phosphorus, nutrient assays were variable in time. Nutrient ratios (TN:TP, PN:PP, PC:PP and PC:PN) did not predict short-term deficiencies, notably that nitrogen deficiency occurred in these phosphorus-limited lakes. In one of our study lakes, there was a relaxation of phosphorus deficiency despite phosphate concentrations occurring below traditional detection limits. Following this period, there was an autumn bloom of *Aphanizomenon flos-aquae*. This relationship corresponds with other studies that have found *A. flos-aquae* to be a poor competitor for phosphorus. In contrast, phosphorus deficiency remained high prior to the autumn diatom bloom in our other study lake. Deficiency measures remain an excellent means of assessing physiological status of plankton communities and provide greater insight into species compositional changes, especially when other potential indicators like dissolved nutrient concentrations are inconclusive. Regardless of the nutrient limitation indicator used for a given study, it is critical to consider the appropriate scale of the measure.

**Keywords** Nutrient limitation · Nutrient deficiency · Algal blooms · Cyanobacteria · Aphanizomenon · Fragillaria

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## Introduction

Aquatic nutrient limitation experiments typically aim to provide greater understanding and predictability of algal biomass and species composition. It is well established that addition of nutrients can increase phytoplankton biomass and change the composition of phytoplankton communities (Watson et al. 1997). However, debate remains over which nutrients limit phytoplankton production and how nutrients should be managed to control eutrophication (Elser et al. 1990; Schindler et al. 2008; Lewis and Wurtsbaugh 2008). For limitation experiments, it is critical to identify the relevant time scales associated with the different types of limitation measurements because some differences in this debate can be explained by differences in measurement scales and research objectives.

Nutrient limitation of pelagic phytoplankton communities has been the subject of numerous studies and a focal area of research for decades (Goldman et al. 1979; Vollenweider and Kerekes 1982). Nutrient limitation of

phytoplankton communities can, for argument sake, be viewed at two general time scales. The first is aimed at understanding long-term constraints on potential algal biomass. Ecologists infer which nutrients limit phytoplankton productivity and the severity of nutrient limitation through the use of proxy measures for algal biomass, such as total and particulate nitrogen (N) or phosphorus (P) and the ratio of these nutrients (N:P) (Hecky et al. 1993). Use of total and particulate nutrients can also be used to predict general characteristics of algal composition and species dominance (Smith 1983; Watson et al. 1997). It is generally recognized that total and particulate nutrient limitation is implied to mean a restriction of biomass attained in proportion to the nutrient in limited supply and is considered to apply to longer times scales (Schindler et al. 2008; Sterner 2008).

The second approach is to assess the immediate needs of the extant plankton community and assess factors affecting its composition. Physiological assessments aim at identifying such nutrient limitation at proximate scales. Phytoplankton physiological requirements operate at relatively short temporal scales (Yentsch et al. 1977; Healey and Hendzel 1980; Davies et al. 2004a) and differ among species despite overall nutrient limitation constraints to total biomass (St. Amand et al. 1989; Davies et al. 2004a). Plankton physiological status can be assessed using various nutrient deficiency bioassays. These are distinctly different from biomass limitation assays and therefore avoid many of the issues inherent with incubating plankton communities in bottles for extended periods under artificial conditions (Schindler et al. 2008).

The relationship between total and particulate measures of limitation and measures of deficiency has been examined and there is general agreement that nutrient ratios and physiological status measures concur (Hecky and Kilham 1988; Guildford and Hecky 2000). However, there is limited information on the relationships between total and particulate measures of limitation, short-term deficiency measures, and the seasonal dynamics and development of lake phytoplankton communities.

The current study assessed nutrient limitation and deficiency measurements seasonally in two lakes over a 2 year period. Both lakes are classified as meso-eutrophic and have similar TN:TP ($\sim$50:1 by moles; Table 1), which is indicative of P-limitation of the phytoplankton community (Guildford and Hecky 2000). However, the algal community of one lake was dominated by the cyanobacterium *Aphanizomenon flos-aquae* (Linnaeus) Ralfs ex Bornet and Flahault, while the other lake was dominated by the diatom *Fragilaria crotonensis* var. *crotonensis* (Kitton). The goal was to assess the congruency between traditional limitation and less commonly assessed deficiency measurements in relation to the ecology of these two lakes of similar average nutrient compositions but different algal assemblages.

**Table 1** Physical and chemical properties of Cusheon and Elk Lakes

| | Cusheon Lake | Elk Lake |
|---|---|---|
| Surface area (ha) | 31 | 246 |
| Max depth (m) | 9.5 | 20 |
| Mean depth (m) | 4.4 | 7.7 |
| TP (μg $L^{-1}$) | 16.1 | 17.6 |
| TN (μg $L^{-1}$) | 418 | 402 |
| SRSi (μg $L^{-1}$) | 7.43 | 0.84 |
| PC:PN | 8.8 | 9 |
| PC:PP | 202 | 200 |
| PN:PP | 24.2 | 21.8 |
| DOC (mg $L^{-1}$) | 5.2 | 5.4 |
| pH | 7.6 | 8.2 |
| Alkalinity (μEq $L^{-1}$) | 560 | 1,047 |

Nutrients, nutrient ratios (by moles), DOC and pH are epilimnetic summer averages (May–September) for the years 2000 and 2001. SRSi (soluble reactive silica), and alkalinity are averages for 2001. The epilimnetic SRSi concentration in Elk Lake for September 2001 was 0.55 (μg $L^{-1}$)

## Methods

The study lakes are on southern Vancouver Island (Elk Lake, 48°32′N 123°24′W) and Saltspring Island (Cusheon Lake 48°49′N 123°28′W), approximately 30 km apart, in British Columbia, Canada. The lakes are classified as warm monomictic with summer surface temperatures typical of north-temperate lakes at their latitude, but neither lake has substantial or permanent ice-cover during winter. Detailed sample collection and laboratory processing are described in Davies et al. (2004a, b) and Nowlin et al. (2007). Briefly, epilimnetic water was collected monthly during the summer at the point of maximum depth using a 5 cm-diameter integrated tube sampler during 2000 and 2001, and at least once during winters. Temperature and dissolved oxygen profiles were defined with a YSI model 58. Chlorophyll *a* was determined by filtering triplicate water samples onto separate GF/F filters (Whatman), and extracting chlorophyll from the filters with 95% ethanol. Total and dissolved nutrients were analyzed on a Lachat autoanalyzer (Zellweger Analytics, QuickChem® 8000), except for ammonium, which was analyzed manually with a spectrophotometer. Total phosphorus (TP), particulate phosphorus (PP), and total dissolved phosphorus (TDP) were analyzed on the same Lachat autoanalyzer after digesting samples in the autoclave with potassium persulfate. Water passed through a 0.45-μm membrane filter was analysed for TDP. TN was measured as nitrite using

unfiltered water samples autoclaved in alkaline potassium persulfate solution and reduced using a cadmium column. Total and dissolved nutrients were conducted in triplicate from three independently collected water samples. PP was collected on a 0.2-μm membrane filter. Particulate carbon (PC) and particulate nitrogen (PN) were collected on pre-combusted GF/F filters and analyzed on a CHN analyzer (Costech) using a Delta$^{plus}$ Advantage mass spectrometer as the detector.

Over the course of the study, four nutrient bioassays were conducted: N-debt, ammonia enhanced response (AER), P-debt, and $^{32}$P-turnover. N-debt and P-debt were conducted after Healey and Hendzel (1979, 1980) and represent, respectively, measures of nitrogen and phosphorus deficiency. Briefly, for each assay 100 mL of lake water was incubated in an Erlenmeyer flask in the dark for 24 h. For each sample prior to incubation, $Na_2HPO_4$ was added to one flask (P-debt) and $NH_4Cl$ was added to a second flask (N-debt), each at a concentration of 5 μmol $L^{-1}$. Nutrient analyses were conducted using a spectrophotometer to obtain pre- and post-incubation nutrient concentrations using the ascorbic acid (P-debt) and Berthelot reaction (N-debt). Net uptake rates were corrected for chl *a*. These bioassays are fundamentally different from nutrient enrichment bioassays that involve a factorial nutrient addition design to lake water and subsequent incubation under light saturated conditions for several days or longer. Often these studies use chlorophyll as the end-point biomass assessment. We avoided such nutrient enrichment bioassays because the growth conditions and response times can be unrealistic reflections of both short-term nutrient deficiency and long-term nutrient limitation. AER is a measure of nitrogen deficiency (Yentsch et al. 1977; Davies et al. 2004a). AER bioassays were only conducted in 2001 immediately prior to and during the autumn blooms. $^{32}PO_4$-uptake bioassays (Lean and White 1983; Mazumder et al. 1988) provide a measure of phosphorus deficiency. Uptake constants were calculated from at least duplicate water samples using carrier-free $^{32}PO_4^{3-}$ (activity 900–3200 Bq $mL^{-1}$) in 100 ml of whole lake water (Nowlin et al. 2007).

Phytoplankton were enumerated according to Utermöhl (1958) using an Olympus inverted microscope. Larger cells, colonies, and filaments were counted at lower magnification (100–200×) and smaller cells at higher magnification (400–630×). Dominant species were identified to genera/species, while smaller unidentified phytoplankton were grouped by size (most unidentified species were <10 μm). Biomass was determined assuming a volumetric to mass conversion of 1 $mm^3$ = 1 mg. Measurements of cells, colonies and filaments were conducted using imaging software (North Eclipse ver. 6) at the same time as counts were conducted. Phytoplankton shapes used to calculate biomass are after Hamilton (1990).

Heterocysts were counted separately to obtain a greater resolution of their biomass. When they were abundant, at least 100 heterocysts were enumerated. When they were less abundant, the number of heterocysts per at least 300 vegetative cells of N-fixing species was counted. Otherwise, the number of heterocysts was counted for 1,000 total phytoplankton cells. Heterocyst biomass was calculated using plankton shapes (Hamilton 1990) with the conversion factor noted above.

## Results

The average total nutrient concentrations and nutrient ratios for C, N and P were remarkably similar and only SRSi differed between lakes (Table 1). In contrast to similar average nutrient concentrations between the lakes, the composition of dominant phytoplankton was markedly different. This is highlighted by the late summer/autumn phytoplankton biomass in Cusheon Lake, which was dominated by the cyanophyte *A. flos-aquae*, whereas in Elk Lake it was dominated by the diatom *F. crotonensis* (Fig. 1). In 2000 and 2001, *A. flos-aquae* constituted 97 and 70%, respectively, of the total algal biomass of the September blooms in Cusheon Lake. In Elk Lake, *F. crotonensis* made up 79 and 58%, respectively, of the autumn blooms in 2000 and 2001. The cyanobacteria *A. flos-aquae* and *Anabaena* spp. were also visible at the surface of Elk Lake. In 2001 they made up approximately 40% of the August biomass and 10–14% of the September and October biomass.

Total phosphorus concentrations had similar seasonal patterns between the lakes with increases during the autumn period (Fig. 2). TN concentrations changed more dramatically in Cusheon Lake than Elk Lake, notably in September when TN values increased in both years. TN:TP ratios in both lakes consistently suggested P-limiting conditions during the stratified periods (Fig. 2c). The greatest biomass of heterocysts (Fig. 3) was observed in Cusheon Lake during the autumn bloom of *A. flos-aquae* (Fig. 1), which matches the period of increased TN concentration in the lake (Fig. 2).

Soluble reactive phosphorus (SRP) showed no consistent seasonal pattern in either lake, whereas dissolved inorganic nitrogen (DIN) demonstrated distinct and similar patterns in both lakes in both years, with the lowest concentrations observed in July–August (Fig. 4). Epilimnetic nitrogen deficiency in Cusheon Lake, as determined using the N-debt bioassay, peaked in July–August of both years. Nitrogen deficiency was not noted at other times of the year in either lake. In Elk Lake, N-debt was observed later in 2000 and was more prolonged in the summer of 2001 compared to Cusheon Lake (Fig. 4b). Periods of nitrogen

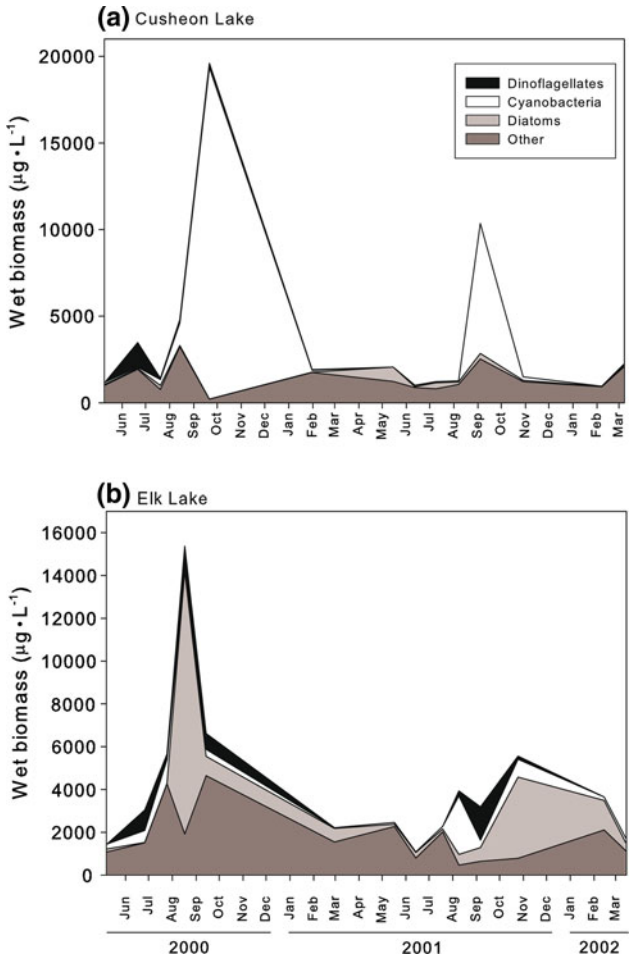


**Fig. 1** Biomass of phytoplankton groups for **a** Cusheon Lake and **b** Elk Lake. *Aphanizomenon flos-aquae* dominated the late summer/autumn bloom in Cusheon Lake, whereas *Fragilaria crotonensis* dominated the algal biomass in Elk Lake. The predominate dinoflagellate in Elk Lake was *Ceratium hirundinella*. Most species grouped in the 'other' category were small (<10 µm) phytoplankton

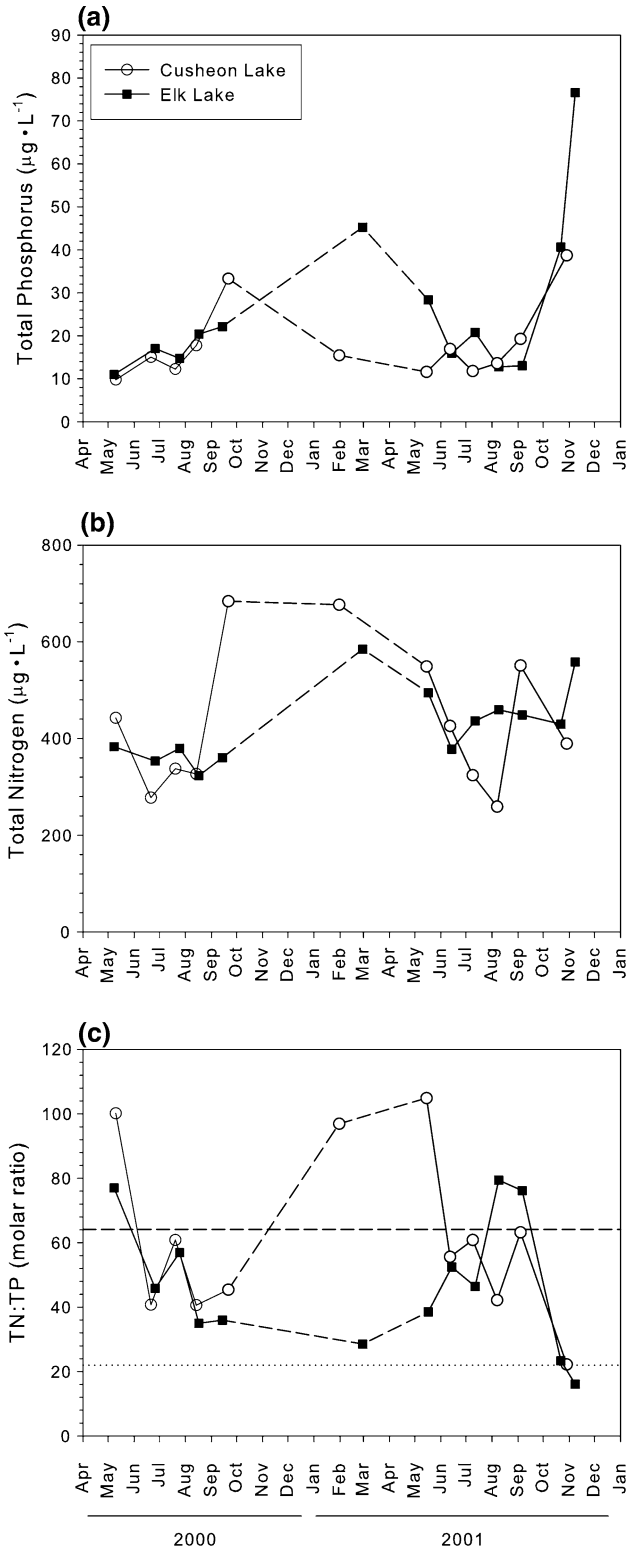


**Fig. 2** Temporal trends of **a** total phosphorus, **b** total nitrogen and **c** TN:TP ratio for Cusheon and Elk Lakes. The *dotted line* in **c** at TN:TP = 22 corresponds to the P-limiting threshold defined in Guildford and Hecky (2000) and the *dashed line* at TN:TP = 64 is the ratio of Smith (1983), above which cyanobacteria blooms are not expected to bloom

deficiency match well with the decreased concentrations of DIN (Fig. 4). The 2001 AER ratio suggests that, compared to Cusheon Lake, Elk Lake had greater nitrogen deficiency, and in agreement with N-debt, this deficiency was prolonged in 2001 (Fig. 4c).

Patterns of phosphorus deficiency, as measured using P-debt, differed between Cusheon and Elk Lake. Cusheon epilimnion was typically phosphorus deficient during summer months, except for August when, according to the P-debt bioassay, it effectively disappeared in both years (Fig. 4e). Elk Lake remained phosphorus deficient throughout the middle of the summer. For the most part, $^{32}PO_4^{3-}$-uptake times corresponded well with P-debt bioassays (Fig. 4f), where an increase in $^{32}PO_4^{3-}$ uptake time suggests a relaxation of phosphorus deficient conditions. Elk Lake had rapid (low) $^{32}PO_4^{3-}$-turnover times (deficient

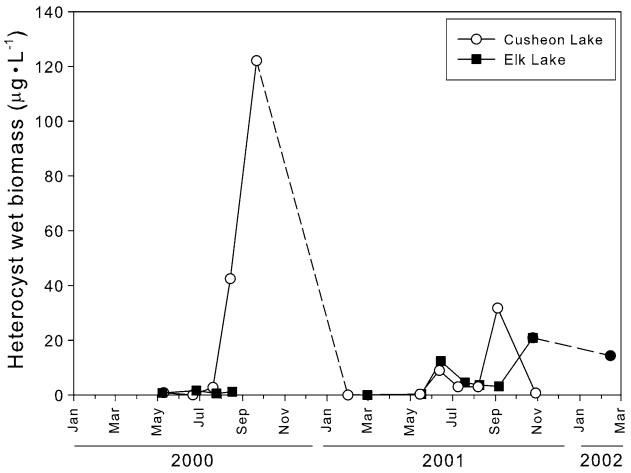


**Fig. 3** Temporal trends in heterocyst biomass for Cusheon and Elk Lakes

condition) during summer months, although this was relaxed somewhat in August 2000.

PC:PP and PN:PP ratios in Cusheon Lake peaked during the September *A. flos-aquae* bloom, suggesting that the blooms were phosphorus limited (Fig. 5a). These ratios varied between years in Elk Lake, in 2000 the ratios were not suggestive of severe P-limitation for much of the summer except in autumn, whereas in 2001 they were suggestive of severe P-limitation for most of the summer but not in late summer/autumn. PC:PN ratios in Cusheon and Elk Lake were not suggestive of severe nitrogen limitation.

The $\delta^{15}$N of the larger particulate organic matter (POM) fraction (>41 µm) decreased during late summer-autumn in both lakes (Fig. 6). In Elk Lake, the two POM size fractions demonstrated similar temporal trends, whereas in Cusheon Lake the pattern between the two size fractions were distinct. There was also a larger range in measured POM $\delta^{15}$N in Elk Lake (approximately 14‰) compared to the approximately 6‰ variation in Cusheon Lake over the 2 year study.

## Discussion

Many studies over the past several decades have investigated plankton nutrient limitation across the spectrum of temporal scales from examination of ecosystem carrying capacity, to population dynamics and individual species physiological response. Our study lakes had moderate to high levels of excess nitrogen relative to phosphorus (i.e. TN:TP), yet we observed physiological nutrient shortages of nitrogen in addition to expected phosphorus shortages in the plankton communities. This is evidence for the temporal uncoupling between ultimate limitation measures and proximate nutrient deficiencies. Thus, while assessment of ultimate limitation examines ecosystem carrying capacity, nutrient deficiency measures may provide insight into species successional patterns.

Despite similar total nutrients in our study lakes, species composition was substantially different in both non-bloom and bloom conditions. According to stoichiometric predictions, N:P values in both lakes were sufficiently low so as to be suitable for cyanobacterial growth (Smith 1983; Pick and Lean 1987; Smith et al. 1995). Bloom conditions are of particular ecological interest and management concern, so it is worth focusing on the autumn period in these two lakes and explore possible reasons for the difference in species composition. Since nutrient deficiency bioassays reflect changes in nutrient supply ratios in relation to the physiological requirements of the plankton community, they should provide insight into the difference in autumn bloom species. During August, prior to the autumn bloom formation, there was a difference in the P-deficiency status between the two lakes. Diatoms are generally competitive for inorganic phosphorus, whereas cyanobacteria populations of taxa such as *Microcystis*, *Anabaena* and *Aphanizomenon* are less competitive (Reynolds 1999). Likewise, Vahtera et al. (2007) found that cultures of *A. flos-aquae* were dependent on ample supplies of inorganic dissolved phosphorus and Riddolls (1985) suggested *A. flos-aquae* bloomed under conditions of low nitrate and plentiful phosphorus supplies. While our study did not undertake competition outcome studies per se, the reduction in phosphorus deficiency provided suitable conditions for *A. flos-aquae* to compete for P and establish critical density necessary for bloom development. In contrast, Elk Lake maintained high phosphorus deficiency during this period. This condition suggests that species more competitive for phosphorus, such as diatoms, should have a competitive advantage in Elk Lake. The findings of Reynolds (1999) and Vahtera et al. (2007) may be interpreted as contrasting with those of Ferber et al. (2004) who suggested a general condition of P-limitation is an accepted condition of cyanobacterial bloom formation. However, reconciliation between these positions may reside in scale-dependant interpretation. For example, in our study Cusheon Lake showed both conditions of ultimate P-limitation, while demonstrating a proximate release from P-deficiency prior to bloom formation. Furthermore, we found an increasing PN:PP ratio prior to and during the bloom. The increase in PN:PP may, in part, be a consequence of depleted phosphorus reserves (Vahtera et al. 2007). Regardless, it is critical to separate the conditions resulting after bloom formation from those necessary to form one (Reynolds 1999).

Dissolved nutrients in many lakes represent the readily available nutrient source for plankton. When

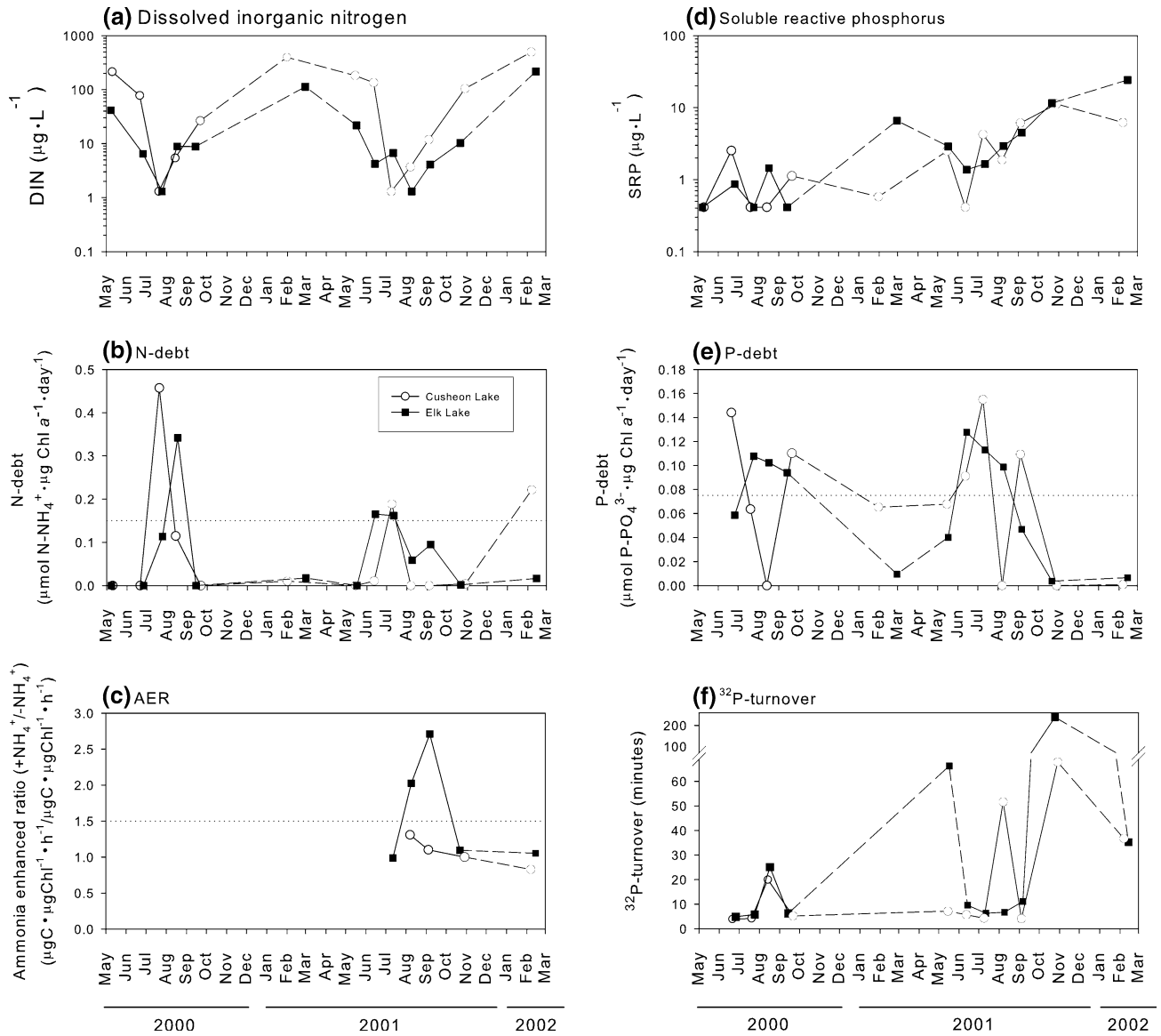


**Fig. 4** Epilimnetic temporal trends of **a** dissolved inorganic nitrogen (*DIN*), **b** N-debt, **c** ammonia enhanced response (*AER*), **d** soluble reactive phosphorus (*SRP*), **e** P-debt, and **f** $^{32}$P-turnover time. N-debt and AER are both measures of nitrogen deficiency where greater values reflects greater plankton community deficiency. P-debt is a measure of phosphorus deficiency, like N-debt greater values represent greater plankton community deficiency. $^{32}$P-turnover is a measure of phosphorus deficiency, unlike N-debt, AER and P-debt, higher values reflect lower measures of deficiency. The *dashed lines* in **b**, **c**, and **e** correspond to deficiency thresholds defined in Guildford and Hecky (2000) and Davies et al. (2004a)

concentrations of SRP or DIN are sufficient, P and N, respectively, should not be limiting (Prepas 1983; Auer et al. 1986; Cooke et al. 1993; Interlandi and Kilham 2001; Davies et al. 2004a). In our study, the seasonal patterns for DIN and N-debt were clearly aligned (Fig. 4). Researchers have concluded that conducting physiological assays provides little additional information over water chemistry (Hameed et al. 1999). However, the two do not always correspond. In our study, SRP provided little useful information on the phosphate pool and in fact represents substantial overestimates of the phosphate concentration (Nowlin et al. 2007). In contrast, P-debt and measures of $^{32}$P-turnover time provide important data on the relaxation of phosphorus in the plankton community. Since *A. flos-aquae* is not considered a strong competitor for phosphorus and SRP values remained low in Cusheon Lake, the physiological assays provide the basis, which is consistent with previous studies, for better understanding the conditions conducive for *A. flos-aquae* bloom development in Cusheon Lake.

Both lakes exhibited peaks of epilimnetic nitrogen deficiency during the summer, but no peaks in nitrogen

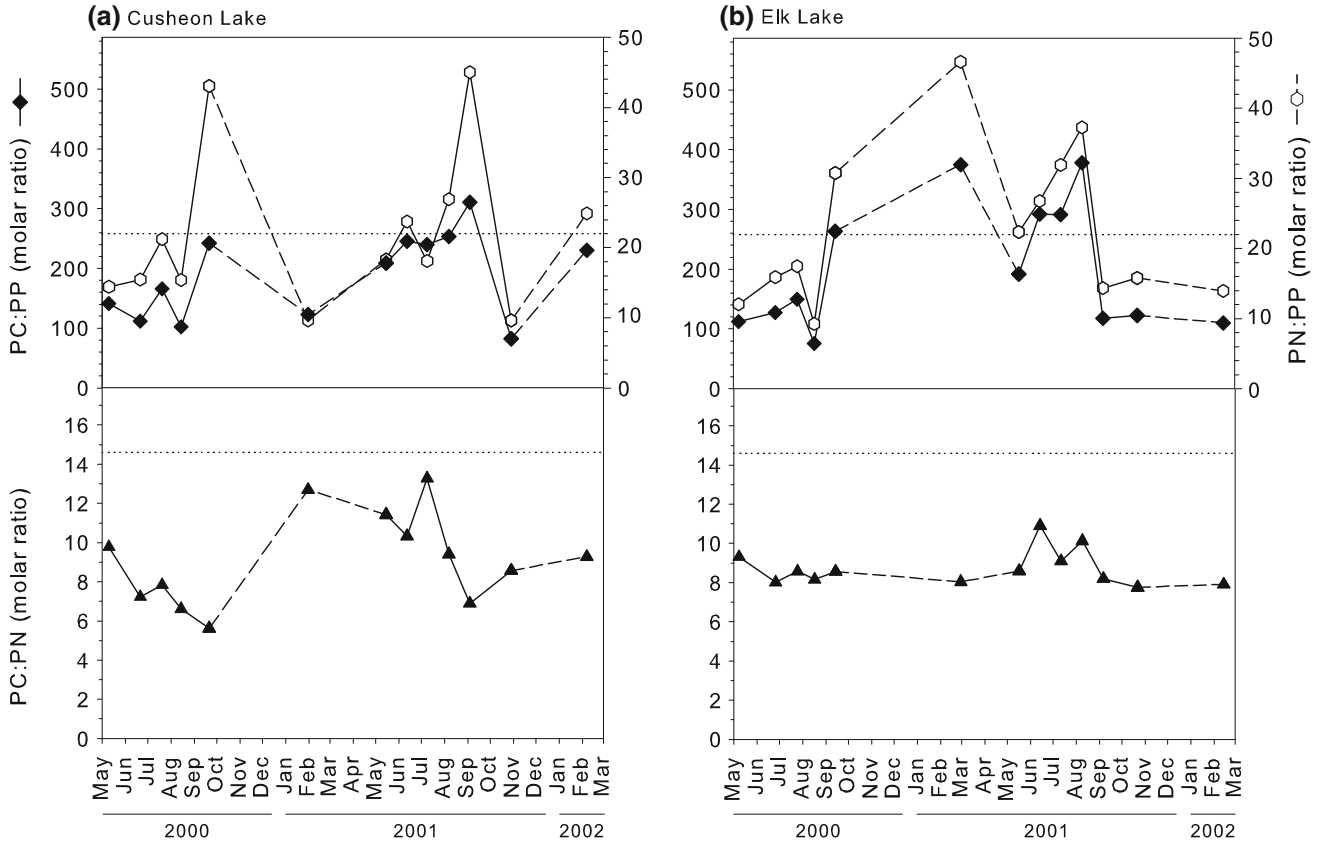


**Fig. 5** Temporal trends of particulate carbon to particulate phosphorus (PC:PP), particulate nitrogen to particulate phosphorus (PN:PP) and particulate carbon to particulate nitrogen (PC:PN) ratios for **a** Cusheon and **b** Elk Lakes. The *dashed lines* correspond to limitation thresholds defined in Guildford and Hecky (2000) and Davies et al. (2004a)

limitation (e.g., PC:PN), again implying discontinuity between measures of nutrient limitation and nutrient deficiency (Healey and Hendzel 1979; Davies et al. 2004a). It has been suggested that nitrogen deficiency is important in cyanobacterial bloom formation because of their ability to store and growth-moderate N and fix $N_2$ (Simon 1971; Allen and Weathers 1980; Carr 1988; Blomqvist et al. 1994). However, evidence supporting the importance of DIN in cyanobacterial bloom formation is equivocal: Lathrop (1988) found that additions of $NH_4^+$ and $NO_3^-$ did not prevent blooms of *Microcystis*, Stockner and Shortreed (1988) added $NH_4^+$ and $NO_3^-$ to an oligotrophic lake with low N:P and stimulated a bloom of *Anabaena*, whereas Varis et al. (1989) found a negative relationship for several cyanobacteria species (including *A. flos-aquae*) and ammonia, and Philips et al. (1997) found decreasing levels of DIN corresponding to cyanobacterial blooms. The similar temporal pattern of N-deficiency in both lakes also suggests that nitrogen deficiency was not a principal factor in the different composition of the autumn forming bloom species.

In 2000 both study lakes were observed to have a substantial increase in the PN:PP ratio during the autumn bloom, this was also observed in 2001 in Cusheon Lake. Unlike phosphorus, determining the source of nitrogen inputs to epilimnetic phytoplankton is more challenging because of N-fixation. An estimate of the nitrogen contributed to Cusheon Lake from N-fixation using September 2000 heterocyst biomass and a relationship between heterocyst biomass and N-fixation (Gondwe et al. 2008) suggests that approximately 60 μg N $L^{-1}$ $month^{-1}$ was added. However, total input from N-fixation generally increases with lake trophic level (Howarth et al. 1988), so this value may be underestimated since the relationship used (Gondwe et al. 2008) was derived from an ultraoligotrophic lake. The heterocyst biomass data, increase in TN and concomitant decrease in $\delta^{15}N$ values during autumn blooms in Cusheon Lake supports that N-fixation was an important source of N during this period. This agrees with MacGregor et al. (2001) who associated seasonal declines in $\delta^{15}N$ with N-fixation. Compared to Cusheon Lake, Elk Lake demonstrated greater decreases in $\delta^{15}N$ during the autumn. However, heterocyst counts in Elk Lake do not support widespread N-fixation in the pelagia nor was there an autumn increase in TN concentration.

The decrease of POM $\delta^{15}N$ in Elk Lake is of interest because it appears to largely be unrelated to N-fixation. In a

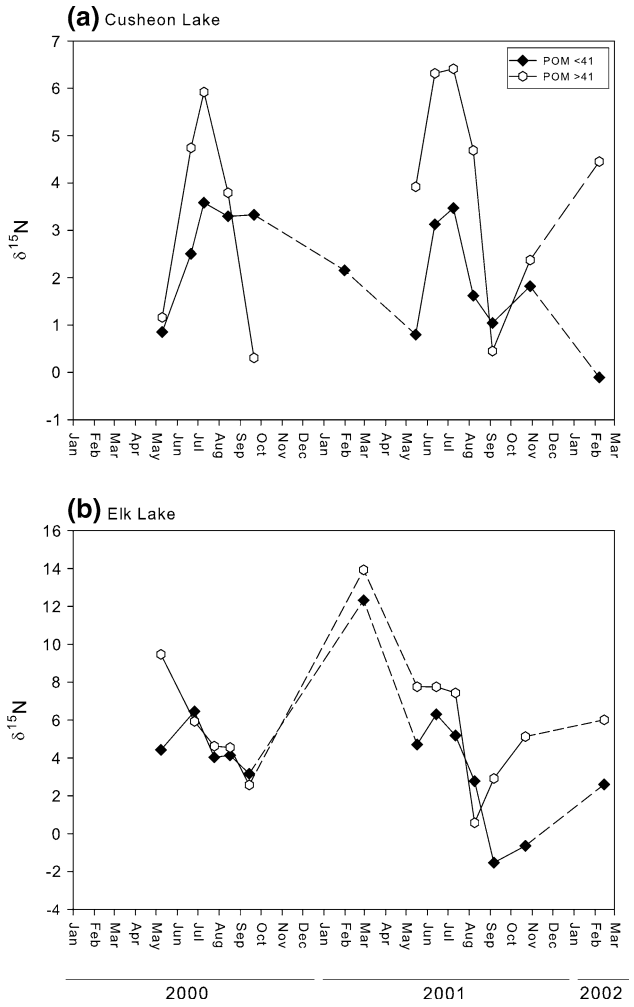


**Fig. 6** Temporal trends of $\delta^{15}$N in two size fractions for **a** Cusheon and **b** Elk Lakes

closed system, Rayleigh fractionation models can be used to detect times of the year when N-fixation may be an important source of pelagic nitrogen (see Fig. 10 in Lehmann et al. 2004). According to Rayleigh distillation kinetics, the $\delta^{15}$N of POM should decline with increasing DIN, leading to a linear relationship between POM $\delta^{15}$N and Log(DIN). Periods of N-fixation can be detected based on unexpectedly low POM $\delta^{15}$N for a given DIN concentration (Lehmann et al. 2004). In Elk Lake, the periods of low POM $\delta^{15}$N in the autumn, notably in 2001, correspond to increases in DIN. In 2001 there was a strong correlation between log DIN and POM $\delta^{15}$N at a lag of $-1$ ($r = 0.77$, $n = 6$). Unlike Cusheon there were no large corresponding increases in TN during the autumn and there were few heterocysts. Furthermore, the $\delta^{15}$N of the <41 μm fraction decreased below zero. Collectively this data suggests that change in $\delta^{15}$N was largely driven by different mechanisms in Cusheon and Elk Lakes.

PC:PN and PC:PP ratios represent the biomass normalized inverse of the Droop equation cell quota value (Droop 1974; Sommer 1991). These indicators are therefore related to limits on growth when N and P, respectively, are limiting. In contrast, low N:P cannot be used alone as an indicator of nitrogen limitation because P can be stored in plankton cells. Although the importance of nitrogen limitation as a precursor to dominance by N-fixing cyanobacteria has been argued (Horne 1979; Tilman et al. 1982), PC:PN data in Cusheon Lake did not indicate N-limitation prior to the cyanobacterial bloom. The decrease of $\delta^{15}$N associated with N-fixation during the autumn blooms of *A. flos-aquae* also did not correspond to conditions of either N-deficiency (bioassays) or N-limitation (PC:PN). The incongruence between PC:PN and N-deficiency assays during mid-summer in our P-limited lakes suggests that nutrient ratios will not always reflect short-term changes in nutrient deficiencies and are not always directly related to community growth. As argued above regarding the relationship between P-deficiency and the autumn bloom of *A. flos-aquae* in Cusheon Lake, N-deficiency measures should also provide greater insight into species competition and successional processes.

Many factors affect phytoplankton species composition and successional patterns (Sommer et al. 1986; Blomqvist et al. 1994) including nutrients (Tilman 1982), grazing (Brett et al. 1994), light (Flöder et al. 2002) and water column stability (Li 2002). Over longer periods, the potential attainable community biomass should therefore be set by the nutrient concentration that cannot be moderated within the ecosystem. However, individual species may respond to changes in nutrient supplies and forms of nutrients. Thus, as we observed with the deficiency measures in our study, it is expected that studies examining community level limitation should find co-limitation among species (e.g., Danger et al. 2008). We argue that nutrient assays provide a basis for better understanding species compositional changes because they provide a means of estimating cellular demand. Increased accessibility of field assessment methods such as in situ fluorescence should improve the application and scope of this approach (Beardall et al. 2001).

Some current nutrient limitation paradigms would benefit from greater critical analysis. Although they are fundamental to understanding nutrient limitation, many indicators are inappropriately applied and should be considered in the proper context, which includes scale dependency of respective measures, the inherent constraints of various measures, and the necessity of defining experimental scope and scale. Studies examining limitation over multiple years or more are often interested in understanding drivers of the ecosystem capacity. Measures of short-term nutrient limitation (deficiency) are focused on

physiological assessments. The ecosystem capacity and physiological scales both influence plankton population dynamics. Since it is frequently the plankton species that is of interest, as opposed to overall biomass, taking a balance approach to understanding nutrient limitation will serve to benefit both ecological and management purposes.

**Acknowledgments** We thank K. Walters, M. Deagle and I. Patchett for sampling assistance, K. Howell and K. Bannar-Martin for assistance with phytoplankton and heterocyst counts and Y. Liang for conducting nutrient analysis on the autoanalyzer. W.D. Taylor offered insightful comments on an early draft and two anonymous reviewers provided comments that substantially improved this paper. This research was supported by grants from NSERC/IRC and industrial partners (Capital Regional District of Victoria, Galloway Lumber Ltd. and Tembec Inc.) to A.M.

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