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Long-term Variation of Summer Phytoplankton Communities in an Urban Lake in Relation to Lake Management and Climate Conditions

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Long-term Variation of Summer Phytoplankton Communities in an Urban Lake in

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by

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Abstract

Eutrophication is one of the primary factors causing harmful cyanobacteria blooms in freshwater lakes; climate change such as warmer temperature can potentially further increase both frequency and intensity of blooms. This study investigated the long-term changes in water quality and summer phytoplankton assemblages in Oswego Lake, OR, in relation to lake management practices (e.g., hypolimnetic aeration and alum treatments), as well as climatic and regional meteorological conditions. Both water quality and phytoplankton assemblages were sampled biweekly during summer seasons between 2001 and 2013. The concentrations of total phosphorus (TP), soluble reactive phosphorus (SRP) and total nitrogen (TN) decreased 66%, 93% and 31%, respectively, in response to the hypolimnetic aeration and alum treatments since 2005. The results of summer phytoplankton assemblages showed a 62% reduction of cyanobacteria biovolume and a switch from cyanobacteria dominance (2001-2005) to diatom and chlorophyte dominance (2006-2013). Cluster analysis identified four statistically different groups of summer phytoplankton assemblages (denoted Groups 1-4). Nonmetric multidimensional scaling analysis indicated that the four groups were associated with different water quality conditions. Group 1 occurred prior to hypolimnetic aeration and was primarily comprised of cyanobacteria, associated with water conditions of high nutrients and high primary production. Group 2, dominated by cyanobacteria and chlorophytes, occurred between hypolimnetic aeration and alum surface application. Group 2 was associated with turbid water conditions. Group 3 was dominated by diatoms, occurring after alum surface application. Group 4 included R-strategist phytoplankton that quickly respond to environmental changes, occurring in the

years following alum injection, drawdown and inflow alum treatment. Both Group 3 and 4 were associated with reduced nutrients in the lake. The results demonstrated a strong temporal relationship between the long-term changes in water quality and summer phytoplankton assemblages and the lake management practices. The Pacific Decadal Oscillation (PDO) index, an El-Niño-like pattern of Pacific climate variability, showed a statistically significant correlation with the summer phytoplankton dynamics, while the multivariate ENSO index (MEI) and regional meteorological variables (air temperature, rainfall, wind speed, wind direction and solar radiation) were not significantly related to the changes of phytoplankton communities during the study period. In conclusion, the study results suggest that the lake management practices had strong effects on both production and community compositions of phytoplankton, and suggest the need for a future study on large-scale climate impacts on lake ecosystems and best management practice.

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1. Introduction

Urban lakes have a high societal value, providing valuable access to water for local residents. However, increasing human population and urban development have substantially accelerated the eutrophication process in urban waterbodies (Birch & McCaskie, 1999; J. W. Moore, Schindler, Scheuerell, Smith, & Frodge, 2003). Urban watershed runoffs from diverse sources, such as wastewater treatment plants, fertilized residential lawns and impervious road surfaces, introduce large amount of nutrients, sediments and other contaminants to urban lakes (Carpenter et al., 1998; Roy et al., 2008). Consequently, many urban lakes have experienced algal blooms and reduced water quality associated with noxious odors, depletion of dissolved oxygen, cyanobacteria toxins, impaired recreation, and degraded drinking water supplies (Dodds et al., 2009; Heinzmann & Chorus, 1994; Kotak et al., 1993; Oberholster, Botha, & Cloete, 2006; Qin et al., 2010).

Excessive nutrient input is one of the primary causes of lake eutrophication that results in algal blooms (Huisman & Hulot, 2005; Paerl et al., 2016; Schindler & Vallentyne, 2008). Both nutrients, nitrogen (N) and phosphorus (P), are essential elements for the growth of phytoplankton, and they are usually limiting nutrients in control of phytoplankton growth in lakes (Butusov & Jernelöv, 2013; Reynolds, 2006). Enrichment of N and P promotes the growth of phytoplankton in general (McCauley, Downing, & Watson, 1989). Particularly, in lakes in temperate regions, during the summer growing season, N and P inputs were found to be responsible for the shift in phytoplankton communities towards dominance by cyanobacteria (Downing, Watson, & McCauley, 2001; Watson, McCauley,

& Downing, 1997) and for the production of cyanobacteria toxins (Davis, Berry, Boyer, & Gobler, 2009; Rapala, Sivonen, Lyra, & Niemelä, 1997; Rolland, Bird, & Giani, 2005).

In order to control algal blooms due to eutrophication, the greatest efforts have been made to reduce nutrients available to phytoplankton production (Ibelings, Bormans, Fastner, & Visser, 2016; Paerl et al., 2016). Since certain cyanobacteria are able to fix atmospheric N, management strategy for reduction of P is more feasible than that of N (Elser et al., 2007; Lewis, Wurtsbaugh, & Paerl, 2011; Schindler et al., 2008). Watersheds are the ultimate source of P input to lakes, so reduction in supply of P in watersheds is a sustainable approach to prevent blooms (Hamilton, Salmaso, & Paerl, 2016). However, a great deal of evidences has shown that P can remain in high concentrations in lake water after significant reduction of P from watershed loading, due to continued release of excess legacy P from lake sediments (Søndergaard, Jensen, & Jeppesen, 2003; Van der Molen & Boers, 1994). Ideally, reduction of nutrients to control algal blooms should focus on P in both watersheds and lake sediments (Cooke, Welch, Peterson, & Nichols, 2005).

Control of algal blooms in urban lakes is a challenge (Birch & McCaskie, 1999; Huser, Futter, Lee, & Perniel, 2016). While reduction of nutrient loading can be partly achieved through watershed point-source permit management based on Total Maximum Daily Loads (TMDLs) according to the Clean Water Act (1972) (National Research Council, 1992), management of nutrient loading from non-point sources (NPS) is usually complex and difficult due to diverse NPS pollution that are transmitted via overland, underground and atmospheric paths and varies according to weather effects (Carpenter et al., 1998).

Effective NPS management is based on best management practices (BMPs), such as development and maintenance of wetlands, retention ponds and vegetation buffer strips (Paerl et al., 2016). However, for urban lakes, those strategies often face implementation challenges, such as fragmented responsibilities in a single watershed and resistance to change in developed urban areas (Roy et al., 2008). For example, Aguiar et al. (2015) found that effective riparian buffer zones were about 12-meters in width, with woody vegetation removing 43% of TN and 36% of TP, shrubs 41% and 32%, and grasses 21% and 17%, respectively. However, especially in developed urban areas, it becomes very costly to change urban settings in order to retroactively construct riparian/wetland buffer zones (Huser, Futter, et al., 2016; Roy et al., 2008). Thus, in-lake management is more practical and cost efficient compared to watershed nutrient management in urban lakes (Huser, Futter, et al., 2016).

In-lake nutrient management mostly targets bioavailable phosphorus resuspended from sediments to the water column (Bormans, Maršálek, & Jančula, 2016; Søndergaard et al., 2003; Van der Molen & Boers, 1994). Hypolimnetic aeration and aluminum sulfate (alum) treatment are both traditionally and commonly used approaches to reduce P in urban lakes (Bormans et al., 2016; Huser, Egemose, et al., 2016; Welch & Cooke, 1999). During periods of summer thermal stratification, hypolimnetic anoxia supports sediment phosphorus release to the water column (Søndergaard et al., 2003). Hypolimnetic aeration reduces hypolimnetic anoxia by increasing dissolved oxygen content of the hypolimnion without de-stratifying the lake (Soltero, Sexton, Ashley, & McKee, 1994). Alum treatment of lake water forms aluminum hydroxide floc that is highly coagulated to phosphorus. It not only precipitates phosphorus from the water column, but also adsorbs

and retains phosphorus from sediments when it settles down to the bottom, thus blocking internal phosphorus loading, which effect can last for about 13 to 15 years on average (Huser, Egemose, et al., 2016; Welch & Cooke, 1999). Alum floc has high adsorption capacity and can directly adsorb and precipitate particles, such as cyanobacteria cells, from the water column (Cooke et al., 2005). Combined with hypolimnetic aeration technology to maintain oxic conditions above the sediments, phosphorus can be immobilized by alum in the sediments and become unavailable to fuel blooms (B. C. Moore et al., 2012). Furthermore, in the case where the major tributary to the lake contributes inflow water carrying high phosphorus loading, alum directly applied to the inflow water can remove phosphorus prior to discharge to the lake (Heinzmann & Chorus, 1994; Pilgrim & Brezonik, 2005).

Climate change poses new challenges for the control of algal blooms (Jöhnk et al., 2008; Paerl et al., 2016). Climate warming, resulting in warmer water temperatures, enhances the strength and duration of thermal stratification and increases internal nutrient loading, favoring buoyancy regulated cyanobacteria with optimal growth at warmer temperatures (often > 25 °C) (Kosten et al., 2012; Rigosi, Carey, Ibelings, & Brookes, 2014; Robarts & Zohary, 1987). Furthermore, climate warming increases variability in reginal meteorological conditions, such as extreme rainfall and extensive droughts, which modifies watershed hydrologic dynamics in increasing external nutrient loading, further favoring bloom formation (Paerl et al., 2016; Rigosi et al., 2014). Climate warming has been widely recognized as an important factor in the increased frequency and intensity of cyanobacteria blooms, and therefore, has an impact on current bloom control strategies (Kosten et al., 2012; Magnuson et al., 1997; Paerl & Paul, 2012; Shimoda et al., 2011;

Winder & Sommer, 2012). For example, a physical bloom control approach, artificial mixing, had successfully controlled blooms of *Microcystis*, a cosmopolitan cyanobacterium of eutrophic freshwater, in Lake Nieuwe Meer, a recreational lake in the city of Amsterdam, since 1993 until an extreme summer heatwave occurred in Europe in 2003, the hottest summer recorded during the last century. During the experiment in Lake Nieuwe Meer in 2003, artificial mixing was switched on and off in order to reduce the energy costs of artificial mixing without inducing blooms. However, in August, at the peak of the summer heatwave, there were almost instant blooms of *Microcystis* as soon as artificial mixing was switched off, providing evidence that the heatwave, as a result of climate warming, promoted the bloom and caused the intermittent mixing strategies in control of the bloom to fail (Jöhnk et al., 2008). Intergovernmental Panel on Climate Change (IPCC, 2014) predicted that climate warming is unequivocal and is projected to rise 2.6-4.8 °C over the 21st century. Although the interaction effect between eutrophication and climate change on cyanobacteria blooms is still uncertain, the general understanding is that eutrophication and climate change act as two key factors favoring the blooms (Carey, Ibelings, Hoffmann, Hamilton, & Brookes, 2012; Kosten et al., 2012; Paerl & Huisman, 2008; Rigosi et al., 2014). Bloom management cannot be successful without properly addressing the challenges of both nutrients and climate conditions in favoring bloom occurrence.

Our understanding of phytoplankton communities in response to nutrient management practices and climate change is insufficient. Many studies have reviewed the efficacy of bloom control practices (e.g., hypolimnetic aeration and alum treatment), but most evaluations were based on a single approach and the short-term effects on nutrient

concentrations, chlorophyll *a* and/or total phytoplankton abundance (Huser, Futter, et al., 2016; Suikkanen, Laamanen, & Huttunen, 2007). Reports of integrated multiple practices are only available for a few lakes (B. C. Moore & Christensen, 2009; Soltero et al., 1994). The responses of phytoplankton communities were often reported briefly, probably due to lack of long-term, comparable data of taxonomic identifications for phytoplankton species (Suikkanen et al., 2007). Changes of phytoplankton compositions usually indicate shifts in environmental conditions (Reynolds, 2006). In perspective of the goal of nutrient management in control of blooms, the responses of phytoplankton communities to the changes of environmental conditions is more informative. In addition, previous studies have found the impacts of large-scale climatic events, such as El Niño Southern Oscillation (ENSO) and Pacific Decadal Oscillation (PDO), on phytoplankton community dynamics (Harris & Baxter, 1996; McGowan, Patoine, Graham, & Leavitt, 2005; Winder & Schindler, 2004; Winder & Sommer, 2012). But the significant climatic effects were related to phytoplankton spring communities and annual succession patterns, less focused on summer growing seasons. Furthermore, large-scale climatic events can modify reginal meteorological conditions that can possibly impact summer phytoplankton in long-term aspect (McGowan et al., 2005). Therefore, study of climate change focusing on summer phytoplankton communities is helpful to understand climate influence on summer cyanobacteria blooms and provides implications for bloom mitigation.

In this study, I characterized the changes in summer phytoplankton assemblages and their relationships with environmental variables in Oswego Lake, an urban lake in Oregon, USA, over a 13-year period (2001-2013). Specifically, I want to (1) characterize the

changes in summer phytoplankton assemblages in terms of dominant species and indicator species; (2) relate the changes of phytoplankton communities to water quality, lake management practices, large-scale climatic conditions, using the multivariate ENSO index (MEI) and PDO index, and regional meteorological conditions characterized by air temperature, rainfall, wind speed, wind direction and solar radiation. I therefore hypothesized that lake management practices, large-scale climatic conditions and regional meteorological conditions would have effects in driving the changes of phytoplankton communities, with nutrient-targeted management practices expected to have stronger influence on water quality and phytoplankton than climate and meteorological conditions. With the purpose of understanding the basic response mechanism of phytoplankton communities to lake management and climate change, this study may provide important information for future urban lake management to effectively predict, prevent and control harmful cyanobacteria blooms.

2. Background

2.1 Phytoplankton communities

Phytoplankton is the foundation of the aquatic food web, and phytoplankton communities are diverse and dynamic, spatially and temporally, in aquatic habitat (Hutchinson, 1961; Reynolds, 2006; Sommer, Gliwicz, Lampert, & Duncan, 1986). The dynamics of phytoplankton communities is the outcome of phytoplankton species competitions for light and nutrients, and of avoidance from grazing, determined by species-specific biological features and growth mechanics. For example, Sommer et al. (1986) used the Plankton Ecology Group (PEG) model to describe the annual succession pattern of phytoplankton composition and abundance due to seasonal changes of temperature, nutrient fluxes, light availability and grazing pressure. Furthermore, Reynolds (2006) and other ecologists developed the idea that the predominant community species are predictable in relation to their habitat characteristics because the most-favored species traits, survival strategies and community assemblages will occur (Grime, 1977; Keddy, 1992; Southwood, 1977). Changes of phytoplankton community compositions usually indicate shifts in environmental conditions (Reynolds, 2006).

2.2 Cyanobacteria blooms

Cyanobacteria are the oldest known fossil organisms, possibly dated back to 3.5 million years ago (Golubic & Seong-Joo, 1999). They are highly diverse photosynthetic bacteria in unicellular or multicellular forms, living in freshwater, ocean, and terrestrial environments (Whitton & Potts, 2007). Cyanobacteria are generally considered to beneficially contribute to photosynthesis and nitrogen fixation and they perform an important ecosystem function as primary producers in many food chains (Whitton & Potts, 2007). However, certain cyanobacterial genera, such as *Microcystis*, *Anabaena* (recently renamed *Dolichosermum*) and *Oscillatoria*, can produce toxins (cyanotoxins) that are harmful to human health, other organisms and the environment (Carmichael, 1997). Harmful cyanobacteria blooms are a severe problem with rapidly increasing worldwide concerns due to their negative impacts on water quality, human health and ecosystem as well as the complexity of their prevention and remediation (Dodds et al., 2009; Kosten et al., 2012; Paerl & Huisman, 2008).

Understanding the environmental factors causing cyanobacteria's successful growth and increasing presence in water bodies is essential to effectively predict, prevent and control cyanobacteria blooms. It is no surprise that a great number of research projects and studies have been conducted to identify the causative factors that contribute to cyanobacteria blooms. No single factor drives cyanobacteria blooms; in reality, it is due to the complex combination and interactions of physical, chemical and biological phenomena that favor the establishment of blooms (Kosten et al., 2012). In general, the

growth of cyanobacteria favors nutrient-rich, high water temperature, sufficient light, low CO_2/ high pH, and stratified water conditions (Figure 1).

The enrichment of nutrients, primarily nitrogen and phosphorus, in freshwater lakes is responsible for cyanobacteria blooms (Huisman & Hulot, 2005; Paerl et al., 2016). Nitrogen and phosphorus are both essential elements for growth of phytoplankton. Nitrogen is a major component of chlorophyll a and amino acids, while phosphorus is a fundamental building block of cell membranes, DNA and RNA, and ATP and ADP (Butusov & Jernelöv, 2013; Reynolds, 2006). They are both usually considered as limiting nutrients in control of phytoplankton growth in lakes (Reynolds, 2006). However, some cyanobacteria are able to fix atmospheric nitrogen and nitrogen fixation can be sufficient to support phytoplankton growth; thus, phytoplankton growth in lakes is more likely limited by phosphorus (Schindler et al., 2008).

Cyanobacteria growth is warm temperature-dependent. Paerl and Huisman (2008) used vivid language, "Blooms like it hot", to describe cyanobacteria metabolism requirement of warm temperature. Thus, blooms often form in summer in temperate freshwater lakes. Although different cyanobacteria species response to temperature in various degrees, higher growth rates are generally observed in higher water temperature conditions, until optimal temperature value is reached because each species has its own set of temperature range that it can tolerate. Robarts and Zohary's (1987) analysis showed that cyanobacteria has accelerating growth rate between 25 and 40 °C based on a large number of published field and laboratory data. Warmer water temperature is a favorable condition for growth of cyanobacteria in general.

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Cyanobacteria perform photosynthesis and utilize light energy to fix carbon dioxide to self-supply nutrients for life activities. Therefore, light availability is essential to the growth of cyanobacteria. Cyanobacteria have two mechanisms that enable them to effectively harvest light as a result of adaption to environment: pigmentation and buoyancy (Carey et al., 2012; Reynolds, 2006). Cyanobacteria contain photosynthetic pigments of chlorophyll *a* and phycobiliproteins. Chlorophyll *a* that is common in phytoplankton absorbs wavelengths of 400-450 nm and 650-700 nm. In addition to chlorophyll *a*, cyanobacteria have phycobiliproteins to absorb wavelength of 400-650 nm that is more prevalent in deeper water (Glazer, 1997), resulting in greater efficiency in light capturing for photosynthesis than other phytoplankton. Cyanobacteria also contain gas vesicles that allow cyanobacteria to regulate buoyancy in the vertical water column to uptake light in epilimnion and nutrients in hypolimnion during summer stratification (Reynolds, Oliver, & Walsby, 1987). The buoyancy-regulation mechanism benefits cyanobacteria in competition with other phytoplankton (Reynolds et al., 1987; Walsby, 1994).

The low CO_2/ high *p*H-hypothesis by King (1970) states that cyanobacteria are more efficient in obtaining carbon dioxide than phytoplankton. Photosynthetic activity makes strong demands on carbon dioxide. As photosynthesis increases, dissolved carbon dioxide in water decreases, resulting in increased water *p*H. Reduced supply of dissolved carbon dioxide can be a limiting factor in the growth of phytoplankton. However, cyanobacteria have evolved an effective photosynthetic CO_2 concentrating mechanism (CCM) that is perhaps the most effective CO_2-uptake mechanism in any photosynthetic organism (Badger & Price, 2003). Shapiro (1997) manipulated Squaw Lake, Wisconsin

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by injection of carbon dioxide for a study of the relationship between carbon dioxide and cyanobacteria. The result of this study suggests that the growth of cyanobacteria is not dependent on the condition of low CO_2/high pH, but rather that cyanobacteria are able to continually grow in conditions of low CO_2/high pH in which the growth of most phytoplankton is limited. The continuing growth of cyanobacteria extends the decreasing rate of dissolved carbon dioxide, and therefore maintain their dominance over other phytoplankton.

Summer vertical thermal stratification in freshwater system favors cyanobacteria. The stability of water is resistant to vertical mixing of epilimnion and hypolimnion, and this resistance is due to water density gradients that are induced by water temperature (Wetzel, 2001). Cyanobacteria are able to regulate buoyancy in the water column, which allows them to obtain light in epilimnion and nutrient in hypolimnion. Cyanobacteria blooms form thick layer scums at the surface of water as a canopy, attenuating light penetration into the water, which suppresses non-buoyant phytoplankton and enhances their domination in the water system (Paerl & Huisman, 2008). However, artificial mixing, as an example, increases turbulence in water and reduces water stability, which results in cyanobacteria losing their advantage of buoyancy (Visser, Ibelings, Bormans, & Huisman, 2016).

Cyanobacteria abundance is directly regulated by aquatic food web structure (Shapiro, Lamarra, & Lynch, 1975). Phytoplankton is a primary producer, while zooplankton and phytoplanktivorous fish are primary consumers of phytoplankton (Reynolds, 2006). However, planktivorous fish, feeding on zooplankton, usually allow opportunity for the

growth of phytoplankton; on the contrary, piscivorous fish, feeding on planktivorous fish, increase grazing pressure of zooplankton on phytoplankton (Reynolds, 2006). This trophic structure has been well studied theoretically, such as in the Trophic Cascade Model (Carpenter, Kitchell, & Hodgson, 1985) and the Top-down or Bottom-up Theory (Horppila, Peltonen, Malinen, Luokkanen, & Kairesalo, 1998), and practically in biomanipulation management on control of cyanobacteria blooms (Triest, Stiers, & Onsem, 2016). Interestingly, cyanobacteria can produce mucilage and/or toxin, and form large colonies or filaments in resistance to grazing. For instance, Drenner et al. (2002) reported that although the stocking of piscivorous fish increased zooplankton abundance and decreased phytoplankton density, the phytoplankton dominant community shifted to inedible cyanobacteria.

Watershed land use types have been acknowledged as a strong factor to freshwater phytoplankton structure (Katsiapi, Mazaris, Charalampous, & Moustaka-Gouni, 2012; Paul et al., 2012). Previous studies suggest that urban lakes are generally more prone to have cyanobacteria blooms than non-urban lakes (Katsiapi et al., 2012; Schueler & Simpson, 2001). Urban lakes tend to be rather small and shallow with large watershed to lake area ratio, indicating that their watersheds exert a strong influence on the lakes (Schueler & Simpson, 2001). Urban watersheds are often areas draining water from wastewater treatment plants, fertilizers from golf course and residential lawns, and storm run-off from impervious cover into the lake, introducing great amount of nutrients, sediments and other contaminants to the lakes (Carpenter et al., 1998; Roy et al., 2008). External loading leads to increasing sedimentation of nutrients and organic materials in

lakes and subsequently reinforcing internal cycling of nutrients, fueling cyanobacteria blooms (Søndergaard et al., 2003).

Climate warming has the potential to directly affect the physical, chemical, and biological characteristics of lakes, and to indirectly affect lakes through its influences on watershed ecosystem. "Blooms like it hot" (Paerl & Huisman, 2008) suggests that cyanobacteria eventually benefit from the consequences of global warming. Temperature increase driven by global warming extends the period of stratification, which is a favorable condition for cyanobacteria growth (Livingstone, 2003; Livingstone & Dokuli, 2001; Magnuson et al., 1997; Shimoda et al., 2011; Winder & Schindler, 2004). Severe occurrences of precipitation and drought driven by climate change also lead to favorable condition for cyanobacteria growth (Carey et al., 2012; Paerl & Huisman, 2008; Reichwaldt & Ghadouani, 2012; Settele et al., 2014). For example, heavy storms increase the turbidity of a water body by causing suspension of sediment in the water and flushing ambient sediment into the water. This condition disturbs cyanobacteria communities in the short-term, however, favor cyanobacteria growth in the long-term (Reichwaldt & Ghadouani, 2012).

2.3 Effects of management and climate change on phytoplankton in urban ecosystems

2.3.1 Approaches for managing cyanobacteria blooms

There are a number of methods for preventing, controlling and mitigating cyanobacteria blooms, and many case studies of the success or failure of these methods have been well documented (Ibelings et al., 2016; Paerl et al., 2016) (Figure 2). Bloom management is usually at first based on limiting the availability of nutrients for cyanobacteria development (Cooke et al., 2005; McCauley et al., 1989; Schindler & Vallentyne, 2008), because the enrichment of nutrients, primarily nitrogen (N) and phosphorus (P), in freshwater lakes is responsible for cyanobacteria blooms (Huisman & Hulot, 2005; Paerl et al., 2016). Both N and P are usually considered to be limiting nutrients in control of phytoplankton growth in lakes (Reynolds, 2006). However, some cyanobacteria are able to fix atmospheric N; thus, phytoplankton growth in lakes is more likely limited by P (Schindler et al., 2008). The 16-year long experiment of a whole-lake nutrient enrichment by Schindler et al. (2008) demonstrated that P was the limiting nutrient alone and N reduction induced the growth of N-fixing cyanobacteria. Although other studies showed that N reduction in control of blooms was equally important due to consideration of downstream eutrophication in N sensitive estuarine and coastal waters (Elser et al., 2007; Lewis et al., 2011), nutrient management in lakes to control blooms is usually focused on P first.

A number of studies have shown that P remained at high concentrations in the water column after significant reductions of P from watershed loading, which was due to

continued release of excess legacy P from lake sediments (Søndergaard et al., 2003; Van der Molen & Boers, 1994). Internal recycling of phosphorus plays an important role in natural processes of lakes, because phosphorus is an essential nutrient to organisms and has been recognized as the least abundant element in comparison to other major nutrients, such as carbon, hydrogen, nitrogen, oxygen, and sulfur (Wetzel, 2001). Exchange of phosphorus between sediments and the overlying water continuously occurs in lakes. Particulate phosphorus in water slowly sinks to the bottom, while dissolved phosphorus is absorbed quickly by bacteria, algae and plants in the water column and eventually deposits as organic detritus to the sediments. Simultaneously, phosphorus in sediments returns to the water under an array of physical, chemical and biological processes. This release of phosphorus from sediments is generally called internal loading to be distinguished from external loading. Dissolved phosphorus diffuses across the sediment-water interface physically due to concentration gradients. Also, the movement of benthic invertebrates and fish results in disturbance of sediments, which generates resuspension of phosphorus particulate to the water. Similarly, wind-induced turbulence, typically in shallow lakes, can cause phosphorus resuspension as well. During summer in deep stratified lakes, phosphorus can release from sediments to the water as a result of redox reactions under anoxic condition, in which iron (III) reduce to iron (II) and release bonded phosphate to soluble forms. Some phytoplankton species contribute phosphorus recycling when they migrate from their resting stage in the sediment to life stage in the water (Pettersson, 1998). More detailed processes of internal phosphorus loading are illustrated by Wetzel (2001). Among various mechanisms of internal phosphorus loading, chemical equilibria process contributes the most significant release in comparison to other processes (Wetzel, 2001). Among various lake management

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practices in control of internal phosphorus loading, applications of alum and

hypolimnetic aeration have been found the effective methods in precipitation and

inactivation of phosphate in sediments.

2.3.1.1 Alum

Alum represents a group of aluminum salts. Alum commonly used as a clarifying agent

(coagulation-flocculation reactant) of water is aluminum sulfate ($Al_2(SO_4)_3$) (Cooke et

al., 2005). Alum is soluble in water. When alum is applied to water, the aluminum ion

dissociates in water and undergoes a series of hydrolysis reactions. Aluminum hydrolysis

reactions are pH dependent. Large colloidal floc of aluminum hydroxide ($Al(OH)_3$) is the

predominant product between pH 6 and 8, while soluble $Al(OH)_2^+$ and Al^{3+} occur at

lower pH and soluble $Al(OH)_4^-$ occurs at higher pH (Cooke et al., 2005). Thus, the

reactions between alum and phosphorus are also pH dependent. At low pH, such as

sewage water, phosphorus is removed from the water column primarily by precipitation

of $AlPO_4$. But in lakes, usually with higher pH, phosphorus and particles are primarily

removed by coagulation and flocculation process of formation of aluminum hydroxide

floc (Cooke et al., 2005). It is very important to determine the optimal dosage of alum in

treatment because excessive additions of alum will decrease pH and increase soluble

aluminum forms, including $Al(OH)_2^+$ and Al^{3+}, producing toxic effects to aquatic systems

(Gensemer & Playle, 1999). The treatment dose can be estimated based on pH and

alkalinity of the lake water and mobile phosphorus (iron bonded phosphorus and loosely

attached phosphorus under anoxic condition) in the sediment (Cooke et al., 2005). In

order to buffer pH in alum treatment, sodium aluminate ($NaAlO_2$) is usually added with

alum content. The buffered treatment can maintain a pH between 6 and 8 and enhance

phosphorus removal (Cooke et al., 2005).

Alum surface application was the first (Jernelov, 1970) and most commonly used method

(Schütz, 2016), which allows aluminum flocs to precipitate phosphorous through the

entire water column and to inactivate phosphorus in the sediments by forming a barrier to

the release of sediment phosphorus. Aluminum hydroxide floc is highly coagulated to

phosphorus. It not only precipitates phosphorus from the water column, but also adsorbs

and retains phosphorus from sediments when it settles to the bottom, thus blocking

internal phosphorus loading, with the effect lasting for more than 10 years (Cooke et al.,

2005). Alum floc has high adsorption capacity and can directly adsorb and precipitate

particles, such as cyanobacteria cells from the water column (Cooke et al., 2005). Later,

the alum hypolimnetic injection method was developed to directly target sediments, the

source of internal phosphorus loading, and lower the adverse effects on aquatic

organisms (Cooke et al., 2005). One-time alum treatment applied to whole lake was

usually employed with a large dose in order to apply adequate aluminum to bond

phosphorus in both water and sediments (Cooke et al., 2005). However, the effectiveness

of phosphorus inactivation is reduced not only by decreasing adsorption capacity of

aluminum floc, but also by burial of active Al-P layer due to new sedimentation and

sediment mixing process (Cooke et al., 2005; Lewandowski, Schauser, & Hupfer, 2003).

Successive low-dose application has been developed for the purpose of maintaining

aluminum floc for high effectiveness (Cooke et al., 2005; Lewandowski et al., 2003; B.

C. Moore, Christensen, & Richter, 2009). In the case where the major tributary to the

lake contributes the inflow water carrying high phosphorus loading, alum directly applied

to the inflow water can remove phosphorus prior to discharge to the lake (Heinzmann & Chorus, 1994; Pilgrim & Brezonik, 2005).

Alum treatment has been commonly used to reduce internal phosphorus loading in lakes of the United States and other countries (Huser, Egemose, et al., 2016; Welch & Cooke, 1999). The review by Welch and Cooke (1999) on alum effectiveness showed that total phosphorus was reduced by an average of 37% (ranged from 13% to 65%) and chlorophyll *a* decreased by an average of 57% (ranged from 28% to 75%), as results of alum treatments to 7 out of 7 stratified lakes. Huser et al. (2016) further analyzed 114 lakes treated with alum and reported that total phosphorus was reduced by an average of 64% and chlorophyll *a* decreased by an average of 62%. Both studies suggest that alum treatment overall is effective in controlling in-lake phosphorus and improvement of water quality based on reduction of chlorophyll *a* concentration.

2.3.1.2 Hypolimnetic aeration

Hypolimnetic anoxia supports sediment phosphorus release to the water column. Hypolimnetic aeration reduces hypolimnetic anoxia by increasing dissolved oxygen content of hypolimnion without de-stratifying the lake (Cooke et al., 2005; Soltero et al., 1994). Hypolimnetic aeration can be accomplished by injection of oxygen or air into hypolimnion. The air-lift aerators, especially the partial air-lift aerators, were commonly used (Soltero et al., 1994). Hypolimnetic aerators receive compressed air at the lake bottom, transport the bottom water up by rising air bubbles and increase dissolved oxygen content in water through the gradient of dissolved oxygen, then return the aerated

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water to the lake bottom and release excess air to the lake surface (Cooke et al., 2005; Soltero et al., 1994).

Hypolimnetic aeration has been used for a number of lakes and reservoirs to reduce blooms and improve water quality. Cooke et al. (2005) reviewed 28 cases and found that hypolimnetic aeration resulted in an increase of hypolimnetic dissolved oxygen (DO) in most cases, often to at least 7 mg DO/L. For example, in Newman Lake, alum addition along with hypolimnetic aeration had improved control of internal P loading (Moore et al., 2012). However, Cooke et al. (2005) found that hypolimnion aeration was not as effective as alum in the reduction of hypolimnetic phosphorus, which might be due to lack of P-binding agents, such as Fe, Mn or Al.

2.3.2 Effects of climate change on phytoplankton

Climate warming has the potential to directly affect physical, chemical, and biological characteristics of lakes, and to indirectly affect lakes through its influences on the watershed ecosystem (Kosten et al., 2012; Paerl et al., 2016; Rigosi et al., 2014; Robarts & Zohary, 1987). Intergovernmental Panel on Climate Change (IPCC) defines climate change as a testable change of climate over an extended period, typically decades or longer, due to natural variability and/or human activity (IPCC, 2007). According to IPCC AR5 (IPCC, 2014), warming of the climate system is unequivocal and a changing climate is having widespread impacts on human and natural systems. Lake surface water temperature is closely correlated with air temperature. Previous studies have showed that warmer air temperatures resulted in warmer epilimnetic temperature, longer duration of

summer stratification, stronger stability of thermal stratification, and deeper thermocline

depth (Livingstone, 2003; Livingstone & Dokuli, 2001; Magnuson et al., 1997; Shimoda

et al., 2011; Winder & Schindler, 2004). Climate effects on nutrients are complex

because climate has effects on physical, chemical and biological processes of nutrient

cycles. For example, prolonged and stable stratification will result in hypolimnetic

anoxia that increase internal phosphorus loading (Carey et al., 2012; Settele et al., 2014).

At the catchment level, warming stimulates rock weathering and soil erosion, and

intensified precipitation due to warming increases runoff of sediments and nutrients to the

receiving lakes (Carey et al., 2012; Paerl & Huisman, 2008; Reichwaldt & Ghadouani,

2012). Climate change, characterized by conditions such as rising temperature and

changing precipitation patterns, causing extreme weather conditions like storms and

droughts, modifies the abiotic and biotic environment favorable to the growth of

cyanobacteria (Reichwaldt & Ghadouani, 2012). Bloom-forming cyanobacteria contain

gas vacuoles that allow cyanobacteria to regulate buoyancy in the vertical water column

to uptake light in epilimnion and nutrients in hypolimnion during summer stratification

(Carey et al., 2012). Cyanobacteria also have greater growth rate than other

phytoplankton in warmer water temperature conditions (Paerl & Huisman, 2008).

Increasing water temperature results not only in boosting the growth of cyanobacteria but

also increasing strength and duration of summer stratification, thus, enhancing bloom

formation (Carey et al., 2012; Elliott, 2010; Jöhnk et al., 2008; Settele et al., 2014).

The El Niño Southern Oscillation (ENSO) is the most important large-scale hydro-

meteorological phenomenon that contributes to extreme climatic events on interannual

time scales (Wolter, 1989). Multivariate ENSO index (MEI) is a method used to

characterize ENSO events in the first principal component of six observed variables over

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the tropical Pacific Ocean (sea level pressure, surface zonal and meridional wind components, sea surface temperature, surface air temperature, and cloudiness), based on long-term marine records from the Comprehensive Ocean-Atmosphere Data Set (COADS) (Wolter & Timlin, 1993, 1998). It is computed every month, averaging monthly means into bimonthly seasons (Dec/Jan, Jan/Feb, … Nov/Dec). The positive values of MEI represent the warm ENSO phase (El Niño), while the negative values of MEI represent the cold ENSO phase (La Niña). According to Mazzarella et al. (2010), MEI was the most representative meteorological index for ENSO compared to other indices. The effects of ENSO on lake ecosystems have been observed in North America (Harris & Baxter, 1996; McGowan et al., 2005; Winder & Schindler, 2004). For example, McGowan et al.'s (2005) study of six lakes in Saskatchewan, Canada suggests that the maximum summer phytoplankton abundance in 1997 was consistent with the very strong 1997 El Niño (Wolter & Timlin, 1998).

The Pacific Decadal Oscillation (PDO) is another large-scale climate phenomenon that has an impact on the surface air temperature and precipitation of western North America (Mantua, Hare, Zhang, Wallace, & Francis, 1997). The PDO has been described as an ENSO-like pattern of Pacific climatic fluctuation, but distinguished from ENSO by 1) the persistence for more than two decades during the 20th century while typical ENSO events persist for 6 to 18 months; 2) the influences particularly in the North Pacific while ENSO's influences are in tropics; and 3) unclear mechanisms casing PDO while causes for ENSO variability were well-known (Mantua & Hare, 2002). The PDO index is defined as the leading principal component of North Pacific monthly sea surface temperature variability (Mantua & Hare, 2002). Positive PDO index indicates warm

phase, while negative PDO index indicates cool phase. The effects of PDO on the

Northwest North America is that the conditions of warm temperature and low

precipitation are associated with the warm PDO phase, while the opposite conditions are

associated with the cool PDO phases (Mantua & Hare, 2002). The PDO has been

associated with ecological variabilities, such as salmon production (Mantua et al., 1997),

lake ice cover (Bonsal, Prowse, Duguay, & Lacroix, 2006), lake water level (Peterson,

Silsbee, & Redmond, 1999), and plankton community dynamics (Winder & Schindler,

2004; Winder & Sommer, 2012). For example, the long-term study (a period of 40 years

from 1962 to 2002) of climatic effects on phytoplankton pattern in Lake Washington

(about 300 km north of Oswego Lake) showed significant effects of large-scale

meteorological phenomena, including PDO and ENSO, on the phytoplankton spring

blooms through its effect of warming on the extended duration of lake stratification

(Winder & Schindler, 2004; Winder & Sommer, 2012).

3. Methods

3.1 Study site

Oswego Lake (45°24′34″N, 122°41′47″W) is located in northwest Oregon, approximately 13 km south of the city of Portland. The lake, a former channel of the Tualatin River, was formed by the Missoula Floods of the last ice age, rich in flood sediments that were deposited in the lake bed (Foster, 2009). The lake area was rich in iron ore, and ore mining and smelting was the central industry in the lake town in the mid-1800s. The lake was dammed for power generation and the area surrounding the lake was developed as a residential district in the 1900s. Now the lake consists of one deep basin, two shallow basins and two canals (Figure 3). It has a maximum depth of 16.7 m and a total area of 1.7 km^2 (Table 1). The lake's flow system includes one outflow, three inflows, and almost 70 storm water outfalls in its watershed. The lake has a watershed area of 18.6 km^2 entirely within the limits of the city of Lake Oswego. The watershed is mostly comprised of urban areas (68%) and forest cover (19%). The climate in the region is characterized by wet, mild winters and dry, warm summers, directly affected by the Pacific Ocean (Franczyk & Chang, 2009). Between 2001 and 2013, the average monthly temperatures ranged from a low of 4.2 °C in December to a high of 20.2 °C in July, and the average annual precipitation was 962.1 mm. More than 70% of the annual precipitation was during October through March, with less than 10% falling between July and September.

Oswego Lake was previously a hypereutrophic lake and had a long history of cyanobacteria blooms (D. M. Johnson, 1985). Since the 1950's the full water right of 1.6 $m^3\ s^{-1}$ had been withdrawn from the Tualatin River 24 hours a day, year-round to the lake for power generation. Although this practice was reduced since the mid 1990's, the inflow of nutrient-rich Tualatin River water resulted in bringing in large loads of phosphorus-rich sediments into the lake (ODEQ, 2001). For example, from November 1986 to December 1987, 38.9 million cubic meters of water were imported from the Tualatin River, with a loading of 487,000 kg sediment and 14,000 kg phosphorus (SRI, 1987). Oswego Lake is also the destination for the surface run-off from the local watershed managed by the City of Lake Oswego. Additionally, in 1996, the lake experienced a major flood that contributed a great deal of sediment from the Tualatin River. As a result, there was a large reservoir of phosphorus-rich sediment in the lake, which caused significant summer algae growth. A *Microcystis* bloom during late summer 2004 necessitated restricting lake use to non-contact activities because of the potential for illness due to the presence of microcystin toxin.

A number of management practices have been employed in Oswego Lake to reduce and prevent cyanobacteria blooms. Prior to 2001, copper sulfate was used to the lake to reduce the algae concentration. This practice was stopped in 2001 because of the possible adverse effects on the aquatic environment. Between 2001 and 2013, the primary management effort was focused on reduction of in-lake phosphorus to control blooms, which included hypolimnetic aeration, alum surface application, alum injection, inflow water volume reduction and inflow alum treatment. In addition, two water level drawdowns dropped the water level 3 meters in 2006 for 138 days and 7 meters in 2010

for 264 days. Although the purpose of these drawdowns was lake and facility

maintenance, the consequence of water level fluctuations can be important in affecting

the lake ecosystem (Bakker & Hilt, 2016; Pan et al., 2018). Table 2 includes more

detailed information about the in-lake management practices implemented in Oswego

Lake between 2001 and 2013. In addition, current watershed management in place in

order to reduce nutrient loading to the lake, besides the Tualatin River inflow control,

includes several storm water best management practices (e.g., wet retention ponds, dry

detention ponds, swales, infiltration rain gardens, underground injection control systems,

and lined planter rain gardens), and bank stabilization through promotion of native

vegetation and removal of invasive species along stream banks.

3.2 Sampling and measurement

Physical, chemical and biological parameters were measured weekly or biweekly at eight

sampling sites in Oswego Lake for long-term water quality monitoring. The sampling

sites, corresponding with the various aspects of lake morphology, include one site at each

of two shallow basins, the outlet and two canals, and three sites at the main lake basin.

Two sampling sites between July and September from 2001 to 2013 were chosen for this

study, including the main lake center sampling site (Appendix A), where maximum depth

is more than 15 meters and captures summer thermal stratification, and the Oswego Canal

sampling site, which reflects the Tualatin River inflows (Figure 3).

At the lake center, water temperature, pH, percent saturation of dissolved oxygen, and

specific conductivity were recorded at every meter from 0.38 m to 15 m below the

surface using YSI 6600 multiparameter water quality sonde. The values of epilimnetic

water quality parameters were generated by averaging the YSI sonde measurements at

depths between 1 and 5 meters. Turbidity, using grab sample at elbow depth, was

measured using the Hatch 2100Q Turbidimeter.

Water samples for total suspended solids (TSS), nutrients, chlorophyll *a* and

phytoplankton were collected using Kemmerer sampler from specific depths. The

epilimnion samples were the equal-volume composite of water samples taken at discrete

depths of 1, 2, 3, 4, and 5 meters. The hypolimnion samples were the equal-volume

composite of water samples taken at discrete depths of 10, 12, and 14 meters. The canal

samples were collected by placing Kemmerer sampler in the middle of the water column

usually about 1 meter deep.

Epilimnetic TSS samples were stored in capped bottles, kept in a cooler in the field and

in the refrigerator in the Lake Oswego Corporation (LOC) laboratory until the time of

analysis. TSS was measured by filtering 1-liter well-mixed water sample through a

weighted standard glass-fiber filter (Whatman 934-AH filter, 47 mm), and by measuring

the weight of the residue retained on the filter that was dried to a constant weight at

103 °C in the oven (APHA, 2012) (Appendix B1). The difference in weight of the filter

represents the TSS.

Nutrients were sampled for total phosphorus (TP), soluble reactive phosphorus (SRP),

and total nitrogen (TN). Samples were stored in new bottles, kept in a dark cooler in the

field, and shipped overnight with ice packs to Aquatic Research, Inc. (ARI) in Seattle,

Washington. ARI received the samples the next day and analyzed the samples based on the ascorbic acid method to determine TP and SRP (detection limits of 0.002 mg/L for TP and 0.001 mg/L for SRP), and the persulfate digestion method to determine TN (detection limit of 0.100 mg/L) (APHA, 2012; Godomski, 2013).

Chlorophyll *a* samples were obtained by filtering 300-500 mL of sampling water with the addition of 1 mL of saturated magnesium carbonate (MgCO$_3$) solution through one piece of 0.45 µm glass fiber filter. The filter was folded, stored in a capped centrifuge tube, kept in a dark cooler in the field, and stored in a dark freezer in the LOC laboratory until the time of analysis. Chlorophyll *a* measurement was processed in subdued light to avoid degradation. Chlorophyll *a* was extracted by centrifuging the sample with the addition of 90% aqueous acetone. Then the optical densities of the 90% acetone extracts and the acidified extracts with addition of 0.1 mL 0.1*N* hydrochloric acid (HCL) were determined with a spectrophotometer (APHA, 2012) (Appendix B2). The concentration of chlorophyll *a* was calculated as follows:

$$Chlorophyll\ a\ (ug/L) = \frac{26.7\ \times\ [(664b - 750b) - (665a - 750a)] \times\ 19.5}{V \div\ 1000}$$

Where:

   26.7 = absorbance correction constant

   19.5 = volume of extract (mL)

   V = volume of sample (mL)

   664*b*, 750*b*, 665*a*, 750*a* = optical densities of 90% acetone extracts before and
                              after acidification

Phytoplankton samples (500 mL each) were stored in bottles, preserved with 3 mL Lugol's solution per sample, kept in a dark cooler in the field, and shipped overnight with ice packs to WATER Environmental Services, Inc. (WES) in Seattle, Washington. WES determined phytoplankton cell volume dimensions and identifications using a 0.1 mL Palmer-Maloney nanoplankton chamber with a calibrated whipple disc under Leitz compound microscope at 400 X high magnification or at 1000 X magnification with oil immersion. Geometric cell dimensions of at least 10 organisms of each phytoplankton were computed to obtain average cell volume per taxon and used to calculate cell volumes for the lake sample. Phytoplankton identification was made to species level where possible. Species identifications were conducted primarily according to Prescott (1975, 1980), Patrick and Reimer (1966, 1975), Smith (1950), Wehr and Sheath (2003), and recent journal article sources (M. Gibbons, 2013) (Appendix B3).

3.3 Data description

3.3.1 Phytoplankton data

The phytoplankton taxon biovolumes were converted to the percentage of total biovolume of each sample in order to characterize phytoplankton assemblages. The division dataset was created based on the untrimmed phytoplankton species dataset that included 129 taxa (Appendix D). The species dataset was created by removing the rare species that occurred < 3 samples among all samples to reduce noise for further multivariate analysis (Poos & Jackson, 2012), which resulted in a total of 50 taxa (Appendix D).

Since cyanobacteria, diatoms and chlorophytes, accounting for more than 80% of total phytoplankton biovolume, dominated the phytoplankton assemblages over 13 years, the data analysis of divisions was focused on these three largest divisions (Appendix E). Further, cyanobacteria were divided into two groups, nitrogen-fixers and non-nitrogen-fixers, because the occurrence of nitrogen-fixing cyanobacteria may suggest nitrogen limitation in environmental conditions (Schindler, 1977).

3.3.2 Environmental data

In addition to water quality, climate and weather conditions were used to identify the environmental impact on the changes of phytoplankton assemblages. Large-scale climatic effects were characterized by using the multivariate ENSO index (MEI) obtained

from the NOAA (https://www.esrl.noaa.gov/psd/enso/mei/table.html) and the Pacific

Decadal Oscillation (PDO) index obtained from the Joint Institute for the Study of the

atmosphere and Oceans, University of Washington, Washington

(http://research.jisao.washington.edu/pdo/PDO.latest). The MEI ranks (obtained from

https://www.esrl.noaa.gov/psd/enso/mei/rank.html, updated on 6 April 2018) were used

because the MEI ranks are useful to interpret ENSO strengths that are defined by a 1-69

scale. The lowest number (1) indicates the strongest La Niña case while the highest

number (69) indicates the strongest El Niño case. MEI ranks from 1-14-21 denote

strong-moderate-weak La Niña conditions, while 49-56-69 denote weak-moderate-strong

El Niño conditions, respectively. The local weather conditions were characterized by

monthly means of air temperature, wind speed, wind direction and solar radiation on

daily data between 5am and 9pm (summer day time), and monthly sum of rainfall on 24-

hour daily data. The monthly averages of wind speed and wind direction were calculated

using R function 'timeAverage' in package 'openair' to treat wind direction correctly

through vector-averaging (Carslaw & Ropkins, 2018; Grange, 2014). The weather data

were obtained from the weather station on the roof of the LOC's office next to the outlet

of the lake (Figure 3).

3.4 Data analysis

To describe the long-term changes in phytoplankton assemblages, a combination of
hierarchical agglomerative cluster method with Ward's minimum variance method (Ward
Jr., 1963) was used to identify relatively homogeneous groups of phytoplankton
assemblages using the Bray-Curtis dissimilarity index (Bray & Curtis, 1957). Analysis of
Similarity (ANOSIM) was used to test statistical significance of the differences among
the cluster groups (Clarke, 1993). Dominant taxa and indicator taxa were further used to
characterize the cluster groups of similar phytoplankton assemblages. Dominant taxa
were defined as most abundant taxa that account for more than 50% of total
phytoplankton biovolume within the group. Indicator taxa were defined as the taxa,
belonging to cyanobacteria, diatoms and chlorophytes, with an indicator species
index >0.5 (p<0.05) based on Dufrene-Legendre indicator species analysis (Dufrene &
Legendre, 1997). P values of indicator species for all taxa were determined using Monte
Carlo permutation tests (1000 times).

Further, nonmetric multidimensional scaling (NMDS) based on Bray-Curtis dissimilarity
matrix was used to visualize the changes among the groups of phytoplankton
assemblages over time. NMDS is a multivariate ordination technique commonly used in
ecological community analysis (Clarke, 1993). NMDS projects each sample into a
species-defined ordination space with two or more dimensions based on their ranked
dissimilarity. The goodness-of-fit for the NMDS projections was measured as a stress
value which quantifies the deviation from a monotonic relationship between the distance
among samples in the original Bray-Curtis dissimilarity matrix and the distance among

samples in the ordination plot. The NMDS was run 20 times each with a random starting configuration. The final NMDS dimension was selected based on the lowest stress value among the best solutions.

The changes of phytoplankton assemblages were expected to associate with environmental factors. In order to assess this relationship, the environmental variables were related to the NMDS ordination space defined by the species data using "envfit" function in R (Oksanen et al., 2013). The importance of each environmental variable was assessed using a squared correlation coefficient (r^2). Furthermore, the lake management practices were related to the cluster groups of phytoplankton assemblages in a timeline manner for assessing the management effects on the dynamics of phytoplankton communities.

Data analysis was performed using R (R Development Core Team, 2014). Specifically, the packages 'vegan' and 'MASS' were used for the cluster and NMDS analysis, the package 'labdsv' for the indicator species analysis, and the package 'timevis' for the time relationship between lake management practices and the cluster groups of phytoplankton assemblages.

4. Results

4.1 Changes of environmental conditions over 13 years

During the study period, water in the lake epilimnion in general had moderate nutrients (mean TP = 39 µg L^{-1}; mean SRP = 3 µg L^{-1}; mean TN = 460 µg L^{-1}), low specific conductivity (mean = 161 µS cm^{-1}), alkaline pH (mean = 8.5) and moderate primary productivity (mean chlorophyll a = 16 µg L^{-1}) (Figure 4 A & D).

Concentrations of nutrients changed over 13 years. The greatest changes occurred in epilimnetic TP and SRP in 2005 when the alum surface treatment was applied (Figure 4A). Mean epilimnetic TP declined from 72 µg L^{-1} (2001-2004) to 25 µg L^{-1} (2005-2013), a 66% reduction, while mean epilimnetic SRP declined from 8 µg L^{-1} to 0.6 µg L^{-1}, a 93% reduction. Mean epilimnetic TN gradually declined from 585 µg L^{-1} to 403 µg L^{-1}, a 31% reduction. In contrast, mean epilimnetic TN:TP ratio increased from 9 to 18. Meanwhile, the most visible changes of hypolimnetic nutrients occurred in 2001 when hypolimnetic aeration began (Figure 4B). Mean hypolimnetic TP declined from 292 µg L^{-1} (2001) to 69 µg L^{-1} (2002-2013), a 77% reduction, with mean hypolimnetic SRP decreasing from 62 µg L^{-1} to 14 µg L^{-1} (77%) and mean hypolimnetic TN decreasing from 2,083 µg L^{-1} to 880 µg L^{-1} (58%). In addition, concentrations of nutrients in inflow water from the Tualatin River were high during the study period (Figure 4C). Over 13 years, mean inflow TP was 112 µg L^{-1}; mean inflow SRP was 45 µg L^{-1}; and mean inflow TN was 3,667 µg L^{-1}. Both lowest inflow TP (mean = 50 µg L^{-1}) and SRP (mean = 9 µg L^{-1}) occurred in 2012, the first year when inflow was treated with alum.

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Several other epilimnetic water quality variables, such as pH, turbidity and total suspended solids (TSS), varied in a similar pattern, showing an increasing trend in higher values between 2001 and 2004 and then dropping to relatively lower values between 2005 and 2013 (Figure 4D). Mean pH ranged from 9.4 (2001-2004) to 8.1 (2005-2013), with mean turbidity ranging from 10 NTU to 3 NTU and mean TSS ranging from 10 mg L^{-1} to 4 mg L^{-1}. Mean chlorophyll a was higher between 2001 and 2006 except 2002 and lower between 2007 and 2013 (2001 and 2003-2006: 25 µg L^{-1}; 2002: 12 µg L^{-1}; 2007-2013: 11 µg L^{-1}). Specific conductivity, pH, turbidity and TSS strongly correlated to chlorophyll a (Pearson correlation coefficient $r = 0.60$, $r = 0.54$, $r = 0.57$, $r = 0.44$, respectively, $p < 0.01$, n =38). Epilimnetic dissolved oxygen was saturated (mean = 117%). Epilimnetic water temperature varied between 18.1 °C and 24.3 °C with an average of 22.0 °C.

Climate and meteorological conditions were moderate during the study period (Figure 4E). Annual means of MEI ranks between July and September from 2001 to 2013 indicated moderate El Niño in 4 years (2002, 2006, 2009 and 2012, ranks from 50 to 56) and moderate to strong La Niña (moderate: 2007 and 2011, ranks=17; strong: 2010, rank=6), with weak ENSO strength (ranks from 22 to 48) in other years. Annual means of PDO indices between July and September from 2001 to 2013 indicated a cold phase in 2001 followed by a warm phase about 4 years (2002-2005). The PDO phase was then neutral until 2007, following a cold phase from 2008 to 2013. Summer air temperatures were moderate, ranging from 18.0 to 21.4 °C with an average of 20.1 °C. Summer wind speed ranged from 0.04 to 0.38 m s^{-1} with an average of 0.18 m s^{-1}, indicating calm to light air conditions. The prevailing wind directions were west-northwest and east during

the study period, accounting for 17.4% and 15.8% of all wind directions, respectively (Appendix C). Two summers in 2004 (mean rainfall = 92 mm) and 2013 (72 mm) were wetter than on average of other years (15 mm). Summer solar radiation ranged from 263 W m^{-2} to 356 W m^{-2} with an average of 307 W m^{-2}.

4.2 Changes of phytoplankton assemblages in relation to environmental conditions

Between July and September from 2001 to 2013, a total of 129 phytoplankton taxa belonging to eight divisions were identified, including 45 chlorophytes, 31 cyanobacteria, 25 diatoms, and 28 taxa belonging to the other five divisions (Appendix D). Phytoplankton assemblages were numerically dominated by cyanobacteria (mean relative abundance = 45%), followed by diatoms (23%) and chlorophytes (15%) (Appendix E).

Summer phytoplankton assemblages changed year-to-year over 13 years (Figure 5 and Figure 7). Overall, the relative abundance of cyanobacteria biovolume decreased (R^2=0.5, p<0.001) while that of diatom biovolume increased (R^2=0.5, p<0.001). Between 2001 and 2007, summer phytoplankton communities were dominated by cyanobacteria (mean relative abundance = 66%), most comprised of non-nitrogen fixers, *Lyngbya* sp. (now known as *Limnoraphis*) and *Microcystis* sp.. Between 2008 and 2012, relative abundances of cyanobacteria were much lower (mean = 17%). In 2013, cyanobacteria was again dominant (49%). Nitrogen fixers, *Aphanizomenon flos- aquae*, *Anabaena* sp.*iroides* and *Anabaena planctonica*, were most abundant cyanobacteria from 2008 to 2013. Diatoms increased from less than 10% to more than 30% over the study period, primarily comprised of *Melosira* sp. and *Fragilaria* sp.. In addition, two noticeable

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changes occurred in 2002 and 2006, the second years of the operations of 2001

hypolimnetic aeration and 2005 alum surface application. Relative abundance of

cyanobacteria dropped 40% in both 2002 and 2006 compared to the previous years.

Chlorophyte (41%) was high in 2002, while diatoms (35%) was high in 2006.

The cluster analysis identified 4 groups based on phytoplankton assemblages between

July and September from 2001 to 2013 (Table 3, Figure 6, Figure 7A-D and Appendix F),

and the groups were significantly different (p=0.001) among each other based on

ANOSIM. Group 1 (G1) was dominated by cyanobacteria that accounted for, on

average, 81% of the total phytoplankton biovolume, which was mainly comprised of non-

nitrogen fixers (67%). *Lyngbya* sp., a filamentous non-nitrogen fixing cyanobacteria,

was both the dominant species and the indicator species of G1. Group 2 (G2) was

dominated by cyanobacteria (60%) and chlorophytes (24%). The dominant species and

indicator species in this group were *Microcystis* sp. and *Closteriopsis longissimi* that

were both common species found in eutrophic freshwaters. Group 3 (G3) had the highest

relative abundance of diatoms (35%) that was more abundant than cyanobacteria (30%).

G3 was comprised of common summer eutrophic assemblages, including non-motile

diatoms (*Melosira* sp. and *Fragilaria* sp.), motile dinoflagellates (*Ceratium hirundinella*),

nitrogen fixing cyanobacteria (*Aphanizomenon flos-aquae* and *Anabaena* sp.), and

chlorophyte (*Pandorina* sp.). Group 4 (G4) was dominated by chlorophytes (38%) and

diatoms (30%), while cyanobacteria were least abundant in this group. The dominant

species and the indicator species of G4 belonged to cosmopolitan taxa found in various

water trophic conditions, including oligotrophic to mesotrophic *Oocystis* sp. and

eutrophic *Coelastrum* sp.. Regarding species diversity, G3 was most diverse (Shannon's

diversity index = 2.9), followed by G4 (2.4), G2 (2.1) and G1 (1.9).

The NMDS plots revealed that phytoplankton assemblages shifted from cyanobacteria

dominance to diatom and chlorophyte dominance over 13 years along NMDS axis I

(Figure 7E-F). The NMDS axis I primarily reflected the gradients of nutrients (e.g.,

epilimnetic and hypolimnetic TP and TN, and inflow SRP) (Table 4 and Figure 7A-D).

This change corresponded to the four groups identified by the cluster analysis. The

NMDS axis I separated the cyanobacteria dominated G1 and G2 with nutrient-rich

conditions from the diatom and chlorophyte dominated G3 and G4 with lower nutrient

concentrations. Specifically, hypolimnetic nutrients were associated with G1, although

only hypolimnetic TP and TN was significant vectors, while inflow SRP was

significantly associated with G2. Other environmental variables that significantly

correlated with the NMDS space defined by phytoplankton assemblages included *p*H,

dissolved oxygen and chlorophyll *a*, indicating high primary production (G1), as well as

specific conductivity, turbidity and TSS, indicating turbid water condition (G2), and NP

ratio (G4). The PDO index was statistically significant associated with the NMDS space

and positively associated with G2. Climatic MEI, air temperature, rainfall, wind speed,

wind direction and solar radiation did not show significant association with the NMDS

space (Table 4).

4.3 Changes of phytoplankton assemblages in relation to lake management practices

The changes of phytoplankton assemblage cluster groups were well associated with lake management practices in temporal terms (Figure 8). G1 dominated by cyanobacteria was associated with the period before the major management practices (hypolimnetic aeration in August 2001 and alum injection in March 2008). G2 dominated by cyanobacteria and chlorophytes occurred between one year after hypolimnetic aeration and one year before inflow reduction (June 2005) and alum surface application (August 2005). G3 dominated by diatoms and cyanobacteria occurred after alum surface application. G4 dominated by chlorophytes and diatoms occurred in 2009 (the year after alum injection), 2011 (the year after long period of drawdown) and 2013 (the year after inflow alum treatment).

5. Discussion

The phytoplankton communities of Oswego Lake have experienced profound changes in the 13-year study period. The summer phytoplankton assemblages have shifted from cyanobacteria dominance to diatom and chlorophyte dominance (Figure 5 and Figure 8). The changes of summer phytoplankton assemblages were significantly correlated with the changes of physical and chemical water conditions and the large climatic pattern PDO (Figure 7) and strongly associated with lake management practices (hypolimnetic aeration and alum application) (Figure 8).

The results illustrate that alum treatment to Oswego Lake was the major contribution to a substantial reduction of phosphorus in the water column and improvement of water quality overall (Figure 4 and Appendix G). The concentration of epilimnetic TP and SRP declined by 66% and 93%, respectively, compared to the period prior to alum treatment (Figure 4A). Meanwhile, the concentration of epilimnetic chlorophyll a and turbidity decreased by 48% and 69%, respectively (Figure 4D).

The results were comparable to the other cases of alum applications to lakes where alum resulted in reduction of in-lake total phosphorus and chlorophyll a concentrations and improvement of water clarity (Huser, Egemose, et al., 2016; Welch & Cooke, 1999). For example, Mirror and Shadow lakes, WI, were both within residential watersheds. The lakes are eutrophic due to storm sewer loading and internal loading. Alum additions successfully reduced mean TP from 93 to 20 μg L^{-1} (78% reduction) in Mirror Lake and from 55 to 23 μg L^{-1} (58% reduction) in Shadow Lake (Welch & Cooke, 1999).

Similarly, in Dollar Lake, OH, a small urban lake, alum lowered surface TP by 65% from the pretreatment concentration of 82 µg L^{-1} and chlorophyll *a* by 61% from 41 µg L^{-1} for 7 years (Welch and Cooke, 1999). Huser, Egemose et al.'s (2016) study on the short-term effects of alum on 83 urban lakes of varying size, morphology and hydrology also found that alum reduced epilimnetic TP from 101 to 36 µg L^{-1} (64% reduction) and chlorophyll *a* from 43 to 16 µg L^{-1} (63% reduction), and increased Secchi depth from 1.6 to 2.4 m (50% increase).

Few studies report the change of SRP but only TP when evaluating alum effectiveness on nutrient and cyanobacteria bloom control, probably because TP is relatively easy to measure (Huser, Futter, et al., 2016), although bioavailable forms of P, such as SRP, are more important than TP in understanding water quality and cyanobacteria blooms (Hatch, Reuter, & Goldman, 1999). In Oswego Lake, the substantial decline of SRP by 93% between pre- and post-alum treatment may be a strong evidence for successful reduction of P by alum treatment (Figure 4A and Appendix G).

In addition, previous studies have found that alum effectiveness lasted for about 13 to 15 years on average but declined over time due to bioturbation, sediment settling processes, and burial by new sediments (Huser, Futter, et al., 2016; Welch & Cooke, 1999). The concentrations of TP and SRP in Oswego Lake were retained well between 2005 and 2013 at concentrations after the initial alum treatment in 2005, probably due to the annual alum treatments, including surface application, injection and inflow alum addition, and the effect of sediment oxidation by hypolimnetic aeration (B. C. Moore et al., 2012).

While in-lake nutrient concentrations were substantially reduced after the alum application and hypolimnetic aeration, a shift from the phytoplankton communities dominated by cyanobacteria to diatoms and chlorophytes has been observed (Figure 5 and Figure 8). Although there is no single factor that drives phytoplankton community dynamics; in reality, it is due to the complex combination and interactions of physical, chemical, and biological phenomena, such as light, temperature, nutrients and food-web structure (Kosten et al., 2012). However, it has been widely accepted that phosphorus is typically the limiting nutrient in lakes and has strong correlation with phytoplankton production (Carpenter, 2008; McCauley et al., 1989; Prairie, Duarte, & Kalff, 1989; Schindler, 2012; Schindler et al., 2008); therefore, phosphorus is commonly used as a primary trophic indicator (Carlson, 1977). Eutrophic lakes with internal phosphorus loading during vertical thermal stratification are particularly favorable for the massive development of cyanobacteria because cyanobacteria are able to regulate their buoyancy and access nutrient-rich hypolimnetic water (Carey et al., 2012).

Many lake restoration programs focused on reduction of phosphorus have proved to be effective in reducing cyanobacteria blooms (Huser, Futter, et al., 2016; Zamparas & Zacharias, 2014). For example, Newman Lake, WA, is a dimictic lake and has stable thermal stratification between May and September. Cyanobacteria blooms in Newman Lake occurred annually in the 1970s and 1980s primarily due to summer hypolimnetic oxygen depletion and internal phosphorus loading (B. C. Moore et al., 2009). Cyanobacteria had a peak in 1989, accounting for >90% of total phytoplankton biovolume (B. C. Moore & Christensen, 2009). Since 1989, a serial management practices implemented in Newman Lake for control of in-lake phosphorus and

cyanobacteria blooms included whole-lake alum treatment, hypolimnetic oxygenation and microfloc alum injection. Cumulative management efforts have successfully reduced average summer TP from pre-restoration 55 µg L^{-1} to an average of 21 µg L^{-1} over 7 years. Cyanobacteria blooms were no longer evident after the first whole-lake alum treatment, and cyanobacteria comprise <8% of total phytoplankton biomass, while diatoms comprise between 50% and 75% and chlorophytes comprise <15% of total phytoplankton biomass (B. C. Moore et al., 2012, 2009). Species shifts also occurred. Almost exclusive *Microcystis* sp. decreased after the first alum treatment with increasing *Anabaena flos-aquae* in the cyanobacteria assemblage (B. C. Moore et al., 2009). Later, *Melosira* sp. became the most abundant phytoplankton during aeration (Thomas, Funk, Moore, & Budd, 1994). This shift in phytoplankton assemblages from cyanobacteria dominance to diatom and chlorophyte dominance was most likely a response to lower phosphorus availability (B. C. Moore & Christensen, 2009).

The observations of changes of nutrients and phytoplankton communities after alum addition and hypolimnetic aeration in Oswego Lake are very similar to the findings in Newman Lake. Previous lake analysis on Oswego Lake identified that internal loading was one of the most significant sources of phosphorus in the lake (H. Gibbons & Welch, 2004). Therefore, hypolimnetic aeration since 2001 and alum application since 2005 have been implemented in Oswego Lake to reduce internal P loading in control of cyanobacteria blooms. Since 2005 after alum treatment to Oswego Lake, coupled with hypolimnetic aeration, summer phytoplankton assemblages showed a 62% reduction of cyanobacteria biovolume (Figure 5), and cyanobacteria dominated phytoplankton communities gradually shifted to diatom and chlorophyte dominance (Figures 7 and

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Figure 8). Based on the concept of community ecology, the succession of dominant

species is partly the consequence of changes to the environmental conditions (Reynolds,

2006). Compared to the "pre-shift" years of 2001 to 2004, the most remarkable changes

in nutrient levels have occurred since 2005, which may be the most probable explanation

for the presence of the diatom dominant phytoplankton communities (Group 3).

Furthermore, in Oswego Lake, the dominance of *Melosira* occurred in summer

stratification periods, which is very likely a direct result of hypolimnetic aeration.

Melosira is not capable of independently staying within the photic zone, due to its heavy

silicon body, without upwelling water current during summer stratification. It is apparent

that the hypolimnetic aeration in Oswego Lake circulated lake water and generated

vertical mixing of the water sufficient to resuspend *Melosira* in the photic zone for

growth. Lund (1971) observed a significant increase in summer *Melosira* population

during the artificial destratification of a small lake in the English Lake District.

Similarly, Thomas et al. (1994) reported that *Melosira* particularly adapted to

hypolimnetic aeration that induced a turbulent environment and became the most

abundant phytoplankton during summer aeration in Newman Lake. Therefore, the

hypolimnetic aeration in Oswego Lake apparently played an important role in

establishing a dominance of *Melosira* in summer phytoplankton communities.

Results have shown, however, that hypolimnetic aeration alone prior to alum treatment

did not significantly lower nutrient concentrations (Figure 4A), and that the major shift of

dominant species from cyanobacteria to diatoms and chlorophytes occurred after the

alum treatment (Figure 5 and Figure 8), which suggests that hypolimnetic aeration alone

was not sufficient, but the synergistic effect of hypolimnetic aeration and alum treatment

is most likely decisive. Hypolimnetic aeration, as a common management technique, has

been reported to be successful in reducing internal phosphorus loading from the anoxic lake sediments, thus controlling cyanobacteria (Beutel & Horne, 1999; Cooke et al., 2005). In Oswego Lake, the hypolimnetic nutrients (TP, SRP and TN) dropped steeply in 2002, the second year of the operation of hypolimnetic aeration (Figure 4B). However, between 2001 and 2004, the epilimnetic nutrients were still high (Figure 4A). And cyanobacteria were most dominant, accounting for > 80% of total phytoplankton biovolume, with the exception of 2002 when cyanobacteria comprised 51% and chlorophytes comprised of 41% (Figure 5). Moreover, *Microcystis* was most dominant in 2002 and a *Microcystis* bloom occurred in 2004. These observations may reflect high nutrient inputs from external sources (Oberholster et al., 2006).

Oswego Lake, within an urban watershed, has a high watershed/drainage area to lake area ratio of more than 10:1, indicating a strong influence of watershed to the lake (Schueler & Simpson, 2001). The drainage pipe network contains 70 stormwater outfalls surrounding the lake, and 40% of storm water from the City of Lake Oswego drains into the lake directly through the pipes (Rubenson, 2016). In urbanized watersheds, the problem of stormwater runoff from impervious surfaces, such as roofs, roads and driveways, has been widely recognized, which is that piped runoff rapidly transfers nutrients, sediments and other pollutants throughout the urban watershed to the receiving waters, resulting in degradation of water quality in receiving waters (Roy et al., 2008). In the watershed of Oswego Lake, imperviousness has been found to be the primary landscape characteristic influencing phosphorous loading, therefore, stormwater runoff is an important phosphorus source to the lake (Rubenson, 2016).

In addition, three stream inflows contribute significant phosphorus to the lake. Rubenson (2016) found that the concentrations of total phosphorus and total suspended solids were similar in piped drainages and streams, but due to higher flows in streams, streams contribute a higher load of phosphorus to the lake. Annually, approximate 376 kilograms of phosphorus loads into the lake from the Tualatin River and approximate 1023 kilograms from Springbrook Creek (ODEQ, 2001). Eroded riparian and soil along the riverbank input significant phosphorus into Lost Dog Creek that further transports phosphorus to the lake (B. S. Johnson, 2009). Soil erosion is a major source of phosphorus in the watersheds of northwestern Oregon that is between an uplifted subduction complex of the Coast Range and active andesitic volcanoes of the Cascade Range (Abrams & Jarrell, 1995; Retallack & Burns, 2016). Much of the watersheds in northwestern Oregon are underlain by volcaniclastic marine sandstones and siltstones and large areas of the basin floor are covered by nutrient-rich silts from the Missoula Floods dating back to the last ice age (Foster, 2009; Retallack & Burns, 2016). These watershed geological characteristics dictate that the Oswego Lake watershed is a native phosphorus source to the lake. As watershed nutrient loading is still high, hypolimnetic aeration targets internal loading of phosphorus from sediments may not be effective in reduction of phosphorus in the lake (Liboriussen et al., 2009).

It is apparent that reduction of phosphorus in the primary stream tributaries to the lake is necessary to further reduce in-lake phosphorus. The Tualatin River is the primary water supply to the lake during summer in order to maintain water level in the lake for summer water uses, such as recreation and irrigation. During the study period, phosphorus concentrations of inflow water from the Tualatin River were relatively high (mean TP

=112 µg L^{-1}, mean SRP = 45 µg L^{-1}) (Figure 4C). Beginning in 2005, the annual inflow

duration was reduced from 3-5 months (2001-2004) to 2-3 months (after 2005), for the

purpose of reducing inflows; consequently, nutrient loading from the Tualatin River to

the lake were reduced. Although the canal TP and SRP declined after inflow duration

reduction in 2005, inflow volume reduction was not as substantial as reduction of in-lake

TP and SRP when the lake was treated with alum (Figure 4C). This result suggests that

the significant reduction of in-lake phosphorus in 2005 was very likely due to the in-lake

nutrient management practices (alum and hypolimnetic aeration).

Beginning in 2012, the inflow water was treated with alum before entering the lake.

Inflow phosphorus steeply decreased immediately after the inflow alum treatment in

2012, and inflow SRP remained low in 2012 and 2013 (Figure 4C), which suggests that

alum was effective in removing phosphorus from inflow to the lake. Meanwhile, the fact

that chlorophyte and diatom dominated phytoplankton communities (Group 4) occurring

in the following year of inflow alum treatment suggests that inflow alum may contribute

to the change of phytoplankton communities. The effectiveness of inflow alum treatment

in removal of phosphorus prior to discharge into the lake and in improvement of lake

water quality was also found in other studies where the major tributary to the lake

contributes inflow water carrying high phosphorus loading (Heinzmann & Chorus, 1994;

Pilgrim & Brezonik, 2005). For example, in two urban eutrophic lakes in Minnesota,

Tanners Lake and Fish Lake, the treatments to the inflow water with alum resulted in

reduction of in-lake TP, accompanied by decreases in chlorophyll *a* and improvement in

Secchi disk depth (Pilgrim & Brezonik, 2005). In addition, a significant correlation was

found between inflow SRP and in-lake SRP in Oswego Lake (Pearson correlation

coefficient r=0.52, p <0.001, n=38), suggesting that reducing SRP in inflow water may result in reduction of SRP in the lake; thus, inflow alum treatment is necessary in nutrient management in Oswego Lake.

The management activities in Oswego Lake during the study period, besides hypolimnetic aeration and alum addition directly targeting water quality, included two water level drawdowns, which dropped the water level 3 meters in 2006 for 138 days and 7 meters in 2010 for 264 days. Although the purpose of two drawdowns were lake and facility maintenance, the consequence of water level drawdown can be important in affecting lake ecosystem (Bakker & Hilt, 2016; Pan et al., 2018). For example, during drawdown, nutrients and plankton in the lake can be partly removed while the water body is flushed out. Drawdown, in some extend, can be seen as a "reset" and an important disturbance of the lake ecosystem. In Oswego Lake, no significant change in the phytoplankton community was observed after the first drawdown; however, in two years following the second drawdown, chlorophytes reached one of three high relative abundance values (Figure 5) and a more diversified assemblage of chlorophytes and diatoms presented, which indicated that duration and strength of drawdown may affect lake ecosystems. Group 4 probably was a result of the second drawdown because the phytoplankton species in Group 4 are regarded as R-strategist that can respond quickly and positively to environmental changes (Reynolds, 2006).

In this study, the Pacific Decadal Oscillation (PDO) index showed a statistically significant correlation with the summer phytoplankton communities that, however, were not significantly correlated with the multivariable ENSO index (MEI) in Oswego Lake

(Figure 7D and Table 4). This result is comparable to a study in Lake Washington, which found that the impact of PDO on the spring plankton communities was stronger than ENSO (Winder & Schindler, 2004). Winder and Schindler (2004) showed that vernal warming induced advanced stratification onset and phytoplankton spring bloom in Lake Washington by 20 days, which coincided with the warm phase of the PDO. Meanwhile, the delayed stratification termination was described by the regression model of air temperature and the PDO index. Thus, Winder and Schindler (2004) concluded that PDO is an important driver for physical processes in Lake Washington. However, in my study, PDO switched phases from warm to cool during 2005 and 2006, a period that coincided with the beginning of the lake's alum treatment in 2005 (Figure 8 and Appendix H). Although many studies have reported that changing climatic conditions can directly and indirectly affect phytoplankton species succession, structure and composition through altering lake's physical, chemical and biological conditions (i.e., water column thermal stratification, thermocline depth, nutrient availability, and zooplankton succession), as well as watershed hydrological conditions (i.e., basin streamflow) (Livingstone, 2003; Livingstone & Dokuli, 2001; Magnuson et al., 1997; Shimoda et al., 2011; Winder & Schindler, 2004), isolation of climate impacts is very difficult due to complex internal feedbacks to stressors and complicated confounding factors, such as eutrophication and acidification in lakes themselves (Adrian et al., 2009). Without further analysis of the correlation between PDO and a wide spectrum of lake parameters such as annual variabilities and stratification characteristics across a long period of time due to the PDO cycles of 20-30 years (Mantua & Hare, 2002), any conclusions regarding significant PDO impacts on the changes in summer phytoplankton communities would be premature.

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In addition, correlations of weather conditions (air temperature, rainfall, wind speed, wind direction and solar radiation) and summer phytoplankton communities were insignificant during the study period in Oswego Lake (Figure 7H and Table 4). However, it is notable that the high production of cyanobacteria might be in response to high rainfalls and high water temperatures in 2004 and 2013, including a *Microcystis* bloom in 2004 (Appendix J). In the region of the Oswego Lake watershed, the summer is usually characterized as warm and dry. Warming favors blooms of harmful cyanobacteria and likely increases cyanobacteria dominance in phytoplankton communities (Paerl & Huisman, 2008). Wetter summers than usual can increase external nutrient loading to the lake and thus increase cyanobacterial biomass (Reichwaldt & Ghadouani, 2012), especially if the lake is in an urban setting with nutrient rich soil within the watershed.

6. Study limitation

The influence of climate impacts on phytoplankton communities in this study was not conclusive. Firstly, the Pacific Decadal Oscillation (PDO) index showed a statistically significant correlation with the summer phytoplankton communities that, however, were not significantly correlated with the multivariable ENSO index (MEI) and weather conditions (air temperature, rainfall, wind speed, wind direction and solar radiation) in Oswego Lake. Meanwhile, PDO switched phases from warm to cool during 2005 and 2006, a period that coincided with the beginning of the lake's alum treatment in 2005. Without further analysis of the correlation between PDO and a wide spectrum of lake parameters such as annual variabilities and stratification characteristics across a long period of time due to the PDO cycles of 20-30 years (Mantua & Hare, 2002), any conclusions regarding significant PDO impacts on the changes in summer phytoplankton communities would be premature. Secondly, the multivariable ENSO index (MEI) was found no significant correlation with the summer phytoplankton community variabilities in Oswego Lake. It should be noted that this study falls by chance between two extreme El Niño events (1997-1998 and 2015-2016) (Appendix I). Both events have been reported to have strong impacts on phytoplankton in freshwater and coastal waters (Cavole et al., 2016; McGowan et al., 2005). However, in this study from 2001 to 2013, the intensity of both El Niño and La Niña were mostly categorized as weak or moderate. This study found no significant relationship between MEI and the phytoplankton community dynamics, which was probably due to the low variability of ENSO as well. Therefore, I cannot conclude the changes of the phytoplankton communities were due to lake management alone, although that was a major finding of this study.

In addition, this study did not investigate food web structure that directly regulate phytoplankton population (Shapiro et al., 1975). Research on food web networks in lake ecosystems should afford a deeper understanding of phytoplankton dynamics.

7. Conclusion and management implication

This study investigated a long-term change of phytoplankton communities in an urban lake to better understand what factors most likely impacted such change. During the years 2001 to 2013, the summer phytoplankton communities of Oswego Lake shifted in dominance from cyanobacteria to diatoms and chlorophytes. This shift very likely resulted from the reduction of in-lake phosphorus due to alum treatments, coupled with hypolimnetic aeration, because a strong temporal relationship was found between these management practices and the changes of in-lake phosphorus and phytoplankton communities.

Alum treatment in Oswego Lake, as well as in other lake restoration projects, has been effective in reduction of in-lake phosphorus concentration and in control of cyanobacteria blooms (Cooke et al., 2005). Although assessment on management cost was not conducted in this study, other studies have documented that water treatment using chemicals, such as alum, can be very costly (EPA, 2015). Costs of alum application depend on dosage used, area treated, and equipment and labor costs. EPA (2015) reported that the costs associated with alum application could be more than $28 million in capital and $1.4 million in annual operation and maintenance for a dredging and alum treatment plan intended to last more than 50 years in Lake Lawrence (1.3 km^2), Washington.

The results of this study implied that watershed external nutrient loading can be significant to Oswego Lake, which is consistent with Rubenson's (2016) study of the

influence of stormwater drains and landscape characteristics on phosphorus loading in Oswego Lake. Urban watershed loads through stormwater runoff from impervious surfaces and soil erosion lead to increasing sedimentation of nutrients and organic materials in lakes and subsequently reinforcing internal cycling of nutrients, fueling cyanobacteria blooms (Søndergaard et al., 2003). Current watershed management is in place to reduce nutrient loading to the lake. In addition to Tualatin River inflow control, there has been implementation of several storm water best management practices (e.g., wet retention ponds, dry detention ponds, swales, infiltration rain gardens, underground injection control systems, and lined planter rain gardens), and bank stabilization through promotion of native vegetation and removal of invasive species along stream banks. The watershed is the ultimate source of nutrient input to lakes, so reduction in supply of nutrients from the lake watershed is a sustainable and cost-efficient approach for long-term efforts to protect water quality.

The influence of climate impacts on phytoplankton communities in this study was not conclusive. Firstly, the Pacific Decadal Oscillation (PDO) index showed a statistically significant correlation with the summer phytoplankton communities in Oswego Lake. However, PDO switched phases from warm to cool during 2005 and 2006, a period that coincided with the beginning of the lake's alum treatment in 2005. Without further analysis of the correlation between PDO and a wide spectrum of lake parameters such as annual variabilities and stratification characteristics across a long period of time due to the PDO cycles of 20-30 years (Mantua & Hare, 2002), any conclusions regarding significant PDO impacts on the changes in summer phytoplankton communities would be premature. Secondly, no significant correlation was found between the multivariable

ENSO index (MEI) and the variability of the summer phytoplankton communities in Oswego Lake, probably due to the low variability of the climate factor during the study period. However, rising temperatures, increasing runoff of nutrients, and other climate change may favor cyanobacteria and accelerate deterioration of aquatic ecosystems; therefore, climate impacts should be carefully considered in ongoing analysis on both watershed and in-lake management.

The challenges of water quality management in Oswego Lake still exist, including dynamic lake system, expensive alum treatment and hypolimnetic aeration, significant nutrient input from watershed, and uncertain climate impacts on lake ecosystem. Adaptive management could be useful for facilitating decision making on lake management with an emphasis on reducing costs and uncertainties of the system over time in order to improve management. Adaptive management is a learning-based approach, integrating science and management, and incorporating what has been learned from past operations into ongoing management decision making. The modelling process plays an important role in adaptive management, as integrating existing scientific information, understanding of the system, and performance of management practices to frame predictions about impacts of management strategies for helping managers to achieve objectives. This study suggests that managers should consider utilizing a comprehensive water quality and hydrodynamic model that links management practices to watershed and lake ecological processes and climate variabilities, representing benefits and costs in terms of management inputs and outcomes, and allowing evaluation and prediction of management impacts.

Tables

Table 1: Morphometric characteristics of Oswego Lake.

Parameter	Values
Surface area	1.7 km^2
Maximum depth	16.7 m
Mean depth	7.9 m
Volume	12.7x10^6 m^3
Maximum length	5.1 km
Maximum width	0.6 km
Shoreline length	19.2 km
Residence time	2 months
Surface elevation	30 m
Watershed area	18.6 km^2

Table 2: Major management practices implemented in Oswego Lake between 2001 and 2013. (The information was provided by Mark Rosenkranz, the Water Resources Manager of Lake Oswego Corporation.)

	Description	Objectives	Operation period	Nutrient response
Hypolimnetic aeration	Increase oxygen levels near lake bottom in order to reduce P released from sediments. Hypolimnetic aeration circulates oxygenated water from the bottom of the thermocline to the bottom of the lake. This is accomplished in aeration towers that use a bubble plume to circulate low oxygen water up near the thermocline, thus forcing higher oxygen water down near the sediment. Hypolimnetic aeration maintains lake stratification.	Providing more oxygen at the sediment maintains the Fe-P complex, reducing P release to the overlying water and become bioavailable for algae growth.	Since 2001.8, during thermal stratification period	P decreases
Alum	Add aluminum sulfate to water to form hypo-reactive aluminum hydroxide floc that adsorbs and precipitates dissolved phosphate to sediments.	Precipitate P in water column to the sediment, reducing bioavailable P.	Alum surface application: • In 2005.8: used dry powdered compound; since 2006: used liquid alum • applied early spring to late summer yearly; usually started right after rain season Alum injection: Since 2008, usually applied in March until late summer	P decreases

	Description	Objectives	Operation period	Nutrient response
Drawdown	Discharge water from the lake through outlet gate to the Willamette River.	The primary purpose of drawdown is for seawall and dock maintenance. The 2010-2011 drawdown was required for a sewer pipe replacement; and 30,000 cubic yards of sediment was removed.	2006.11: lasted for 138 days ($\approx$ 5 months), drawdown 3 meters. 2010.9: lasted for 264 days ($\approx$ 9 months), drawdown 7 meters.	Major disturbance to the lake ecosystem. Potential effects of drawdown: • Erosion of exposed sediment • Sediment freezing • Potential to freeze aquatic plants • Potential high turbidity during rain events
Tualatin River inflow control	The primary water inflow during summer (June-Sept.) is from the Tualatin River. A headgate on Oswego Canal controls water flow into Oswego Lake from the Tualatin River and timing of water use depends on spring rainfall.	Maintain water level in the lake for the purpose of summer recreation and irrigation use.	2001-2004: opened for 3-5 months yearly; Since 2005: opened for 2-3 months yearly; * Since 2012.8, inflow treated with alum.	Due to relative high concentration of N and P in the Tualatin River, N is expected to increase in the lake during the time of headgate opening. Theoretically, because of alum treatment, P is not expected to be high. However, in practice, P removal is not complete so a fair amount is imported during summer.

Table 3: Phytoplankton dominant taxa and indicator taxa of 4 groups based on the cluster analysis. The assemblage of species composes of more than 50% of total phytoplankton biovolume.

Assemblage	Taxonomic group	Relative Abundance	Indicator value
Group 1			
Lyngbya sp.	non-N fixing Cyanobacteria	52%	0.8
Unicellular cyano	Cyanobacteria		0.5
Group 2			
Microcystis sp.	non-N fixing Cyanobacteria	31%	0.7
Closteriopsis longissimi	Chlorophyte	23%	0.9
Group 3			
Melosira sp.	Diatom	25%	0.7
Ceratium hirundinella	Dinoflagellate	10%	
Aphanizomenon flos-aquae	N-fixing Cyanobacteria	9%	
Fragilaria sp.	Diatom	6%	
Anabaena sp.	N-fixing Cyanobacteria		0.5
Pandorina sp.	Chlorophyte		0.5
Group 4			
Coelastrum sp.	Chlorophyte	26%	0.6
Fragilaria sp.	Diatom	21%	0.6
Ceratium hirundinella	Dinoflagellate	9%	
Oocystis sp.	Chlorophyte		0.7
Asterionella Formosa	Diatom		0.5

Table 4: Results from the environmental vectors fitting in the ordination space of the NMDS plot with variable scores along the two ordination axes (NMDS1-2), goodness-of-fit statistic r^2 and its significance (p-value). Results were sorted on r^2.

Variables	Abbreviation	Unit	NMDS1	NMDS2	r^2	p-Value	
Epilimnetic specific conductance	epiSpCond	mS.cm^{-1}	-0.87	0.50	0.60	0.001	***
Epilimnetic pH	epipH		-1.00	0.02	0.55	0.001	***
Epilimnetic total suspended solids	epiTSS	mg.L^{-1}	-0.73	0.68	0.46	0.001	***
Epilimnetic total phosphorus	epiTP	µg.L^{-1}	-0.97	0.23	0.45	0.001	***
Epilimnetic chlorophyll a	epiChla	µg.L^{-1}	-0.98	-0.21	0.44	0.001	***
Epilimnetic total nitrogen	epiTN	µg.L^{-1}	-0.95	0.30	0.42	0.001	***
Epilimnetic turbidity	epiTurbidity	NTU	-0.80	0.60	0.42	0.001	***
Pacific Decadal Oscillation index	PDO		-0.81	0.59	0.41	0.002	**
Hypolimnetic total nitrogen	hypoTN	µg.L^{-1}	-0.81	-0.59	0.33	0.002	**
Epilimnetic nitrogen to phosphorus ratio	epiNP		0.84	-0.54	0.29	0.005	**
Inflow soluble reactive phosphorus	inflowSRP	µg.L^{-1}	-0.93	0.36	0.28	0.008	**
Hypolimnetic total phosphorus	hypoTP	µg.L^{-1}	-0.95	-0.31	0.22	0.006	**
Epilimnetic percent saturation of dissolved oxygen	epiDOpct	%	-0.89	-0.45	0.18	0.034	*
Air temperature	airTemp	°C	0.44	-0.90	0.16	0.063	.
Epilimnetic water temperature	epiTemp	°C	-0.76	-0.65	0.14	0.087	.
Hypolimnetic soluble reactive phosphorus	hypoSRP	µg.L^{-1}	-0.96	-0.27	0.12	0.107	
Epilimnetic soluble reactive phosphorus	epiSRP	µg.L^{-1}	-0.82	0.57	0.11	0.141	
Multivariate ENSO index	MEI		-0.98	0.19	0.10	0.183	
Solar irradiation	Solarrad	W.m^{-2}	0.19	-0.98	0.08	0.245	
Wind speed (u)	Windsp	m.s^{-1}	-0.61	0.79	0.07	0.291	
Wind direction (v)	Winddir	degree	0.33	-0.95	0.07	0.296	
Rainfall	Rain	mm	0.11	0.99	0.04	0.516	
Inflow total phosphorus	inflowTP	µg.L^{-1}	-1.00	0.10	0.03	0.599	
Inflow total nitrogen	inflowTN	µg.L^{-1}	0.93	0.36	0.03	0.577	

Significance codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

p-values based on 1000 permutations.
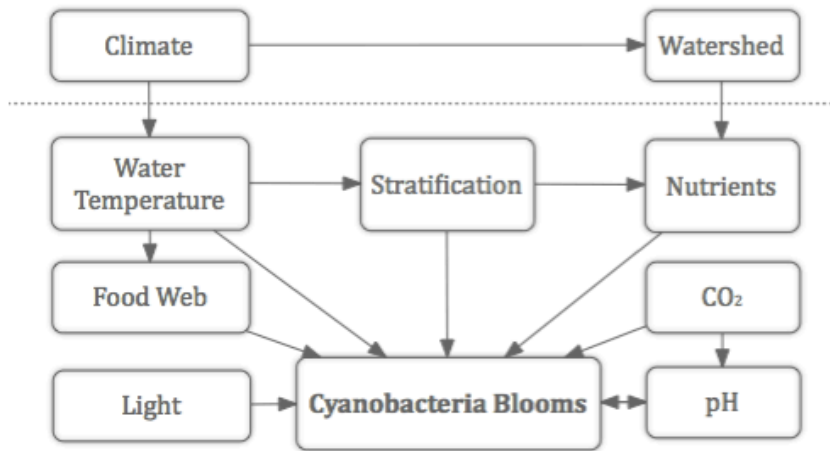
Figures

Figure 1: Conceptual diagram showing the major factors that affect cyanobacteria blooms.
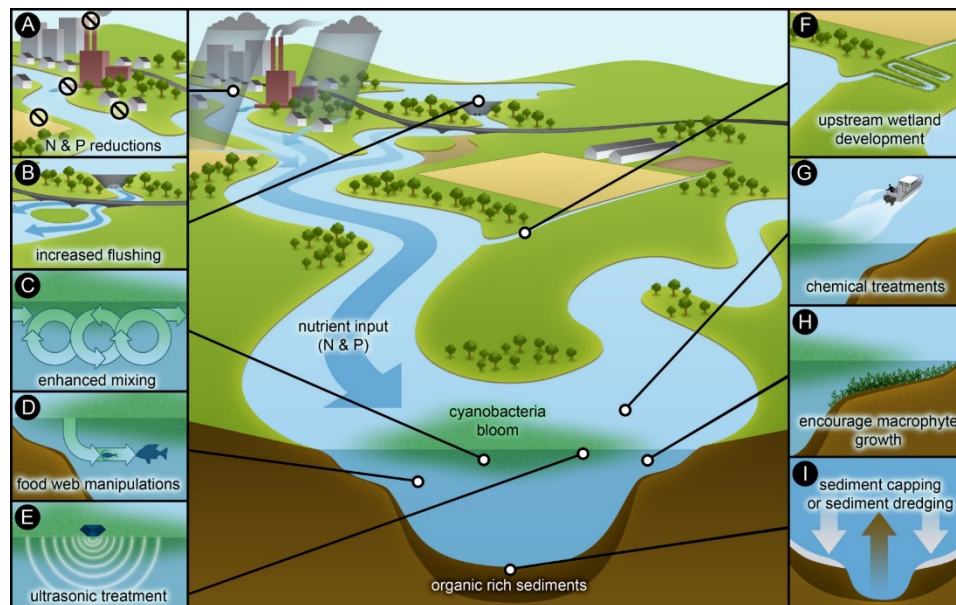

Figure 2: Conceptual illustration of various approaches currently in use to control cyanobacteria blooms (from Paerl et al., 2016).
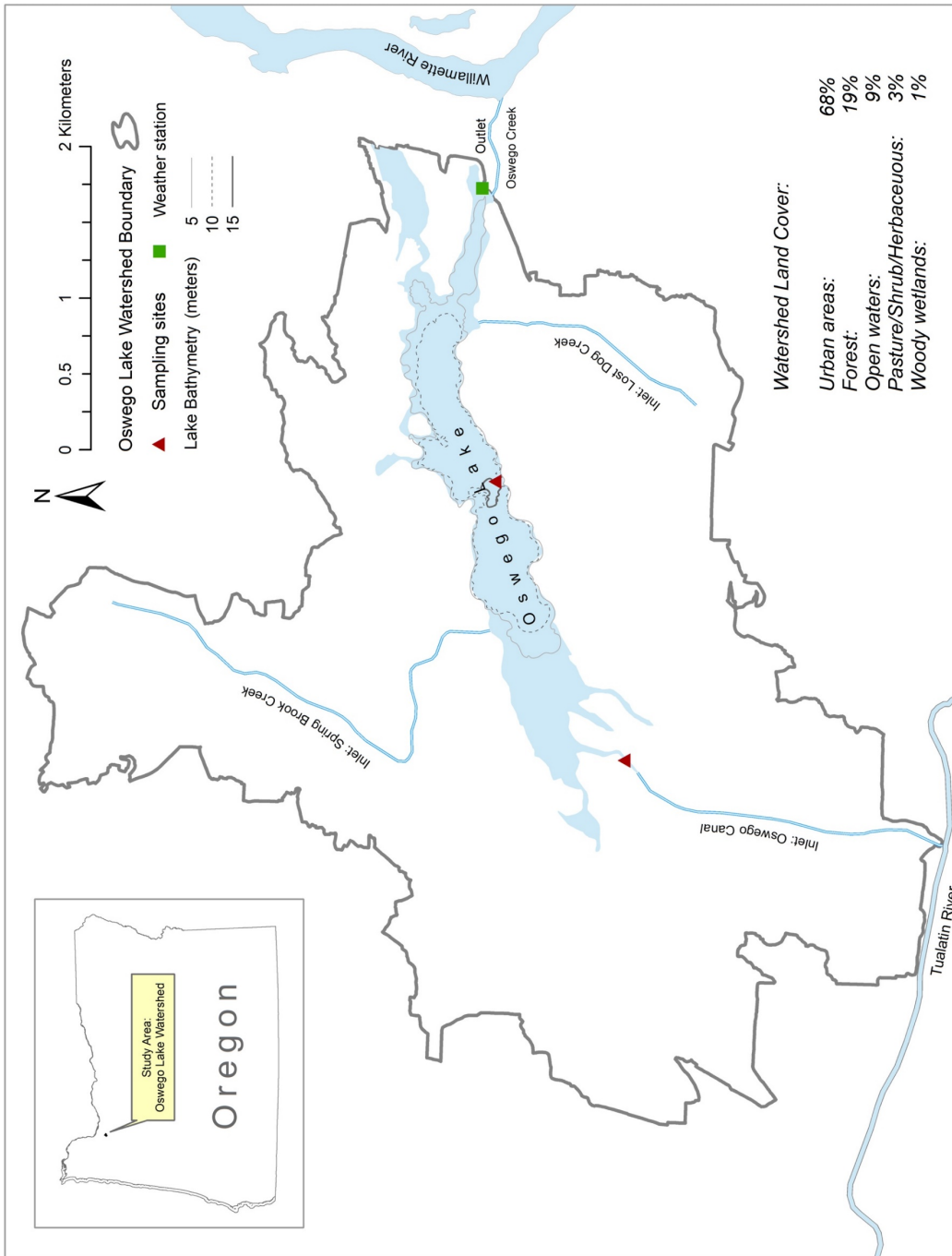
Figure 3: Location of Oswego Lake within its watershed.
Data sources: National Geographic World Map, Oregon Spatial Data Library, and Lake Oswego Corporation.
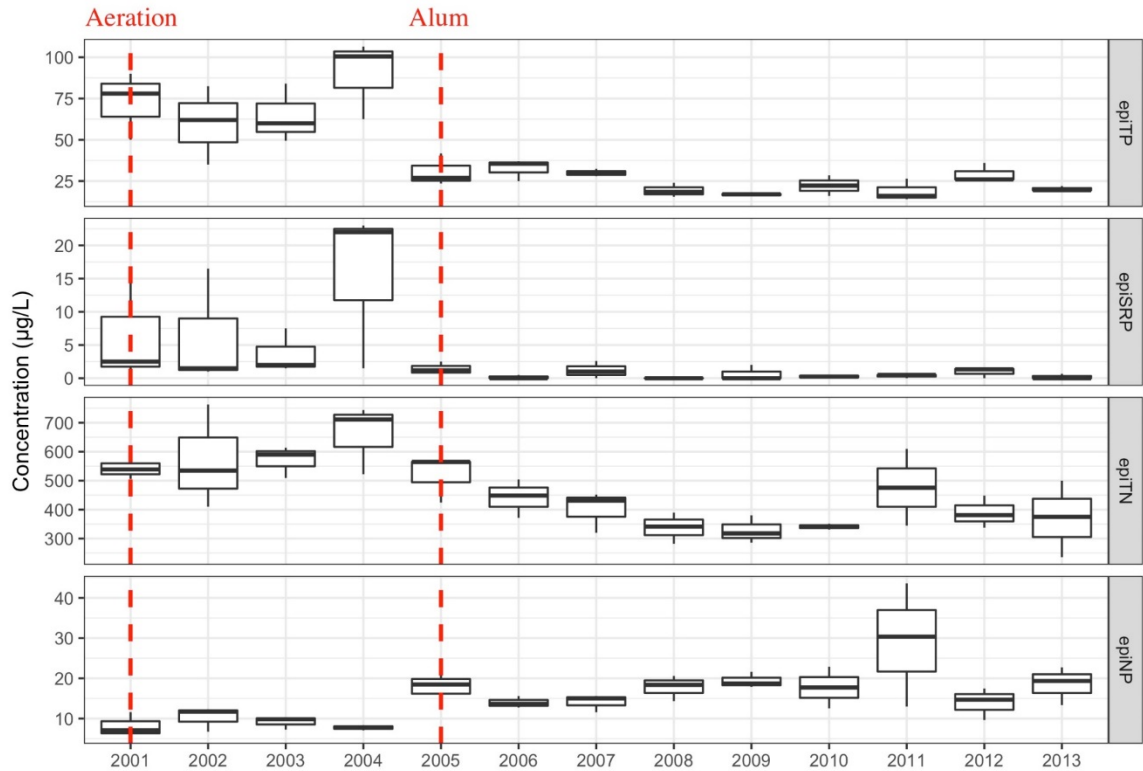
(A)

Figure 4: Boxplots showing year-to-year variations of the selected environmental variables between July and September from 2001 to 2013 in the deep basin of Oswego Lake. (A) epilimnetic nutrients. Figure continued on the following pages.

The box includes the data between first and third quantiles and the median (black bar). The upper whisker extends to the largest value no further than 1.5 IQR (the interquartile range), while the lower whisker extends to the smallest value at the most 1.5 IQR.

Abbreviations: epi – epilimnion; hypo – hypolimnion; TP – total phosphorus; SRP – soluble reactive phosphorus; TN – total nitrogen; NP – nitrogen to phosphorus ratio; Temp – temperature; SpCond – specific conductance; DOpct – percent saturation of dissolved oxygen; TSS – total suspended solids; Chla – chlorophyll a; MEIrank – ranks of the multivariate ENSO index (MEI); PDO – the PDO index based on Mantua et al. (1997); Windsp – wind speed (wind component u); Winddir – wind direction (wind component v); Solarrad – solar radiation.
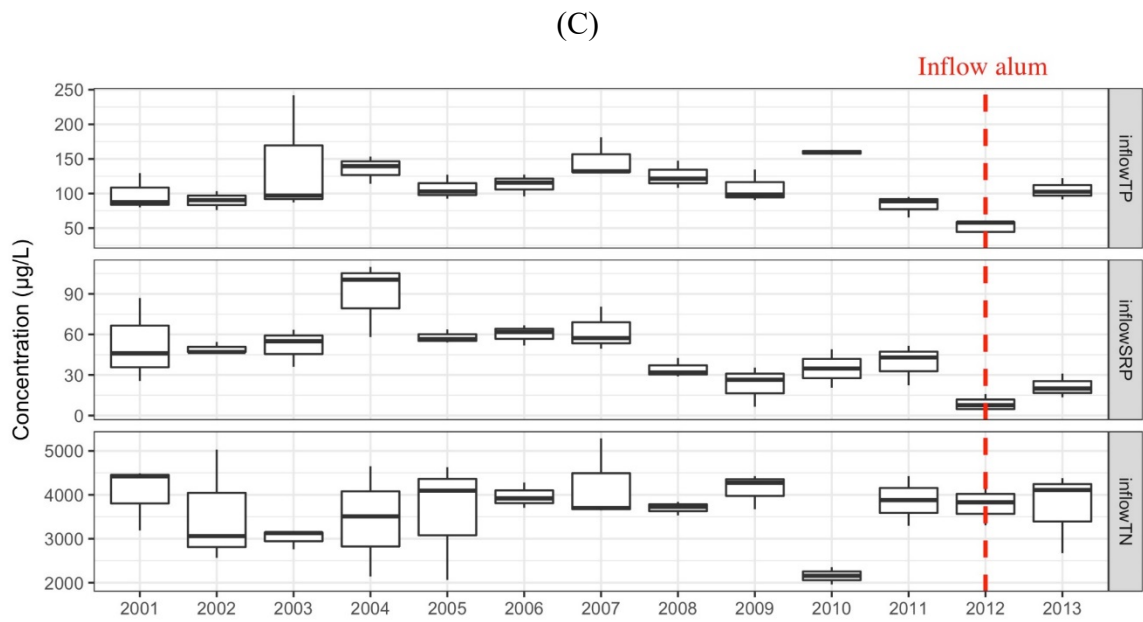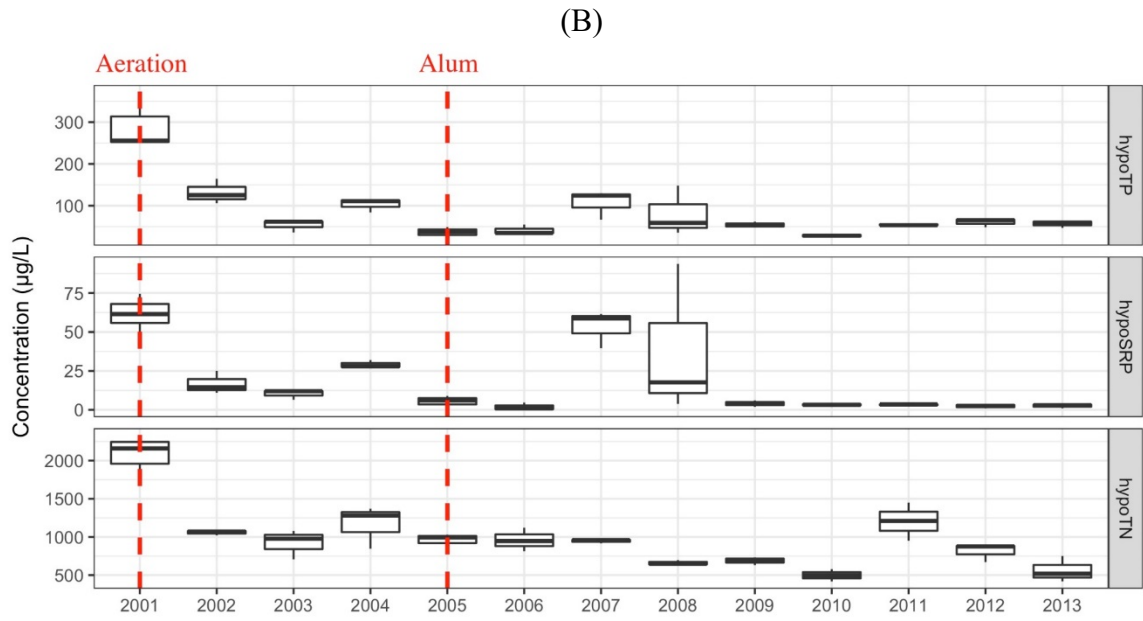
(B)

(C)

Figure 4: *Continued*. (B) hypolimnetic nutrients, (C) inflow nutrients.
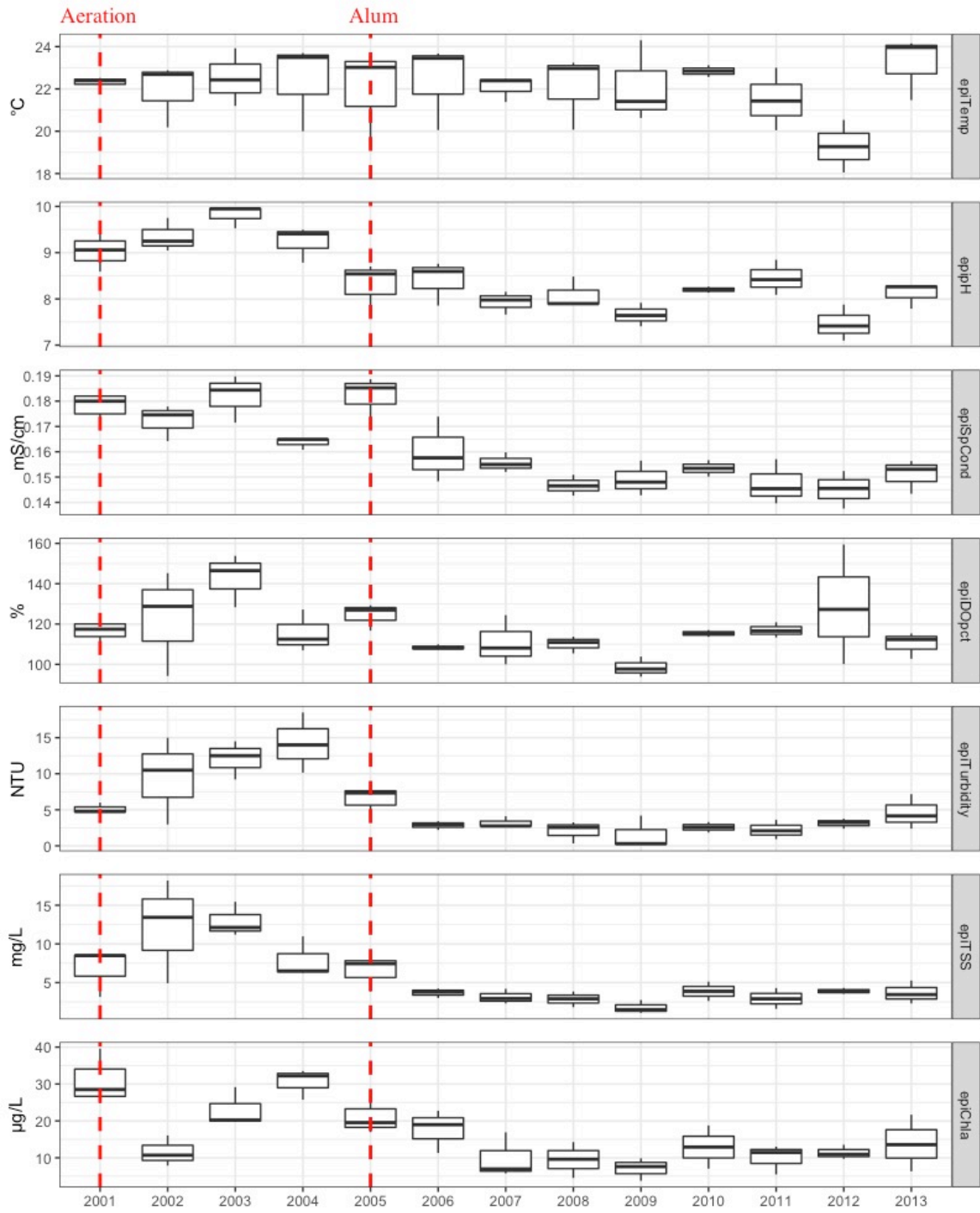
Figure 4: C*ontinued*. (D) other epilimnetic water quality variables.

(E)

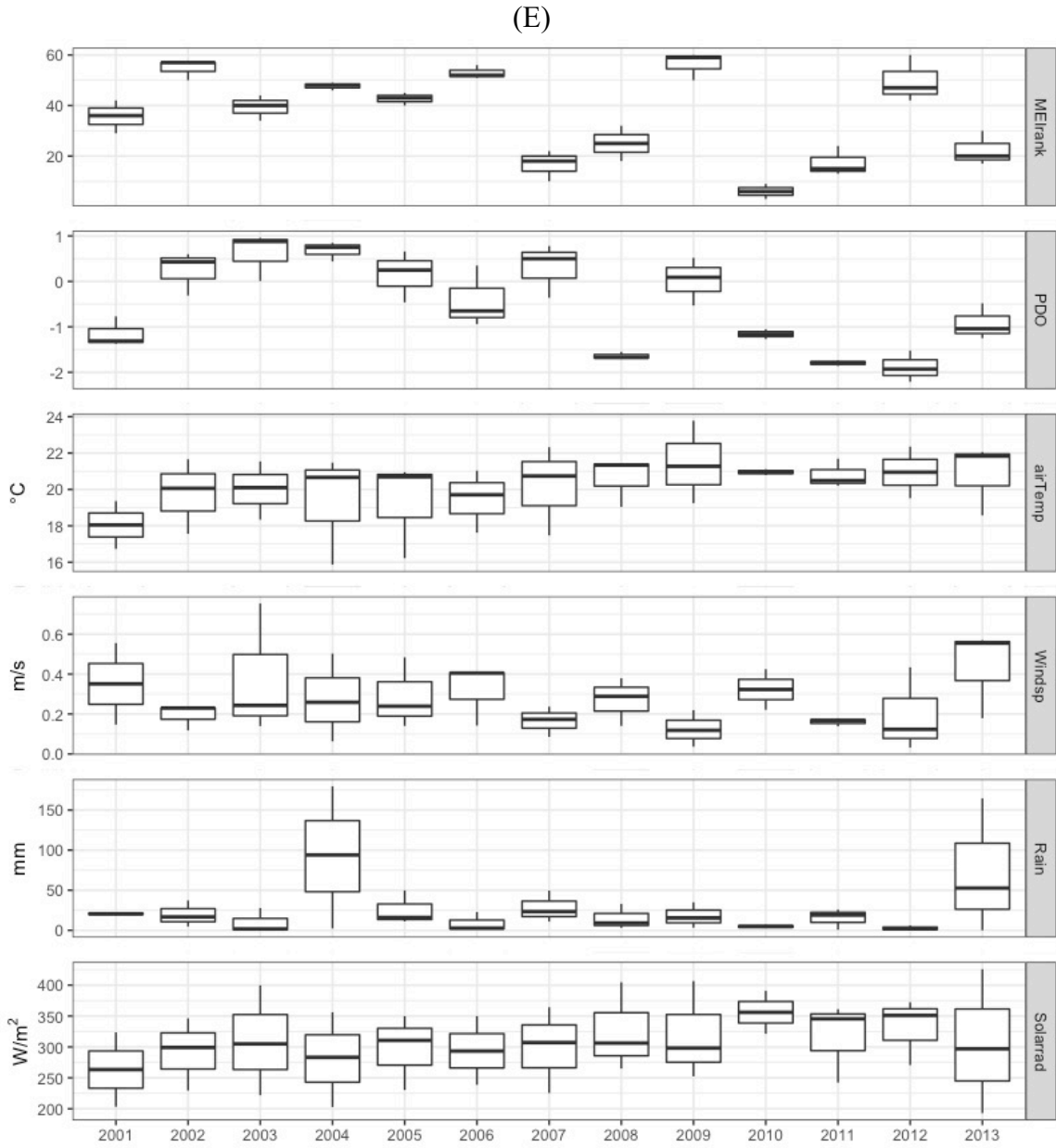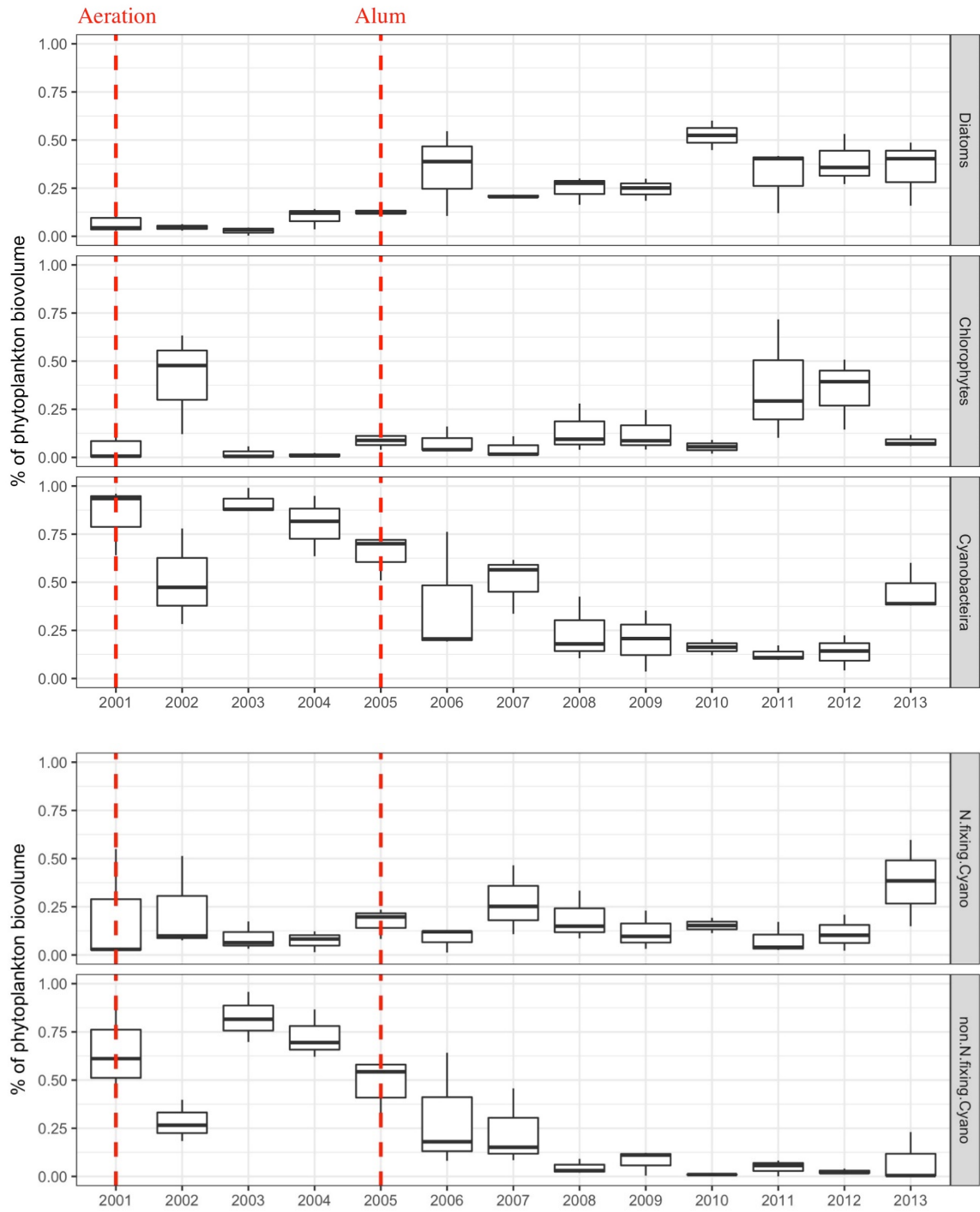Figure 4: C*ontinued*. (E) climatic and meteorological variables.

Figure 5: Boxplots showing year-to-year variations of the phytoplankton assemblages among the major taxonomic groups between July and September from 2001 to 2013 in the deep basin of Oswego Lake.
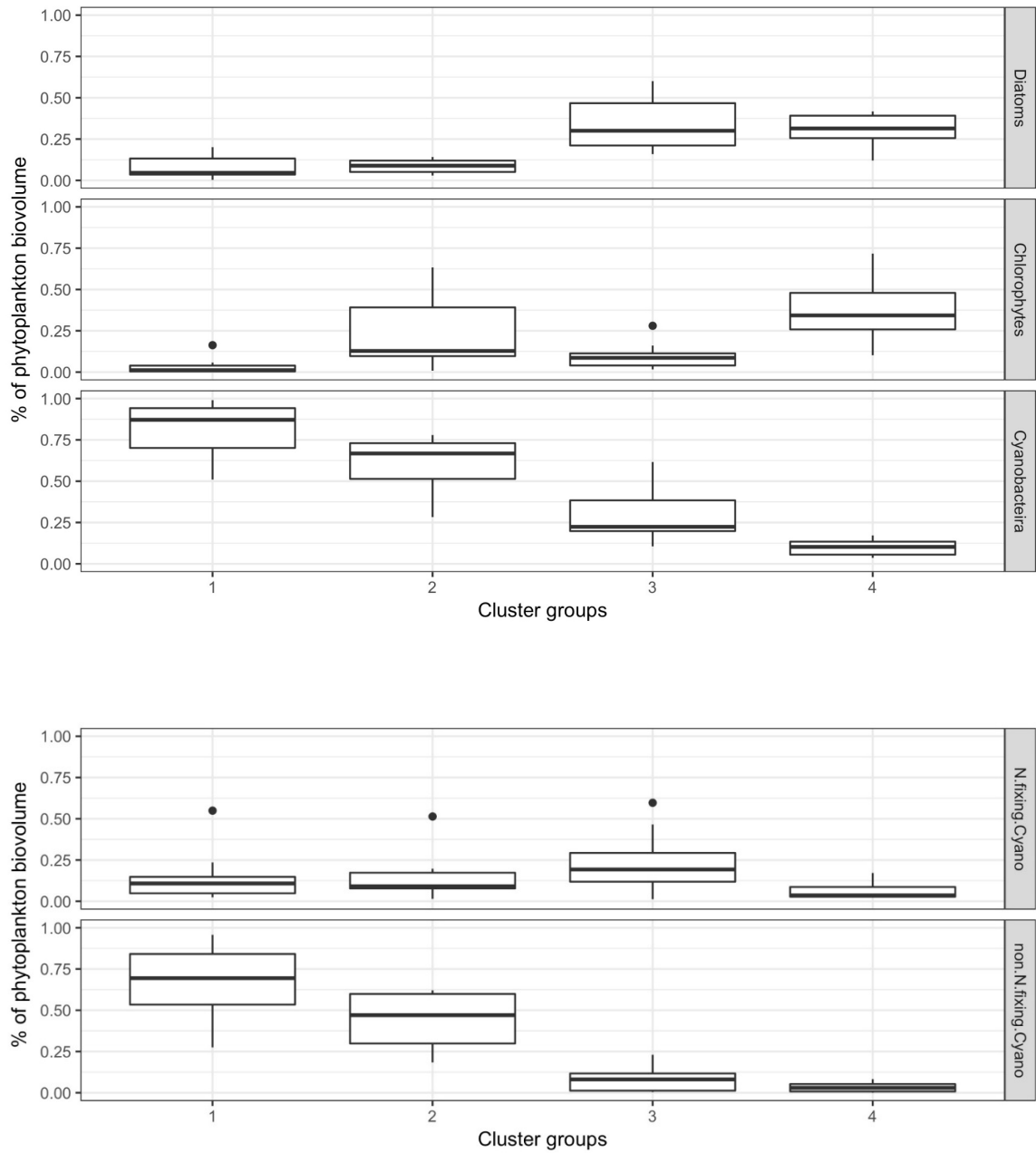
Figure 6: Boxplots showing comparison of relative abundance of the phytoplankton taxonomic groups among 4 cluster groups based on the cluster analysis.
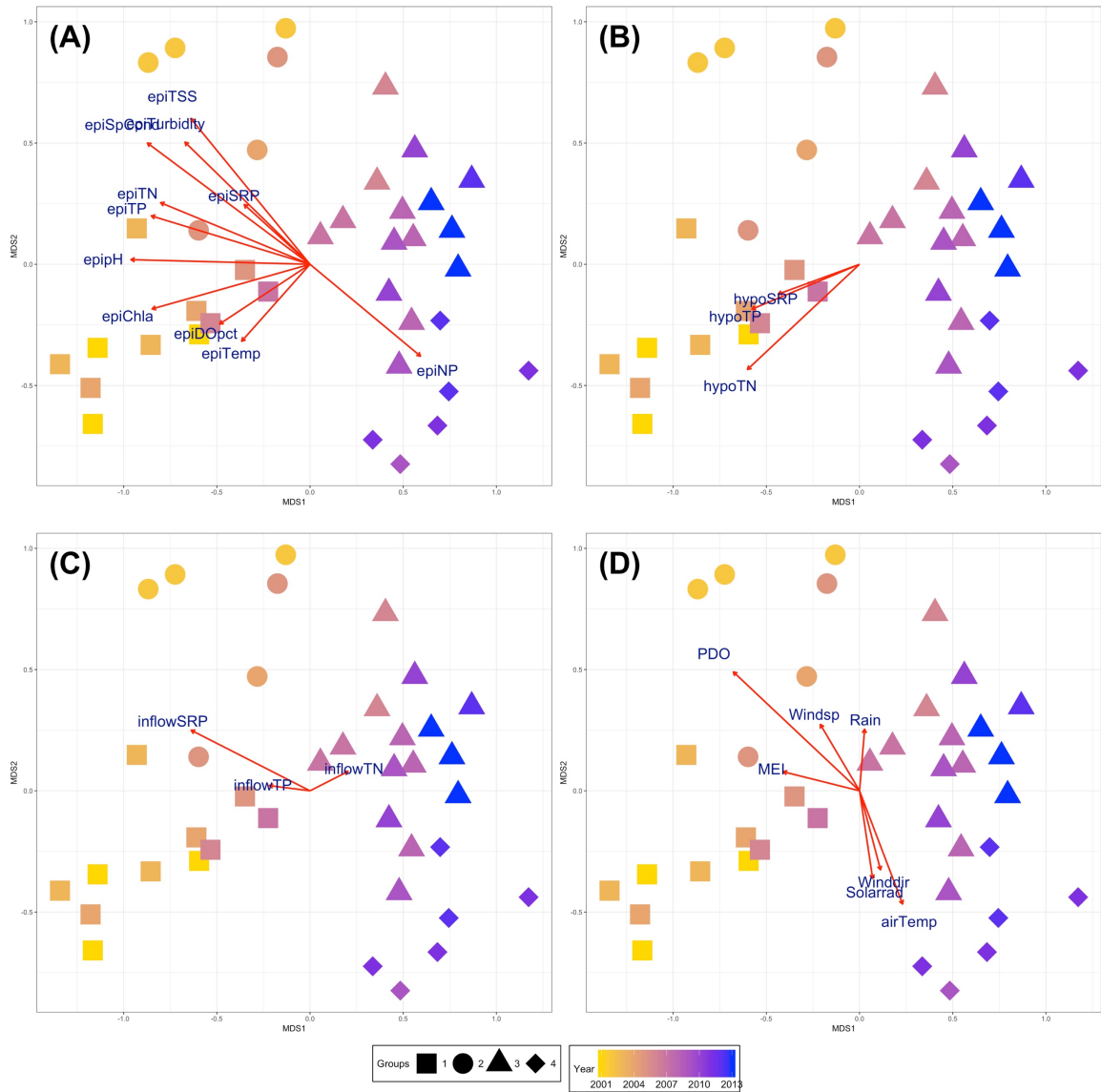
Figure 7: Plots of non-metric multidimensional scaling (NMDS) analysis showing temporal variation of the phytoplankton assemblages between July and September from 2001 to 2013 in the deep basin of Oswego Lake, superimposed with the cluster groups and with relation to major environmental variables of epilimnion (A), hypolimnion (B), Tualatin River inflow (C), and climate and meteorological conditions (D). Figure continued on the next page.
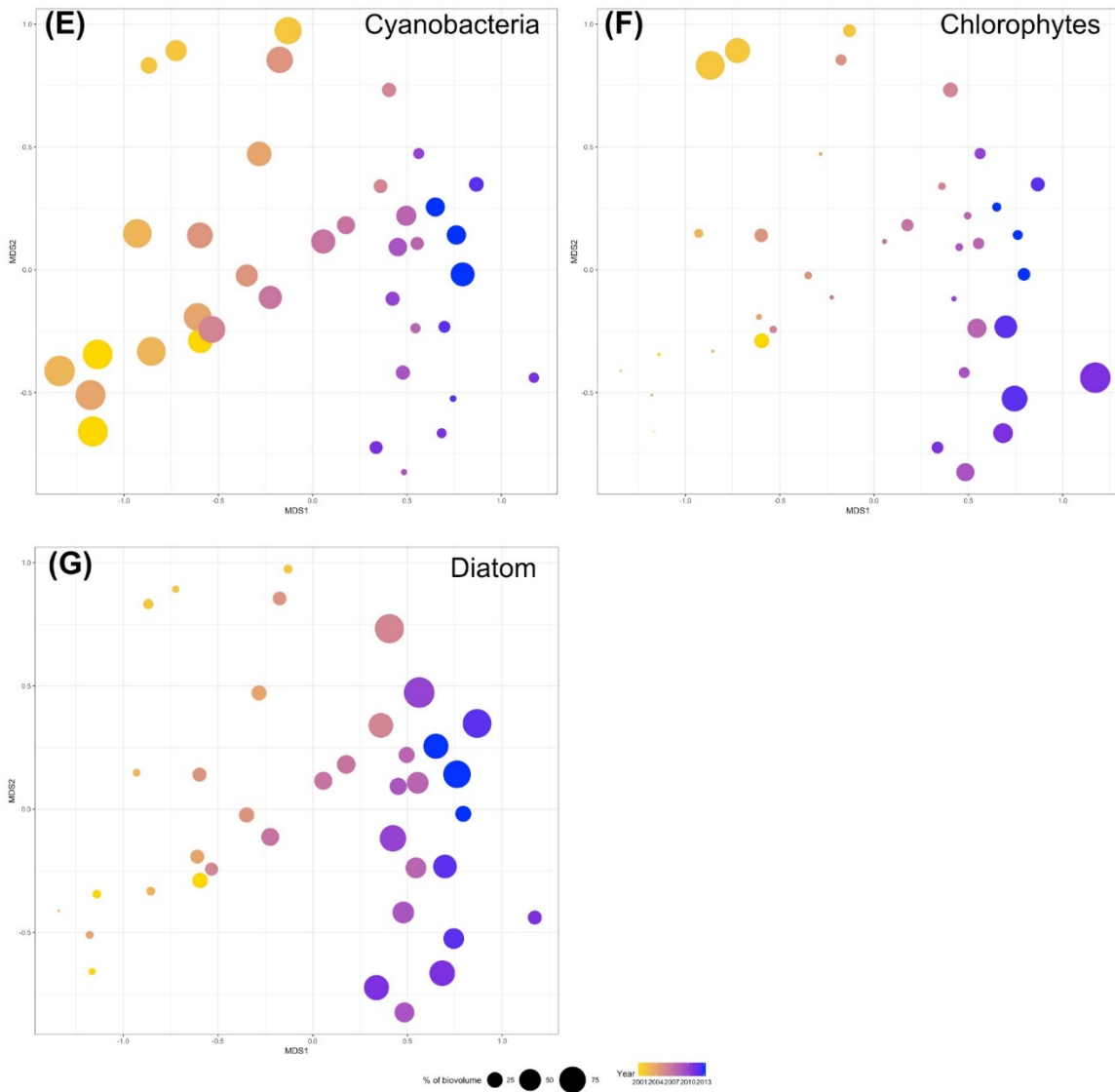
Figure 7 *Continued*. (E)-(G) are the same NMDS plot superimposed with % of cyanobacteria (E), chlorophytes (F) and diatoms (G) in each sample (bubble size is proportional to the relative abundance of total phytoplankton biovolume).

Figure 8: Timeline of the lake management practices and the occurrence of the cluster groups based on phytoplankton assemblages between July and September from 2001 to 2013. Circumscribed numbers (7, 8, & 9) = months (July, August and September)

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Appendices

Appendix A: Water quality parameters between 2001 and 2013 in the deep basin of Oswego Lake.

Variable	Year 2001-2013 (n_stratification=13; n_others=224)					July-September 2001-2013 (n=76)				
	Minimum	Median	Mean	Maximum	Variance	Minimum	Median	Mean	Maximum	Variance
Stratification onset	Mar 16 (2005)	Apr 20	Apr 16	May 24 (2011)						
Stratification breakup	Aug 21 (2012)	Sep 14	Sep 15	Oct 3 (2001)						
Stratification duration	119 (2012)	161	152	182 (2005)						
Water temp (°C)	2.1	16.5	15.3	24.5	42.2	17.3	22.6	22.0	24.5	3.3
pH	6.7	8.1	8.2	10.0	0.5	7.1	8.3	8.4	10.0	0.6
Percent of dissolved oxygen (%)	5.3	111.5	109.9	185.1	563.3	72.6	115.6	117.9	179.2	357.6
Special conductance (mS/cm)	0.1	0.1	0.2	0.2	0.0	0.1	0.2	0.2	0.2	0.0
Total suspended solides - Epilimnion (mg/L)	0.4	3.8	4.7	25.6	11.8	0.9	3.9	5.5	25.6	17.8
Turbidity (NTU)	0.0	2.8	3.6	22.0	11.8	0.0	3.4	5.2	22.0	21.3
Total Nitrogen - Epilimnion (ug/L)	201.0	510.3	587.3	1690.0	69573.0	201.0	424.1	449.4	982.0	22117.3
Total Phosphorus - Ephilimnion (ug/L)	13.0	31.5	39.6	130.0	562.1	13.0	28.5	37.9	129.0	673.6
Soluble Reactive Phosphorus -Epilimnion (ug/L)	0.0	1.0	4.7	63.0	100.6	0.0	1.0	2.7	37.0	40.5
Nitrogen:Phosphorus	4.6	15.7	17.9	73.5	71.9	3.9	13.3	15.2	44.3	61.2
Chlorophyll-a (ug/L)	0.9	12.5	16.0	79.5	144.5	2.3	12.6	15.6	39.6	94.3
Volume Phytoplankton (um3/ml)	310,956	6,988,859	10,011,225	44,623,586		841,870	7,533,211	9,566,173	39,727,119	
Volume Cyanophyta (um3/ml)	0	558,592	2,134,063	22,663,619		41,233	2,303,213	4,633,541	22,663,619	
% volume Cyanophyta of total phytoplankton	0	9.4	23.1	99.4		1.5	44.1	44.9	99.4	
Volume N-fixer Cyanophyta (um3/ml)	0	209,648	674,021	10,622,061		6,649	791,536	1,318,434	10,622,061	
% volume N-fixer of total phytoplankton	0	3.5	9.6	82.3		0.2	11	16.6	82.3	
Volume non-N-fixer Cyanophyta (um3/ml)	0	77,679	1,460,041	21,849,728		0	591,138	3,315,107	21,849,728	
% volume non-N-fixer of total phytoplankton	0	1.3	13.5	96.8		0	11.3	28.3	96.5	

Appendix B: Field sampling and laboratory procedures.

Appendix B1: Total suspended solids

<u>Field sampling</u>
- Use Kemmerer sampler to collect water from specific depth(s)
- Fill 1 L bottle with sample
- Refrigerate when you return to marina until processing

<u>Preparation of glass fiber filter</u>
- Label aluminum cups, 1-10
- Install aspirator to faucet and connect via hose to flask
- Place rubber stopper end of magnetic filtering stage inside neck of filter flask
- Place glass fiber filter disk (Whatman 934-AH filter) wrinkle side up on filtering stage
- Place magnetic filtering cup over filter
- Turn on water to apply vacuum and wash disk with three successive 20 mL portions of D.I. water
- Continue vacuum to remove as much water as possible (3 minutes)
- Stop vacuum, remove filtering cup, and use filter forceps to transfer filter from the filtering stage to numbered aluminum cup in drying oven
- Repeat process until 9 filters have been prepared
- Dry in oven at 103º C for at least 1 hour
- Turn off oven and allow filter papers to reach room temperature (at least 1 hour, typically 90 minutes)
- Use forceps to move cooled cup and filter from oven to electric balance
- Record start weight on water quality lab worksheet

<u>Filtering sample</u>
- Set up filtering apparatus as outlined above
- Remove filter from aluminum cup and place on filtering stage
- Shake water sample to thoroughly homogenize
- Slowly turn on water to apply vacuum
- Start by pouring 300 mL of mixed sample into filtering apparatus
- Repeat until flow starts to slow
- Record filtered volume
- Wash graduated cylinder with 3 successive 10 mL volumes of D.I. water and pour each into filtering apparatus
- Wash interior of filtering cup with one 10 mL rinse of purified water

- Continue suction for at least 1-3 minutes after filtration is complete. If the filter is being rewetted from splash back stop the vacuum, remove the stopper and sample from neck of flask, pour out water from flask, replace the stopper and sample, and reapply vacuum to continue filtering
- Stop vacuum, remove filtering cup, and use filter forceps to transfer filter from the filtering stage to original numbered aluminum cup in drying oven
- Repeat process for all samples
- Dry for at least one hour at 103º C
- Turn off oven and allow filter papers to reach room temperature (at least 1 hour, typically 90 minutes)
- Use forceps to move cooled cup and filter from oven to electric balance
- Record final weight on lab sheet

Appendix B2: Chlorophyll *a*

Field data collection and filtering
- Place GN-6 filter2 on filter plate and screw on filter funnel, attach hand vacuum pump to filter apparatus
- Use Kemmerer to collect water from specific depth
- Use graduated cylinder to measure 500 mL of sample water, add 0.5 mL saturated magnesium carbonate solution3
- Pour 300-500 mL of sample into filter funnel (volume will depend on particulate concentration)
- Do not exceed a vacuum of 15 inches mercury during filtration
- Rinse graduated cylinder with 10 mL volume of distilled water and pour into filter funnel
- Rinse walls of filter apparatus with distilled water
- Remove filter from plate with filter forceps, fold into quarters lengthwise and place in centrifuge tube
- Place tubes in cooler with ice pack
- Rinse graduated cylinder and filter apparatus with distilled water after each use
- Collect 1 QA/QC chlorophyll *a* sample from Outlet for ARI
  - Follow procedures through remove filter from plate with filter forceps
  - Place filter (UNFLODED) in sterile petri dish. Label, cover in foil. Ship with other samples to ARI
- Place tubes in a test tube rack and store in marina freezer with caps off under a towel until processed

Laboratory analysis
- Perform all work in subdued light to avoid chlorophyll degradation
- Fill centrifuge tube to the 10 Ll mark with 90% aqueous acetone4
- Sonicate sample for 10 seconds, rest for several seconds, and then sonicate for another 10 seconds
- Place sample in ice water, shaded from the light (under a towel)
- Repeat previous two steps for each sample, wiping off sonicator between samples with 100% acetone
- Allow samples to steep in ice water for 2 hours
- Remove from ice water and centrifuge samples for 20 minutes

Spectrophotometric Determination of Chlorophyll *a*
- Turn on spectrometer and allow to warm up at least 10 minutes prior to using
- Choose User Programs then Chl a

- Fill cuvette with 3 mL 90% aqueous acetone and place into spectrometer well, frosted sides facing you and black number aligned with well arrow
- Zero spectrometer by pressing Zero. Remove cuvette and place aside for additional zeros
- Pipette 1.5 mL of 90% aqueous acetone into the other empty cuvette
- Pipette 1.5 mL from sample tube into the same cuvette
- This dilutes sample to 19.5mL (the filter accounts for an additional 0.5 mL)
- Agitate cuvette gently then wipe sides with a Kimwipe
- Place cuvette into spectrometer well with the frosted sides facing you
- Press Read, record optical density at 750 nm and 664 nm on WQ lab worksheet
- Remove cuvette and add 0.1 mL (~3 drops) 0.1N HCL, agitate gently, and let stand for 90 sec (use timer)
- The sample will be a little cloudy initially, make sure it is completely clear before reading again
- Return cuvette to spectrometer well, frosted sides facing you and black number aligned with arrow
- Press Read, record optical density at 665 nm and 750nm on WQ lab worksheet
- Empty cuvette into waste acetone container and rinse glass cuvette with 100% acetone
- Repeat for all samples, re-zeroing spectrometer after every three samples using cuvette filled with 3mL 90% aqueous acetone

Appendix B3: Phytoplankton

Field Collection
- Collect phytoplankton sample using Kemmerer sampler from designated depth
- Rinse bottle with sample water and fill completely
- Add 3 mL Lugols solution1 per liter of sample water
- Close tightly and keep in cool, dry location until shipped (marina refrigerator)

Laboratory Procedures
- Place a GN-6 filter on top of filter apparatus support plate
- Connect tubing to water aspirator and turn water on slowly
- Shake sample water
- Rinse 100 mL graduated cylinder with sample water
- Shake sample water again
- Fill graduated cylinder with 50 mL of sample
- Add six drops of lugols to graduated cylinder
- Filter the 50 mL of sample and lugols
- Place filter paper in tent of aluminum foil labeled with sample location, date, and volume filtered
- Allow filter to dry for at least 2 days
- Place clean slide on paper towel
- Label slide with sampling station, date, and volume filtered (50 mL usually)
- Cut out square from a corner of the filter paper inside the X created by filtering support plate
- Put oil on slide and move around until entire filter paper is covered
- Pop any bubbles formed by oil
- Cover with cover slip
- Paint lots of clear nail polish around edges of cover slip
- Place in cabinet to dry
- Count 80-100 algae by moving frames
- Keep track of number of frames, algal species and power used

Taxonomic analyses of phytoplankton samples by WES
(WATER Environmental Services, Inc.)

Microscopic methods, provided on 26 February 2013 by Maribeth V. Gibbons, Pres.
WATER Environmental Services, Inc.

Phytoplankton identifications, enumerations, and cell volume determinations were made
on lake water samples collected by Lake Oswego staff for each study date. All samples
were preserved in the field with standard Lugol's (iodine) Solution and kept cool prior to
shipping to WATER Environmental Services, Inc. for microscopic taxonomic analyses.

Taxonomic analysis was performed on a single 1.0 mL subsample of each well-mixed
lake sample using a Sedgewick-Rafter counting chamber and Leitz compound
microscope (@100X, 400X magnification). The subsample volume held within the S-R
chamber was allowed to settle for at least 15 minutes prior to microscopic viewing.
Initially, the viewing chamber was scanned at low (40X) magnification to confirm an
even distribution of algal cells before intensive identification @100 X power. For
routinely processed samples, a transect counting methodology was used in which
successive horizontal sweeps of the S-R chamber were made under 100X power so that
the entire 1 mL subsample volume was analyzed. The entire chamber is covered by 11
transects with each transect composed of 28 fields of view, resulting in a total of 308
fields of view. For samples containing high cell densities of the most common form, (i.e.,
greater than 50 cells/colonies within one transect pass), at least one half of the volume of
each 1 mL subsample was counted, resulting in a total of 154 fields of view. In that case,
the entire 1 mL S-R chamber was also analyzed to enumerate all rare and very large
forms, like Ceratium sp. Only algal cells presumed to be alive at the time of sampling
(chloroplast reasonably intact in preserved sample) were counted. Replicate subsamples
of a single sample selected from a group of 25 samples were analyzed as a statistical
check for counting precision (e.g., Coefficient of Variation was sufficiently small).
Average counts for each phytoplankton taxon were computed from subsample results,
using appropriate multiplying factors if dilutions are necessary. Algal densities were
typically reported in natural units as numbers of cells or colonies per mL.

For each sample, cell dimensions of at least 10 organisms of each phytoplankter were
computed to obtain average cell volume per taxon based on geometric shape. Cell
volumes for each taxon were calculated for each lake sample analyzed. Determination of
cell volume dimensions and identifications were made with a calibrated whipple disc at
400X (high dry magnification) using a Palmer-Maloney nannoplankton chamber (0.1 mL
volume) or at 1000X(oil emersion). Cell volumes were reported as cubic microns per mL,
and were converted to cubic millimeters per liter (mm^3 /L) for report presentation.
Phytoplankton identifications were made to at least genus level wherever possible.

Species identifications were conducted primarily according to Prescott (1975, 1980), Patrick and Reimer (1966, 1975), Smith (1950), and Wehr and Sheath (2003). Recent journal article sources were also consulted for certain Cyanobacterial identifications due to current revisions in the systematics of this group.

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Appendix C: Wind rose diagram showing wind speed and wind direction across day time (5am - 9pm) between July and September from 2001 to 2013 at the weather station at Oswego Lake.
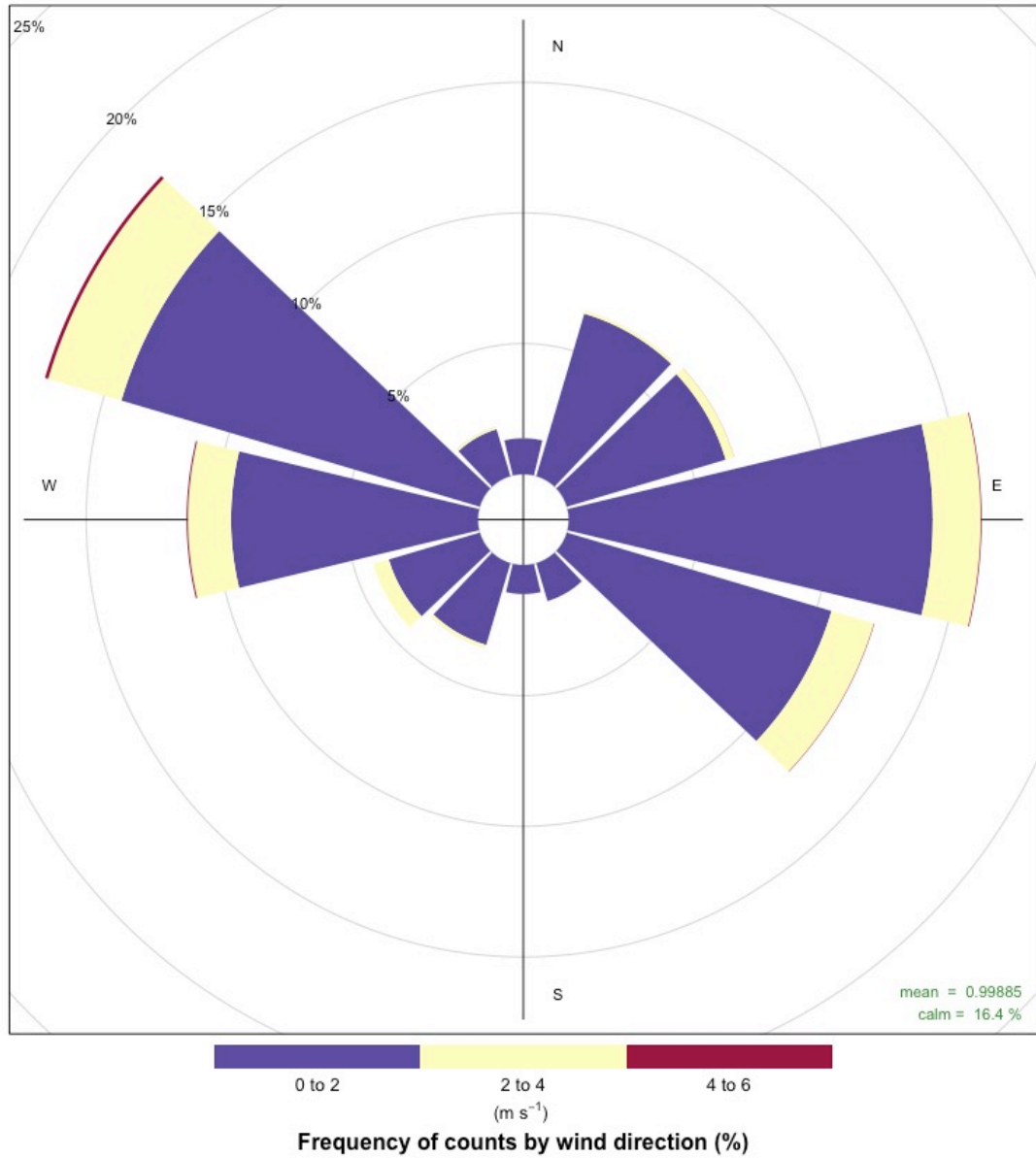
Appendix D: List of phytoplankton taxa and divisions.
(The phytoplankton species included in this study were underlined.)

Division	Taxa	Division	Taxa
Bacillariophyta		Chlorophyta	
	Acanthoceras sp.	*(cont.)*	***Coelastrum* sp.**
	Amphora sp.		*Crucigenia irregularis*
	Asterionella formosa		*Crucigenia* sp.
	Cocconeis sp.		*Dictyosp.haerium* sp.
	***Cyclotella* sp.**		***Eudorina elegans***
	Cymbella sp.		***Eudorina* sp.**
	Diatom sp.		*Kirchneriella* sp.
	Diatom sp. *(pennate)*		*Micractinium pusillum*
	Epithemia sp.		***nannoplankton***
	Eunotia sp.		*Nephrocytium* sp.
	Fragilaria crotonensis		*Oocystis Borgei*
	***Fragilaria* sp.**		*Oocystis crassa*
	Gyrosigma sp.		***Oocystis Eremosphaeria***
	Melosira granulata		***Oocystis* sp.**
	***Melosira* sp.**		*Palmellaceae (nanno)*
	Melosira varians		***Palmellaceae (sph)***
	Navicula sp.		***Pandorina morum***
	Nitzschia sp.		***Pandorina* sp.**
	Stephanodiscus niagarae		*Pediastrum Boryanum*
	***Stephanodiscus* sp.**		*Pediastrum duplex*
	Synedra cyclopum		*Pediastrum simplex*
	Synedra sp.		*Pediastrum tetras*
	Synedra ulna		*Pleodorina* sp.
	Tabellaria fenestrata		*Pseudosp.haerocystis* sp.
	Urosolenia sp.		*Quadrigula* sp.
			Scenedesmus arcuatus
Chlorophyta			*Scenedesmus bijuga*
	Ankistrodesmus falcatus		*Scenedesmus dimorphus*
	Ankistrodesmus sp.		*Scenedesmus quadricauda*
	Asterococcus limneticus		*Scenedesmus* sp.
	Asterococcus sp.		*Schroederia* sp.
	***Botryococcus* sp.**		***Sphaerocystis schroeteri***
	Chlamydomonas sp.		*Tetraedron limneticum*
	Closteriopsis longissima		*Tetraedron* sp.
	Closteriopsis sp.		***undetermined***
	Coelastrum microporum		*Volvox* sp.

Appendix D: *Continued*

| Division	Taxa	Division	Taxa
Cyanobacteira		Charophyta	
			Closterium sp.
N-fixers			***Cosmarium* sp.**
	Anabaena circinalis		*Staurastrum chaetoceros*
	Anabaena flos-aquae		***Staurastrum paradoxum***
	Anabaena limnetica		*Staurastrum pingue*
	Anabaena planctonica		***Staurastrum* sp.**
	***Anabaena* sp.**		*Zygnematales*
	Anabaena spiroides		
	Aphanizomenon flos-aquae	Chrysophyta	
	Aphanizomenon issatschenkoi		*Chrysophyte (colonial)*
	Aphanizomenon sp.		*Chrysophyte (filamentous)*
	Aphanocapsa Grevillei		***Chrysophyte (unicell)***
	Aphanocapsa sp.		*Dinobryon bavaricum*
	Gloeotrichia echinulata		***Dinobryon divergens***
	Nostocaceae		***Dinobryon* sp.**
			***Mallomonas* sp.**
non-N-fixers			*Rhizochrysis limnetica*
	Anacystis sp.		***Rhizochrysis* sp.**
	Chroococcaceae (colonial)		***Synura* sp.**
	Chroococcus sp.		
	Coelosp.haerium Naegelianum	Cryptophyta	
	***Coelosphaerium* sp.**		***cryptomonad***
	Cyanophyte (colonia)		***Cryptomonas erosa***
	Cyanophyte (unicell)		***Cryptomonas ovata***
	Gomphosp.haeria lacustris		***Cryptomonas* sp.**
	Gomphosp.haeria sp.		***Rhodomonas* sp.**
	***Lyngbya* sp.**		
	Merismopedia sp.	Euglenophyta	
	Microcystis aeruginosa		***Trachelomonas* sp.**
	***Microcystis* sp.**		*Trachelomonas volvocina*
	Oscillatoria sp.		
	Oscillatoriaceae	Pyrrophycophyta	
	Oscillatoriales		***Ceratium hirundinella***
	Snowella sp.		***dinoflagellate***
	***Woronichinia* sp.**		*Peridiniales*
			Peridinium sp.

Appendix E: Phytoplankton relative abundance between July and September from 2001 to 2013 in the deep basin of Oswego Lake.
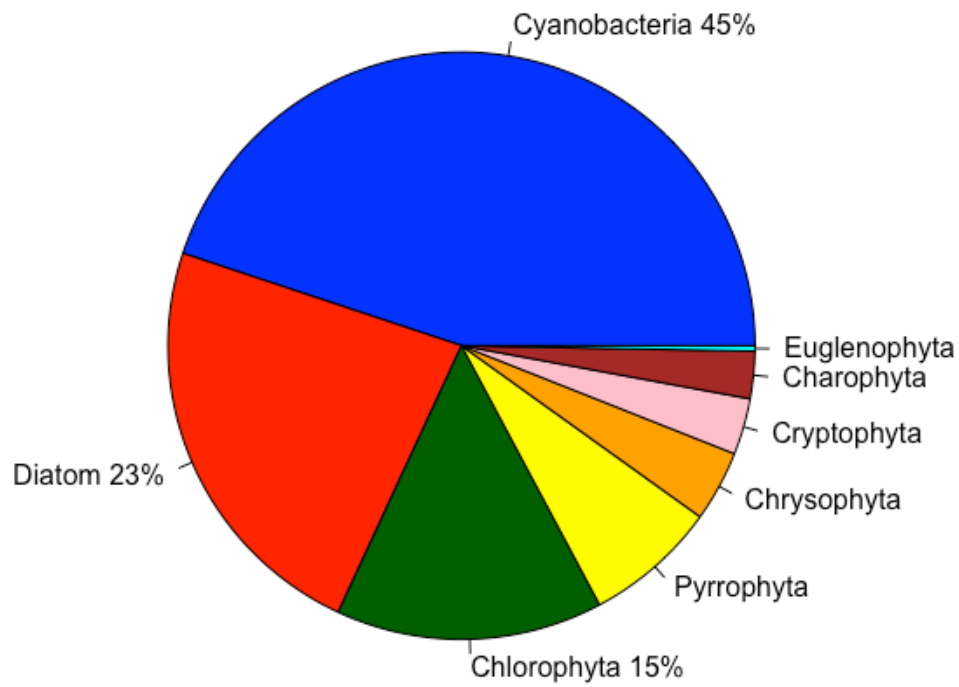
Appendix F: Cluster dendrogram of phytoplankton assemblages showing four groups.

Cluster Dendrogram

dis.sp
hclust (*, "ward.D2")
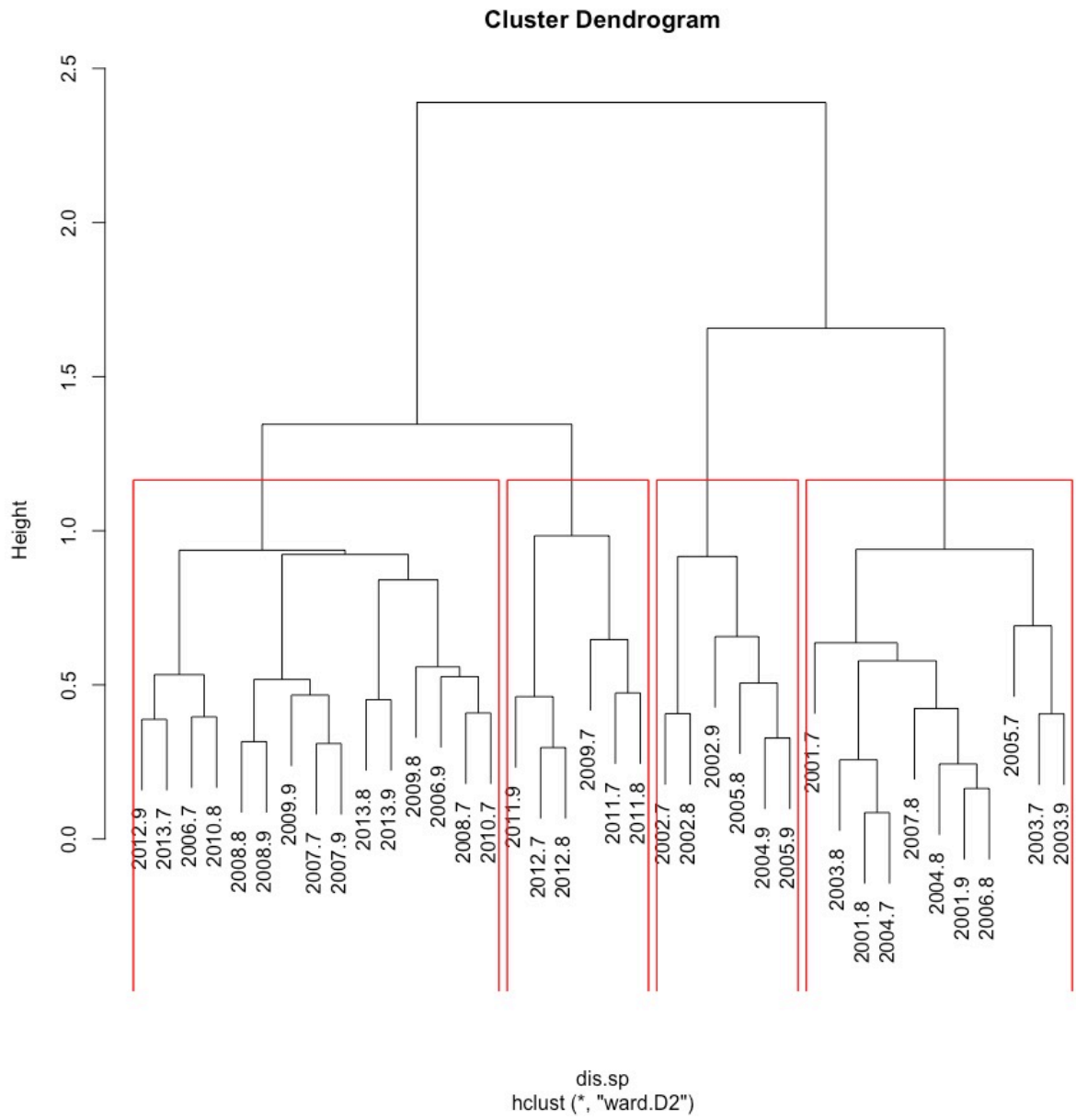
Appendix G: Boxplots showing comparisons of the July-August-September means of epilimnetic TP, SRP and chlorophyll *a* for pre- and post-initial alum treatment in 2005. All differences were significant (*p*<0.05) based on Welch Two Sample t-test.
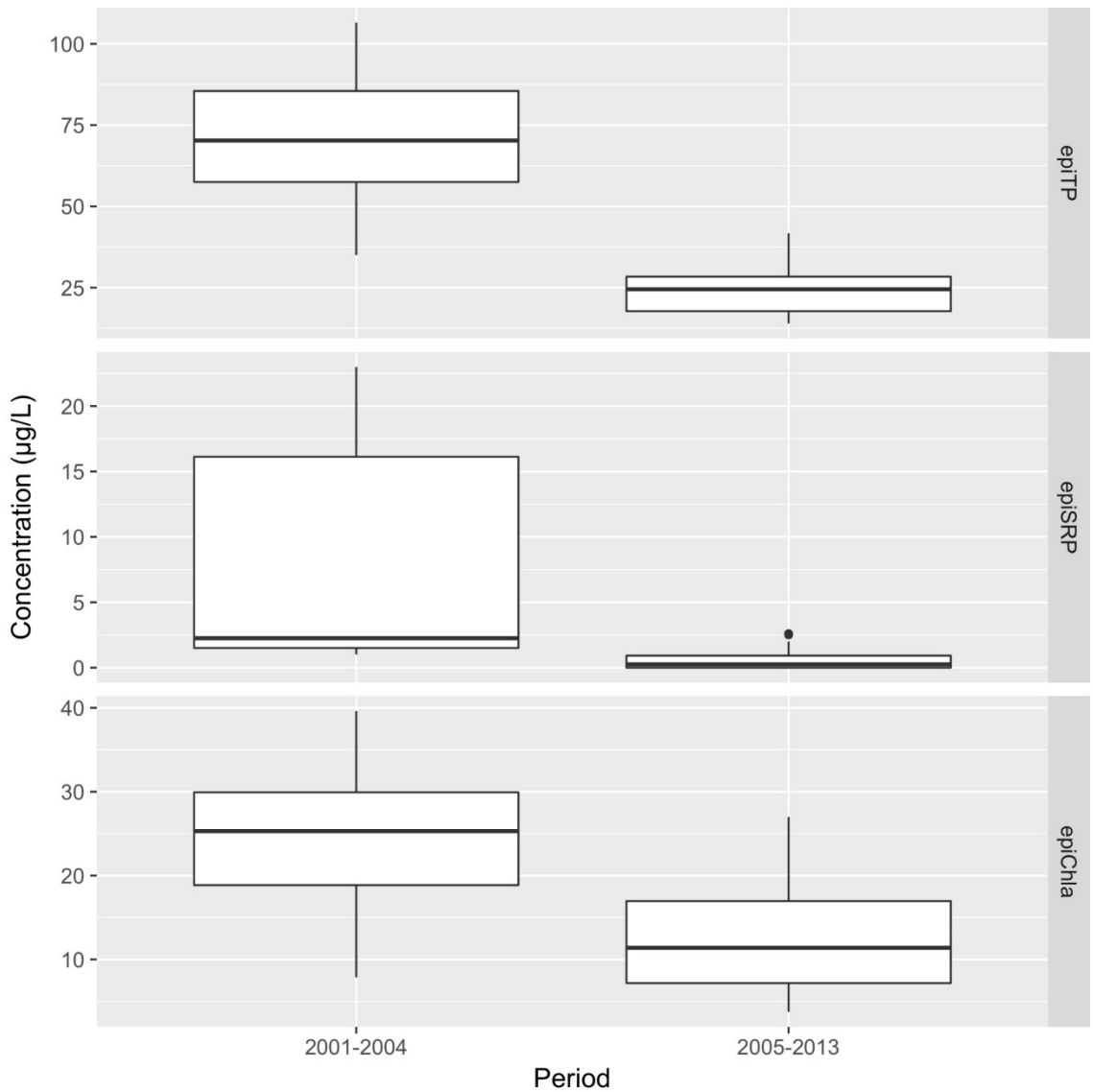
Appendix H: Time series of the multivariate ENSO index (MEI) and Pacific Decadal Oscillation (PDO) index between July and September from 2001 to 2013.

The stacked bar plot of MEI and PDO indices was generated based on Gershunov and Barnett (1998). They demonstrated that PDO seems to have a modulating effect on ENSO teleconnections in North America. They showed that typical El Niño patterns are strong and consistent only during the positive phases of PDO and typical La Niña patterns are consistent only during the negative phases of PDO.
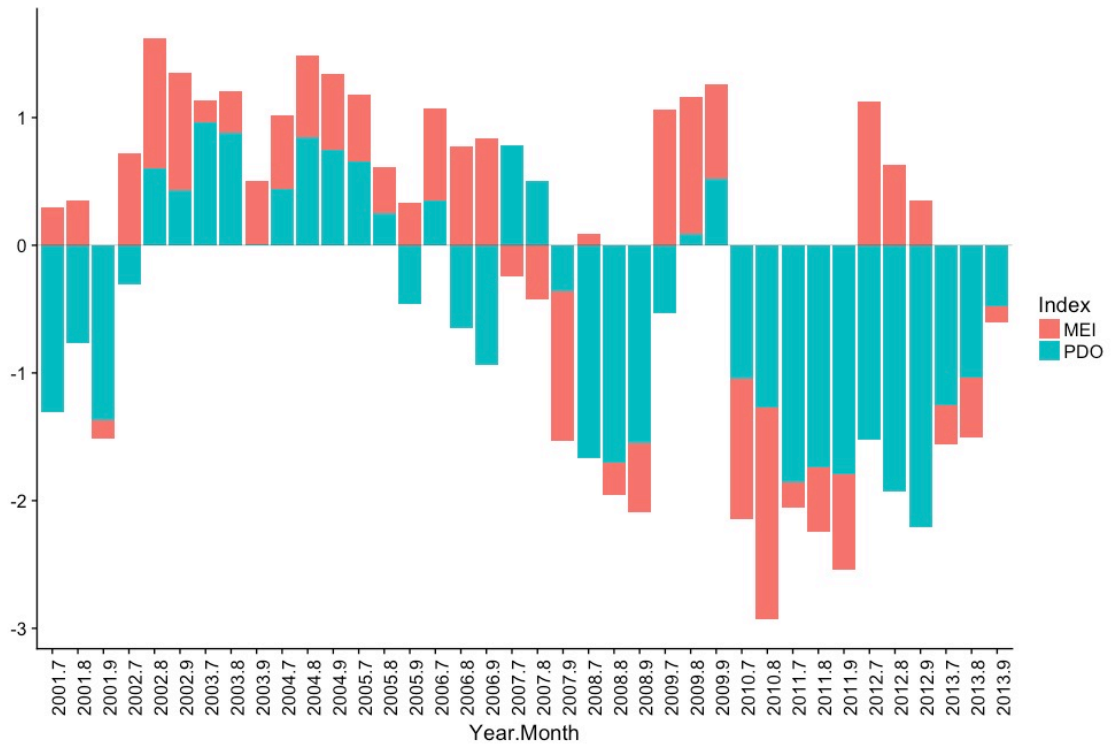
Reference:

Gershunov, A., & Barnett, T. P. (1998). Interdecadal modulation of ENSO teleconnections. *Bulletin of the American Meteorological Society*, *79*(12), 2715-2726.

Appendix I: Time series of the multivariate ENSO index (MEI), showing the study period between 2001 and 2013 and two extreme El Niño events (1997-1998 and 2015-2016).
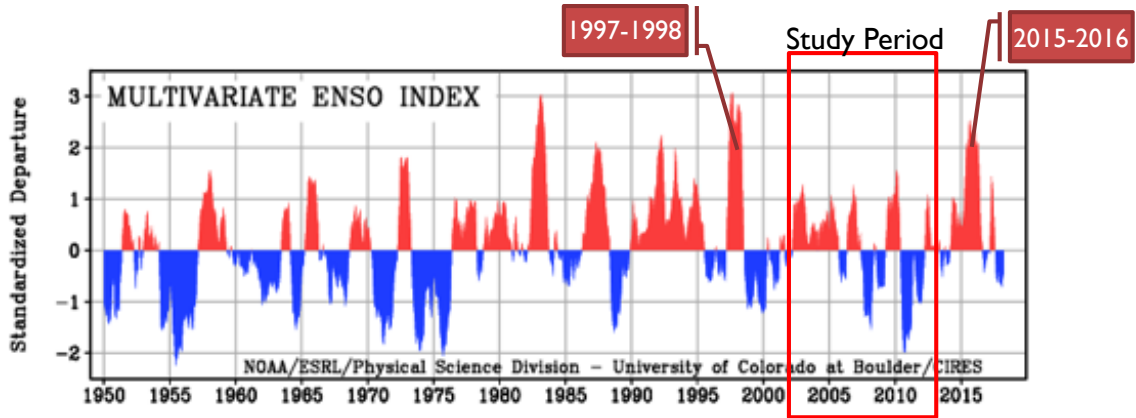
Appendix J: Boxplots of cyanobacteria, epilimnetic water temperature and rainfalls between July and September from 2001 to 2013.

2004 *Microcystis* bloom