

# Source analysis of nutrient loading and their impact on cyanobacteria dynamics in a hyper-eutrophic freshwater system, Lake Agawam, Southampton, NY, USA.

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## Abstract

Nuisance cyanobacteria blooms have plagued Lake Agawam for a number of years. Current monitoring has shown that during peak bloom months (May through October), blooms are dominated by *Mycrocystis* spp. and become nitrogen limited. Recent studies have found toxins produced by *Mycrocystis* spp. within the lake and fish kills from low oxygen levels have also been documented after blooms crash. A nutrient budget was constructed to determine the point and non-point sources for nutrients entering the lake to assist with guiding remediation efforts. Dissolved nitrogen was found to primarily originate from groundwater with high levels also originating from a storm drain at the north end of the lake and benthic flux. Phosphorus inputs, both organic and inorganic, mainly originated from the benthos. These findings suggest that remediation efforts should focus on relieving groundwater inputs but as this can often prove too costly or politically difficult, significant changes can be made by reducing storm water input and possible dredging of the lake bottom. Nutrient dilution experiments suggest that reducing nutrients will not only reduce algal biomass, but may also yield a shift in the phytoplankton community away from cyanobacteria and toward non-phycoerythrin containing-species. Since cyanobacteria blooms are the cause of many environmental problems in Lake Agawam, including fish kills, a focus on nutrient reduction should be a part of any future management plan for Lake Agawam.

## Introduction

The eutrophication of coastal and inland waters continues to pose economic and health risks to communities around the world. Eutrophication is the result of elevated supplies of nutrients to surface waters which results in increased productivity of primary producers, particularly phytoplankton and aquatic plants (Prepas & Charette 2003). There can be many external point and nonpoint sources of nutrients to waters including wastewater effluent, storm sewer outfalls, agricultural and urban runoff and atmospheric deposition (Smith et al. 1999). In fresh-water lakes and slow moving rivers, excess nutrients and resulting phytoplankton blooms can leave waters turbid, foul smelling and often devoid of oxygen.

One of the most prevalent algal species in fresh water systems is cyanobacteria. Cyanobacteria are a large and diverse group of prokaryotic photosynthetic organisms, which can be found in marine, freshwater, and terrestrial environments. Cyanobacteria blooms in freshwater systems are typically associated with eutrophic and poorly flushed waters (Paerl 1988, Philipp et al. 1991, Carmichael 1994, Oliver & Ganf 2000, Paerl et al. 2001). While it is clear that the occurrence of toxic cyanobacteria blooms around the world have increased during recent decades (Chorus 1999a), the underlying causes of such blooms and their increased frequency are poorly understood.

Blooms formed by toxic cyanobacteria can include the genera *Anabaena*, *Aphanizomenon*, *Nodularia*, *Oscillatoria*, and *Microcystis* (Carmichael & Falconer 1993, Carmichael 1994, Chorus 1999a, Fleming et al. 2002). Cyanotoxins associated with these genera fall into two broad categories; hepatotoxins and saxitoxins. Microcystins and cylindrospermopsins are cyclic peptides (hepatotoxins) which inhibit eukaryotic protein serine/threonine phosphatases (Honkanen et al. 1990, Chorus 1999a). Anatoxin-a, anatoxin-a(s), and saxitoxins are three types of neurotoxins which block neuronal signal transmission and can lead to the paralysis of the heart and lungs resulting in rapid death in terrestrial vertebrates (Duy et al. 2000, Briand et al. 2003).

Of all the known cyanotoxins, microcystins are the most widespread and common. They are produced in the majority of blooms formed by *Microcystis* spp., but also can be produced by *Anabaena* spp.,

*Oscillatoria/Planktothrix* spp., and *Nodularia* spp. (Chorus 1999a). Microcystin toxicity can accumulate within the liver of vertebrates causing chronic liver damage (Falconer et al. 1988, Fitzgeorge 1994). They bind irreversibly to protein phosphatases 1 and 2A which play a key role in maintaining homeostasis in the cell (Cohen 1989, Chorus 1999a). The inhibition of these compounds can lead to increased phosphorylation of tumor suppressor proteins which can result in increased signaling and promote cell proliferation, transformation, and tumor promotion (Fujiki & Suganuma 1993, Chorus 1999a).

Human interaction with cyanotoxins is primarily from drinking water; however recreational activities such as swimming or boating can also lead to exposure (Turner et al. 1990, Carmichael 1994, Falconer 1999). Although direct consumption of lake water in the United States is rare, a number of cases of cyanotoxin contamination of raw and treated drinking water have been reported within a number of municipalities (Boyer et al. 2004). Moreover, in developing nations, due to a lack of water treatment plants, raw lake water may be consumed. Symptoms of short-term recreational exposure to microcystin can include vomiting, diarrhea, central abdominal pain, sore throat, and blistering of the lips (Turner et al. 1990). On the other hand, long term exposure to cyanotoxins can be associated with severe health effects. Worldwide, reports of human deaths, liver and colorectal cancers, neurological disorders, such as Alzheimer's disease, and other illnesses have been associated with water contaminated with cyanotoxins (Falconer et al. 1988, Carmichael & Falconer 1993, Bell & Codd 1994, Carmichael 1994, Chorus 1999a, Zegura et al. 2003, Cox et al. 2005). However, in the United States, where residents generally do not rely on surface water for their primary source of drinking water, most reports of cyanotoxin poisonings involve water fowl, cattle and domestic pets which are exposed to cyanotoxins by ingestion of contaminated water.

In light of the environmental problems associated with eutrophication and algal blooms, a multitude of measures have been proposed to remediate such problems. In determining remediation measures for smaller water bodies, it is critical to understand the sources from which excess nutrients emanate. As each river, lake, or estuary is often influenced by multiple nutrient sources, the type and magnitude of each nutrient source must be determined in order to construct an accurate nutrient budget. There are multiple sources of external

nutrients into a body of water including: domestic wastewater, road runoff, fertilizer (residential and agricultural), and groundwater. Management strategies can, and often do, focus on reducing the nutrient input from wastewater, road runoff, and fertilizers, which are known as point sources (Chorus 1999a). A nutrient budget of these sources can then be used to identify which remediation measure(s) will be most successful for alleviating the environmental problems within a given ecosystem.

The goal of this study was to evaluate the point and non-point nutrient sources to Lake Agawam and evaluate phytoplankton community responses to nutrient dilutions in order to better determine effective remediation measures.

## Materials and Methods

Agawam Lake is a small (328 m<sup>2</sup>) shallow (mean depth 1.6 m) fresh water eutrophic lake located at 40° 52' 05" N, 72° 25' 96" W. The lake is oriented north to south and is surrounded by the Village of Southampton. To the north of the lake are well-developed commercial lots. To the east and west are residential lots. To the south is a thin strip of land which separates the lake from the Atlantic Ocean. The lake experiences minimal saltwater intrusion however. Due to urbanization and the raw drainage water entering the lake, along with the lake's physical features it has become a hypereutrophic system by EPA standards (mean chlorophyll  $a = 107 \mu\text{g L}^{-1}$ ). Consequently, this lake has been experiencing chronic dense cyanobacteria blooms which last from May until October and are dominated by *Microcystis* cells.

As part of an ongoing study starting in 2004 (Davis & Gobler 2007), a station located in the longitudinal middle Lake Agawam within the northern half of the lake was sampled weekly to biweekly from May through November in 2007. For each sampling, general physical parameters (temperature, dissolved oxygen, and pH) were measured using a YSI 556 MPS probe. Replicate twenty liter carboys of water were filled from 50 cm below the surface of the lake and returned to the laboratory for analysis within 1 hour. In the lab, nutrient samples were filtered through combusted (2 hr. @ 450°C) glass fiber filters (GFF) and stored frozen. These

were later analyzed for nitrate, ammonia, phosphate, dissolved organic nitrogen (DON) and dissolved organic phosphate (DOP) (Valderrama 1981, Jones 1984, Parsons et al. 1984). *In vivo* samples for chlorophyll *a* (total phytoplankton) and phycocyanin (total cyanobacteria) were size fractionated for whole (glass fiber filters (GFF)) and less than 20  $\mu\text{m}$  (20  $\mu\text{m}$  polycarbonate filters). Filters were placed in 25 ml falcon tubes and stored frozen for 24 hours. A 90% acetone solution was then added to each tube and stored frozen for an additional 24 hours. The supernatant was then analyzed using a Turner Designs fluorometer (model TD-700) by standard fluorometric methods (Parsons et al. 1984).

Nutrient dilution experiments were conducted to evaluate the effects of reduced nutrient loading on the abundance of cyanobacteria populations. Triplicate bottles were set up for all treatments using acid-washed 250 ml polycarbonate bottles. Triplicate samples of 50% whole lake water were incubated with filtered lake water (0.2  $\mu\text{m}$ ) or with a lake ion solution which did not contain nitrogen or phosphorus. Experimental bottles were incubated  $\sim$ 0.3 m in Old Fort Pond (OFP) at the Southampton College Marine Station. Open tidal exchange with Shinnecock Bay keeps OFP well flushed. Temperatures during incubations were similar to that of the station at Agawam Lake. After 48 hours, bottles were filtered for chlorophyll *a* and phycocyanin onto glass-fiber filters.

In addition to these experiments, an attempt was made to assess the relative sources of nutrients to this system. Primary nutrient sources for the lake were determined to originate from a large storm drain at the north of the lake, benthic fluxes, groundwater input, atmospheric input and surface runoff. These input sources were quantified to construct a nutrient budget for the lake. Data used to construct the budget were June through October 2007.

The storm drain contained two outflow openings which measured 170 x 60 cm and 175 x 60 cm. During heavy rain events (precipitation > 2cm) both openings would be seen to release water. During light rain events (precipitation < 2cm) only the larger of the two openings (right facing) was seen to release water. There is a 20-minute delay from the initial start of the rain event to release of water from the storm drain, potentially due to

a catch basin further up the pipe. During rain events, water samples were collected from outflow in acid-washed 250 ml bottles. Flow rates were measured using a General Oceanics Flowmeter (Model 2035). Flow depth was measured with a meter stick from the base of the opening to the middle of the flow to get an average depth. Rain start and end times were also recorded. Water samples were stored frozen and later analyzed for nitrate, ammonia, phosphate, DON and DOP (Valderrama 1981, Jones 1984, Parsons et al. 1984). Total storm drain contribution was also determined by considering the size of the Village watershed (approximated to be 4,839,243 m<sup>2</sup>) and estimating that 20% of this watershed was likely impervious and funneled into the storm drain system due to streets, parking lots, buildings and driveways (Civco et al. 2006). The volumetric contribution of the storm drain determined via this method was similar to volumes determined via several actual measurements extrapolated for the five month study period.

To determine benthic flux, sediment core samples were obtained from three locations in the lake: one at the north sampling station, one at the longitudinal center of the lake and one near the southern portion of the lake. In addition to core samples, general physical parameters of the lake (temperature, pH, salinity and dissolved oxygen) were measured via a YSI 556 MPS probe. Bottom depth and water clarity were determined by secchi disk. Cores were extracted using a box corer dropped from the side of the boat which was then brought to 0.3 m below the water surface. An acid-washed clear polycarbonate tube (length = 26.6 cm, diameter = 9.3 cm) was then inserted through the top of the corer to collect a sediment sample. While the tube was still in the sediment, a plastic cap was placed on the bottom and then the top to capture the sediment sample and lake water immediately above the sediment. Cores were immediately placed in a cooler and transported back to the lab within one hour. A replicate and blank of the North End were also retrieved. Core samples were then incubated in similar light and temperature conditions to those measured at the lake bottom of each site. The samples were also aerated to achieve similar dissolved oxygen levels found in bottom waters of Lake Agawam using an aquarium air pump. Physical parameters were monitored using an Onset® temp/light monitor. Water samples were extracted using an acid-washed 60 ml syringe with 15 cm tubing attached to the end. Water was drawn up slowly from just above the sediment water interface and care was taken to not draw

up sediment. Samples were placed in acid-washed 60 ml bottles and frozen. The incubation was allowed to run for 12 hours with a total of 5 samples obtained per core as a time course during the incubation. Samples were filtered on combusted GFF and analyzed for nitrate, phosphate, ammonia, DON and DOP (Valderrama 1981, Jones 1984, Parsons et al. 1984). As filtered lake water was not added to replace the volume extracted, a mass balance correction was applied using the equation  $(C_0 - C_1) \times V_0 = \Delta m$  where  $C_0$  is the starting concentration,  $C_1$  is the ending concentration,  $V_0$  is the starting volume and  $\Delta m$  is the mass change. This correction was applied to each time point in the series and the results were plotted against time. The resulting slope was used to determine the flux of nutrients out or into the sediment.

The volume of groundwater recharging the Lake Agawam watershed was estimated using the methods of Steenhuis et al. (Steenhuis et al. 1985) who determined that seasonal changes in evapotranspiration is the process which has the greatest influence on groundwater recharge on eastern Long Island. This method has been used successfully in the past to calculate groundwater discharge and recharge rates into Long Island embayments (Gobler & Sañudo-Wilhelmy 2001, Montlucon & Sañudo-Wilhelmy 2001, Gobler & Boneillo 2003) where recharge rates are generally comparable to groundwater discharge rates (Schubert 1998). The watershed area for Lake Agawam was determined from USGS water table maps of Long Island's south fork watershed (Schubert 1998). The area of the lake itself was determined from an EPA Enviromapper® topographic map. A gridded overlay was applied to both maps and used in calculating the area. Grid squares containing > 50% of the relevant shape (lake or watershed) were counted in the area calculation. Squares containing <50% of the relevant shape were not counted. From the north and south end of the lake groundwater was sampled 0.3 m from the shore by shallow (1 m) PVC piezometers with 2.5 cm long, horizontal screened slits along the lower 25 cm. Piezometers were sampled using a low-flow (<100 ml/min.) peristaltic pump equipped with acid-washed Teflon tubing (Gobler & Boneillo 2003). Samples were frozen and later filtered using combusted GFF and analyzed for nitrate, phosphate, ammonia, DON and DOP (Valderrama 1981, Jones 1984, Parsons et al. 1984).

Concentrations of dissolved inorganic nitrogen (DIN) and DON in precipitation were obtained from Cornell et al. (Cornell et al. 1995). Atmospheric deposition fluxes (wet and dry) were calculated according to an average flux obtained from three sources; Paerl and Hu et al. (Paerl 1993, Hu et al. 1998) and data retrieved from the National Atmospheric Deposition Program ([nadp.sws.uiuc.edu/](http://nadp.sws.uiuc.edu/)). Precipitation data was obtained from NOAA Satellite and Information Service. Run-off volumes (5% of precipitation to the watershed) and estimates of DIN concentrations in run-off were obtained from a study of a similarly sized, enclosed embayment on eastern Long Island (Gobler & Boneillo 2003).

## Results

From May to October large phytoplankton blooms occurred with mean chlorophyll *a* (chl *a*) levels in May of  $38.21 \mu\text{g L}^{-1}$ , peaking in late June at  $150 \mu\text{g L}^{-1}$  and also again in late September at  $90 \mu\text{g L}^{-1}$  (Table 1). These levels were slightly lower than previous seasons where bloom densities of  $200 \mu\text{g L}^{-1}$  were observed (Gobler et al. 2007). Similar trends were observed in the phycocyanin concentration levels which also peaked in late June at 75 RFU (Table 1). DIN concentrations were inversely related to chl *a* and phycocyanin levels. As concentrations of chl *a* and phycocyanin rose during June, DIN concentration decreased markedly and remained low through the summer and fall (Table 1). DON and DIP concentrations showed only slight fluctuations throughout the season. DOP concentrations began low ranging from  $0.21 \mu\text{M}$  to  $0.48 \mu\text{M}$  at the beginning of the season, but then increased to  $8 \mu\text{M}$  on average in August to October (Table 1).

Experimental reductions of ambient nitrogen and phosphorus levels were capable of significantly reducing algal biomass during summer months. Nutrient dilution did not alter algal biomass in May, early June, September and October (Figure 2, 3). However, during peak bloom months (mid-June through August) a reduction in nutrients by 50% reduced phycocyanin levels on average by half (Figure 2) and chl *a* levels on average by 40% (Figure 3). In addition to the reductions of phycocyanin being larger than those in chlorophyll, during the late June experiment, phycocyanin levels decreased by 40% while chlorophyll levels were unchanged (Figure 2,3)

Nutrient budget calculation indicated that groundwater is the greatest contributor of inorganic nitrogen to the lake. DIN input from groundwater is calculated to be  $718 \text{ mol day}^{-1}$  (Table 2) which makes up 46% of the DIN supply (Figure 4). This is nearly twice the amount of DIN originating from benthic flux and effluent from the storm drain ( $354 \text{ mol day}^{-1}$  (23%) and  $439 \text{ mol day}^{-1}$  (28 %) respectively) (Table 2, Figure 4). Atmospheric deposition and surface runoff were negligible when compared to these inputs. For organic fluxes of nitrogen, the benthos was the largest source, contributing  $241 \text{ mol day}^{-1}$  DON (Table 2) or 75% of the DON budget (Figure 4). Other source contributions were an order of magnitude less than this. Phosphorus inputs, both organic and inorganic, mainly originated from the benthos (94% DOP, 83% DIP and 89% TDP) with inputs from the storm drain making up 12% of the DIP budget (Figure 4). In comparison, all other sources were minimal.

## Discussion

As part of ongoing monitoring of the lake which includes this study, nutrient amendment experiments have shown that nitrogen becomes the limiting nutrient for the lake during months of high bloom biomass (Davis & Gobler 2007, Gobler et al. 2007). Phosphorus, particularly DOP, is relatively abundant in the lake due primarily to recycling from the benthos. Ninety-four percent of the total dissolved phosphorus input to the lake comes from the sediment flux (Figure 4). Typically, phosphorus levels are targeted to minimize or reduce cyanobacterial densities, but this can be difficult in small, shallow, fresh water lakes as recycling rates are generally high (Chorus 1999b). In light of this, it may be more prudent to target the sources of nitrogen.

For Lake Agawam, with the majority of DIN originating from groundwater flow, upstream remediation measures are needed to lessen the amount reaching the lake. Previous monitoring has shown that DIN concentrations in the groundwater have been high throughout the South Fork of Long Island. USGS wells located north of the lake show average DIN concentrations of  $300 \mu\text{M}$  (National Water Information System; <http://waterdata.usgs.gov/nwis>). County wells within the watershed also have shown high concentrations of DIN (mean =  $480 \mu\text{M}$ ,  $n=3$ ; Suffolk County Department of Health Services) and a recent study documented

levels around the western shoreline of Mecox Bay to be 120  $\mu\text{M}$  (Gobler et al. 2005). This suggests that there is a widespread problem with nitrogen contaminated groundwater in the region which affects not only the watershed supplying Lake Agawam, but the South Fork as a whole. Historically, two possible sources for nitrate contamination include past farming practices and Suffolk County's reliance on septic tanks for wastewater disposal (Laroche et al. 1997, Vitousek et al. 1997, Gobler & Boneillo 2003, Reay 2004, Gobler et al. 2005).

Remediation of groundwater nutrient loads may prove too costly or politically difficult in Suffolk County due to such widespread use of septic tanks within the county. With this difficulty, a focus should perhaps be directed at benthic and storm drain sources of nutrients. Both sources combined make up over 50% of the dissolved nitrogen budget and 89% of the dissolved phosphorus budget (Figure 4). Possible remediation tactics could involve sediment dredging and diversion of storm water runoff. Dredging of the lake has the potential to reduce both nitrogen and phosphorus loads to the lake however care should be taken as in some cases dredging has further exacerbated the problem. China, with its booming economy, is facing many environmental challenges including eutrophication of its lakes, rivers and estuaries. In attempts to combat the large cyanobacteria blooms that are occurring, various remediation measures are used. In South Lake, China, dredging decreased phosphorus loads to the lake but had no effect on nitrogen (Wang & Feng 2007). It was suspected that during the dredging process, ammonia was released into the water column which hindered the short-term effectiveness of dredging (Wang & Feng 2007). However, it is likely that over time, this process improved the longer term nitrogen conditions for this system.

The dilution experiments in this study suggest that reducing nutrient loading to Lake Agawam could reduce bloom intensity. Specifically, a 50% reduction in nutrient levels during experiments resulted in 50% lower densities of cyanobacteria and 40% lower levels of chlorophyll *a* during the peak bloom period of the summer (Figure 2,3). In addition, during late June, the experiment reduced cyanobacteria levels by 40% but had no impact on chlorophyll *a* (Figure 2,3). These two findings suggest that diluting nutrients will not only reduce algal biomass, but may also yield a shift in the phytoplankton community away from cyanobacteria and toward non-phycoerythrin containing-species. Since cyanobacteria blooms are the cause of many environmental

problems in Lake Agawam, including fish kills, a focus on nutrient reduction should be a part of any future management plan for Lake Agawam.

## Tables and Figures

**Table 1.** *In Vivo* record of Temperature, Phycocyanin, Chlorophyll *a*, and nutrient concentrations taken during this study.

	Temperature [°C]	Phycocyanin [RFU]	Chlorophyll <i>a</i> [ $\mu\text{g L}^{-1}$ ]	DIN [ $\mu\text{M}$ ]	DON [ $\mu\text{M}$ ]	DIP [ $\mu\text{M}$ ]	DOP [ $\mu\text{M}$ ]
30-May-07	23.80	7.43	38.21	111.66	-	0.16	0.21
12-Jun-07	22.50	12.87	23.92	45.07	12.31	0.00	0.48
26-Jun-07	26.30	75.00	150.43	1.21	21.18	0.28	0.14
17-Jul-07	28.01	11.80	44.06	1.29	14.66	0.08	0.45
31-Jul-07	29.29	19.30	52.13	1.74	23.07	0.13	0.20
13-Aug-07	25.51	19.70	36.76	2.38	17.69	0.09	7.89
29-Aug-07	-	12.30	28.85	1.11	15.98	0.08	8.08
27-Sep-07	22.90	40.10	90.25	0.91	24.33	0.23	8.03
11-Oct-07	18.80	64.90	16.10	14.22	-	0.04	8.01

**Table 2.** Nutrient contributions to Lake Agawam in mol day<sup>-1</sup>.

	DIN	DIP	DON	DOP	TDN	TDP
<b>Surface Runoff</b>	7	0	10	1	17	1
<b>Groundwater</b>	718	5	14	2	733	7
<b>Benthic Flux</b>	354	129	241	173	594	302
<b>Storm Drain</b>	439	19	44	7	483	26
<b>ATM</b>	35	2	11	1	47	3

**Figure 1.** Initial Chlorophyll *a* and Phycocyanin concentrations.

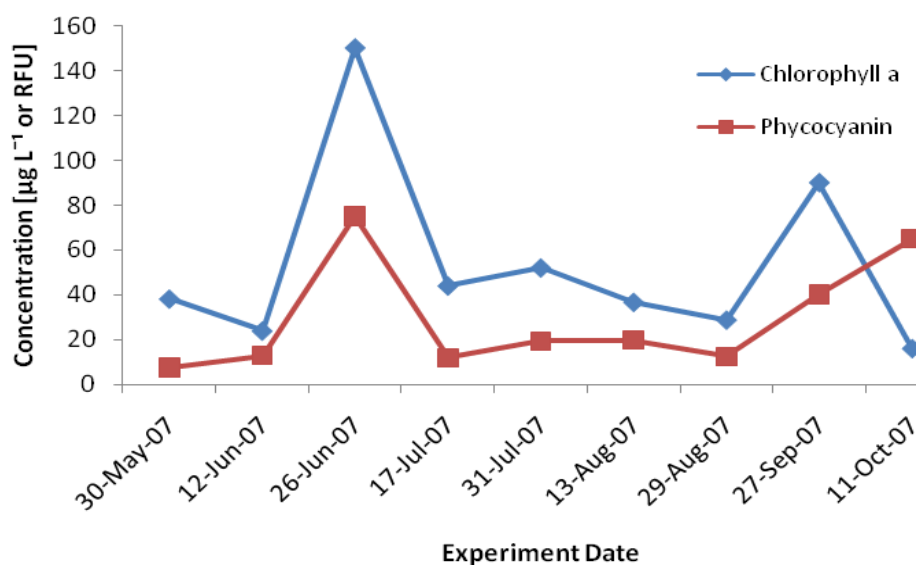


Figure 2. Results of nutrient dilution experiments showing changes with addition of nutrients in phycocyanin concentration. Percent change indicated above peak bloom dates.

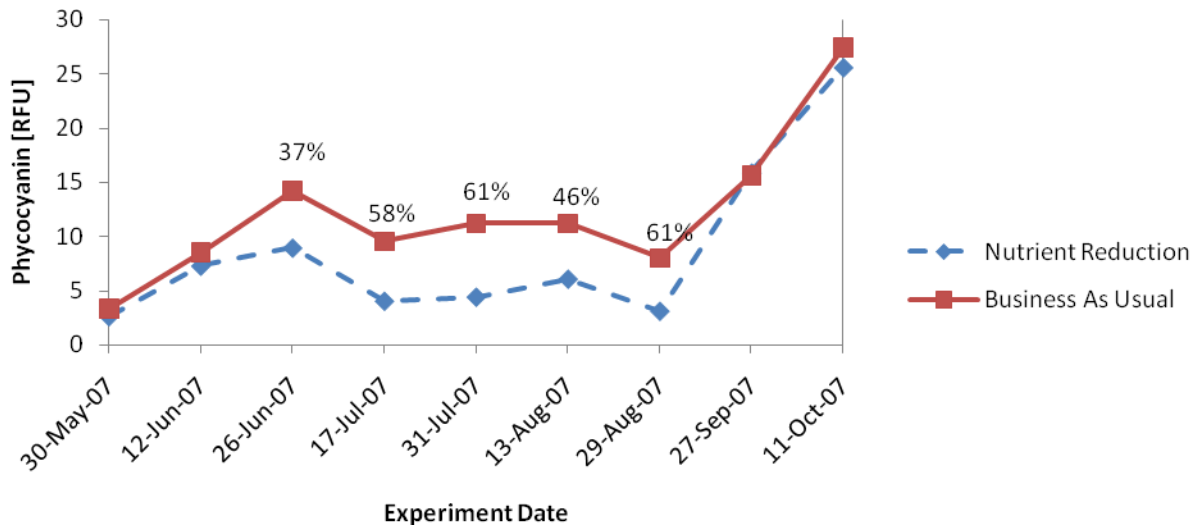


Figure 3. Results of nutrient dilution experiments showing changes with addition of nutrients in chlorophyll *a* concentration. Percent change indicated above peak bloom dates.

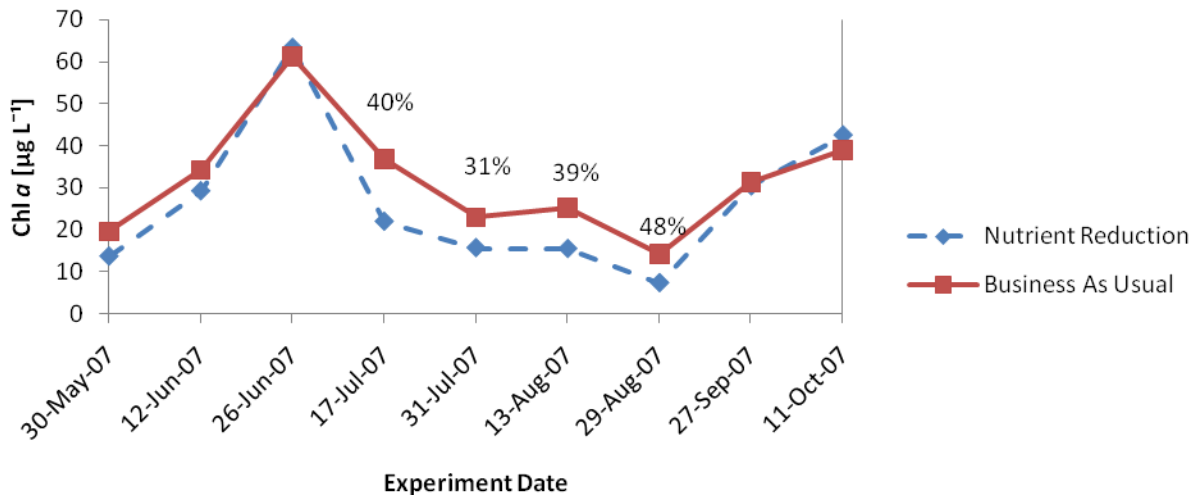
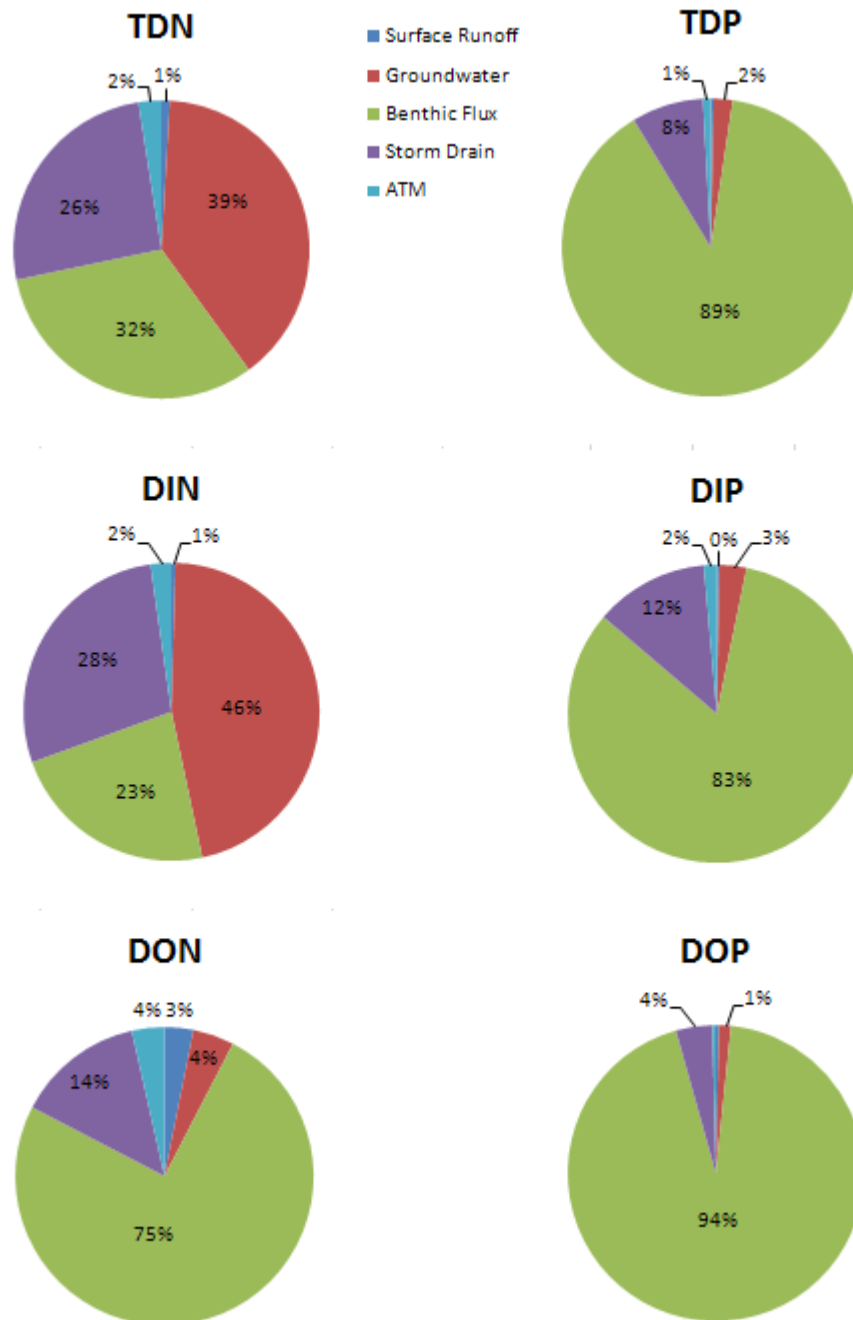


Figure 4. Relative contribution of nutrient species entering Lake Agawam.



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