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Cyanobacterial Harmful Algal Blooms Vary Within and Across Years at Lake Fayetteville, Arkansas

Alyssa Ferri
University of Arkansas, Fayetteville

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Alyssa Ferri
University of Arkansas
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_____
Brian Haggard, Ph.D.
Thesis Director

_____
Mary Savin, Ph.D.
Committee Member

_____
Lisa Wood, Ph.D.
Committee Member

Abstract

The occurrence of cyanobacterial harmful algal blooms (HABs) and toxins are a nationwide concern. Although Lake Fayetteville likely experienced HABs for many years, microcystin, an algal toxin, was not monitored until 2019. The objectives of this study were to: 1) observe temporal variation in water quality and total microcystin concentrations, 2) quantify thresholds with microcystin and nutrients, algal parameters, and environmental factors, and 3) evaluate complex relationships between total microcystin and nutrient supplies, algal parameters, and environmental factors using a classification and regression tree model (CART). Three sites (dam, inlet, and mid) at Lake Fayetteville were sampled weekly to monthly from 2019 to the end of 2021. Water samples were analyzed for temperature, pH, dissolved and total nutrients, chlorophyll-a, chlorophyll raw fluorescence units (RFU), phycocyanin (RFU), and total microcystin. Peak mean total microcystin concentrations (1.9 to 6.2 μg/L) and the longevity of toxin production varied between years. Many parameters had significant thresholds with total microcystin across the time periods in this study including temperature, algal biomass, inorganic N concentrations, and molar TN:TP ratio. Significant nCPA thresholds with total microcystin concentration varied across years, and the parameters with the greatest R^2 values were increased water temperatures, elevated algal biomass, low inorganic N concentrations, and an intermediate molar TN:TP ratio. The CART models for the all data and each study year varied in the number of splits, the parameters included, and amount of variation in total microcystin explained, and the 2020 model explained the most variation (76%) in the total microcystin concentrations. Overall, the factors triggering a HAB and toxin production are not well known, but monitoring a lake over multiple years and developing waterbody specific guidelines can help guide future management decisions and help protect human health.

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Introduction

Worldwide, anthropogenic eutrophication is altering aquatic ecosystems through the proliferation of harmful algal blooms (HABs) (Carmichael, 2001). Furthermore, global climate change, the potential warming of surface waters, and changes in precipitation could result in an increase in frequency and magnitude of HABs and potential toxin production (Paerl et al., 2011). Blooms can be composed of numerous taxa, but blue-green algae, or cyanobacteria, are one of the most wide-spread and problematic taxa of freshwater HABs.

Cyanobacterial blooms degrade water quality through dissolved oxygen depletion, aesthetic nuisances (odor, scums, and unsightliness), and unpalatable and potentially unsafe water supplies (Paerl, 1988). Additionally, cyanobacteria can produce toxins (e.g., microcystin) which are harmful to biota including invertebrates, finfish, shellfish, and vertebrate consumers of water, including pets and humans (Paerl et al, 2001). Cyanobacterial toxin production may vary widely spatially, between and within an aquatic system, and temporally, during a single sampling season and between years (Jacoby et al., 2000).

Many cyanobacteria (e.g., *Microcystis, Anabaena,* and *Dolichospermum*) can produce hepatotoxic, neurotoxic, and/or cytotoxic compounds that are a human health risk in freshwaters (Carmichael, 1994; Sarkar et al., 2020). These toxins primarily exist within cyanobacterial cells, but these toxins can be free in the water when excreted or during senescence of the algal bloom (Jacoby et al., 2000). Microcystin is one of the most frequently measured cyanotoxins which is produced by many genera of cyanobacteria (Yuan et al., 1999). Microcystin production can be triggered by various environmental conditions such as nutrient dynamics (Neilan et al., 2013).

Nutrients such as nitrogen (N) and phosphorus (P) are an important driver of cyanobacterial HABs and microcystin production. In lakes on continental scales, microcystin concentrations are positively correlated with total nitrogen (TN) and total phosphorus (TP) (Yuan et al., 2014). The dominance of different cyanobacteria depends on nutrient availability and ratios in the water (Paerl et al., 2001). The greatest toxin production typically occurred when N and P were balanced in intermediate molar TN:TP ratios (between 26 and 44) (Scott et al., 2013; Otten et al., 2012; Paerl et al., 2011). Some studies observed that specific N forms (i.e., nitrate and ammonium) in the water can increase the growth of cyanobacteria and subsequent toxin production (O'Neil et al., 2012; Ha et al., 2009). Other studies suggest that the N form did not matter, but rather the magnitude of the N supply did (e.g., Wagner et al., 2021). P supply also plays a role in toxin production since microcystin is positively correlated with soluble P at low concentrations (<10 μg/L). While nutrients play a significant role in cyanobacterial growth and toxin production, other environmental and hydrologic conditions can influence cyanobacterial HABs and toxin production.

Cyanobacterial blooms typically occur from spring to late fall throughout the United States due to elevated water temperatures and increased vertical stratification (Paerl and Huisman, 2008). Optimal cyanobacterial growth occurs at increased water temperatures (30 to 35°C), while cellular toxin production may be greatest at lesser temperatures (20 to 25°C) (O'Neil et al., 2012; Giannuzzi, 2018). Cyanobacterial blooms grow and produce toxins at a typical water pH range (six to nine), but microcystin concentration is positively correlated with pH (Caraco and Miller, 1998; Francy et al., 2016). Cyanobacteria can regulate their buoyancy in the water column for growth to actively seek optimal conditions (i.e., low turbulence, light, and nutrients) (Carey et al., 2012). Toxin production may occur to limit growth of competing algal species or cyanobacteria

predators (Paerl et al., 2001). Overall, HABs and toxin production are influenced by a variety of interconnected factors, and an understanding of these across freshwaters will be key to improve water quality and protect human health from the adverse effects of HABs.

A multi-year monitoring study began at Lake Fayetteville, Arkansas in April 2019. On May 7, 2019, a HAB at the dam site was detected with a total microcystin concentration that exceeded 8 µg/L (EPA recreational guideline) (USEPA, 2019). Monitoring of the lake continued through 2021 with each growing season also experiencing cyanobacterial HABs. The primary objectives of this study were to: 1) observe temporal variation in water quality and total microcystin concentrations from 2019 through 2021, 2) quantify thresholds with microcystin and nutrients, algal parameters, and environmental factors, and 3) evaluate complex relationships between total microcystin and nutrient supplies, algal parameters, and environmental factors using a classification and regression tree.

Methods

Study Site Description

Lake Fayetteville (36°08'11.5" N, 94°07'46.7" W) is a 0.604 km^2 hypereutrophic and shallow (mean depth 3 m and maximum depth 10 m) reservoir in northwest Arkansas (Grantz et al., 2014). The reservoir was constructed in 1948 on Clear Creek, and the average residence time for the reservoir is 164 days (Grantz et al., 2014). Lake Fayetteville now serves as a recreational source for the surrounding population with an adjacent trail system, a marina, and a disc golf course, among other amenities.

The Lake Fayetteville watershed has a total area of 24 km^2 and in 2019 consists of: 43.5% agriculture (pasture, grasslands, and cultivated crop uses), 44.4% urban (developed open space, low, medium, and high intensity uses) and 8.4% forested (deciduous, mixed, and evergreen forests). Land use was estimated using the 2019 National Land Cover Database in Model My Watershed (https://modelmywatershed.org/).

Many studies have been conducted on Lake Fayetteville since the reservoir's creation in 1949. A study observed that cyanobacterial blooms occurred in spring and into the fall, showing that this lake has always had a dominance of cyanobacteria in the phytoplankton community (Meyer, 1971). Past studies showed that nutrient supplies vary seasonally since 1971, where nitrate-N which ranged 1.30 to 1.60 mg/L in the winter to 0.11 mg/L in summer. Nitrogen from the inflows and atmospheric deposition is not the only source because nitrogen fixation which is 27% of the total N input into Lake Fayetteville (Scott and Grantz, 2013; Grantz et al., 2014). Furthermore, the reservoir retains 90% of P inputs, N retention is a net zero because of

denitrification (Grantz et al., 2014). Overall, these observations suggest that cyanobacterial growth in the lake should be colimited if during the growing season (see Haddock et al., 2022).

Since March 2019, water samples were collected weekly to monthly at three public access sites along the northern windward side of the reservoir (Figure 1). Water samples were typically collected from 11:00 AM to 2:00 PM (central time) just below the water surface (depth of 0.15 m). At each site, one 1 L and one 125 mL bottle were filled by triple rinsing the collection bottle with the lake water before the final water collection. Water temperature, dissolved oxygen (DO; YSI Model 85, Yellow Springs, Ohio), pH (pH Testr30, Vernon Hills, Illinois), and conductivity (Hach Pocket Pro, Loveland, Colorado) were measured on site following sample collection.

Analytical Methods

Upon return to lab, the 125 mL raw water samples were acidified to pH < 2 with H_2SO_4. The water was used in persulfate autoclave digestions (APHA 4500-P J), and then the digests were analyzed for total nitrogen (TN) and total phosphorus (TP) on a Skalar Sans++ Wet Chemistry Autoanalyzer (Skalar inc., Buford, Georgia) (Table 1). A subsample of 20 mL from the 1 L bottle was filtered through a 0.45 μm syringe filter and acidified to pH < 2 using H_2SO_4, and the acidified filtrate was analyzed for dissolved nutrients (nitrate plus nitrite-N (hereafter, NO_3-N); ammonium-N (NH_4-N); soluble reactive phosphorus (SRP)) on the Skalar San++ Wet Chemistry Analyzer. Dissolved inorganic nitrogen (DIN) was calculated by adding NO_3-N and NH_4-N. TN:TP molar ratios were calculated by multiplying the ratio by (31/14), and particulate N:P (PNPP) ratios were calculate by subtracting dissolved nutrients from total nutrients; the supply N:P ratios were on a molar basis using only dissolved nutrients.

Chlorophyll (CHL) and phycocyanin (PC) raw fluorescence units (RFU) were measured on a CyanoFluor Handheld HAB Indicator (San Jose, California) starting June 25, 2019. The RFUs were corrected using filtered (0.45μm) water, which removed interference from dissolved organics. Then, PC values were divided by CHL to get a ratio (PC:CHL).

For chlorophyll-*a* and pheophytin, 25 to 50 mL, depending on algal biomass, of sample was filtered through a Whatman 0.7 μm glass microfiber filter. The filter was put into a glass vial, preserved with 90% acetone, and stored in a freezer. After at least 24 hours, samples were analyzed on a Turner Design Fluorometer (APHA 10200 H3; San Jose, California).

Raw water was used in total microcystin analysis. From 2019 through June 2020, 2 mL of lake water was used in 6 mL amber vials; after June 2020, 20 mL of lake water was used (Austin and Haggard, 2022). These samples went through three freeze thaw cycles to lyse cyanobacteria cells, releasing intracellular microcystin. Following these freeze thaw cycles, total microcystin was quantified using enzyme linked immunosorbent assays (EPA Method 546; Eurofins Abraxis; Warminster, Pennsylvania).

Seasons and precipitation were also used in this study. Seasons were classified as Spring (March 1 to May 31), Summer (June 1 to August 31), Fall (September 1 to November 30), and Winter (December 1 to February 28). Growing season was split into two categories, growing season (April 1 to October 31) and off-season (November 1 to March 31). Daily precipitation data was downloaded from the National Water Information System (NWIS; https://waterdata.usgs.gov/nwis) for Mud Creek (USGS 071948095); rainfall data was compiled into 24-hour, 72-hour and 1-week values prior to each sampling date.

Statistical Data Analysis

Temporal changes in all physicochemical properties were evaluated qualitatively using graphs of data over time. The mean of the three sites was used since the data were not highly variable between sites. A microcystin concentration of at least 2 μg/L was used for longevity based off the World Health Organization guidelines for recreational contact (WHO, 2003). Box and whisker plots were created with all the data by month, season, and algal growing season. A Wilcoxon Rank Sum test was used to determine significant differences between months, seasons, and growing seasons with an α of 0.05.

Nonparametric change point analysis (nCPA; RStudio) was used to assess nonlinear responses or thresholds in water quality parameters with total microcystin. The dataset was then split into two subgroups which results in the greatest reduction in deviance from the mean of the response variable (Qian et al., 2003). Thresholds were evaluated using all data and for each calendar year with an α of 0.05 and a bucket size of seven. (King and Richardson, 2003; Qian, 2003).

A classification and regression tree (CART) model was applied to the physicochemical dataset, which included month, year, and site in addition to previously mentioned parameters, to identify thresholds and hierarchical structure with total microcystin concentrations. Thresholds and relationships were evaluated for all years combined and for each calendar year (2019, 2020, and 2021). A CART model builds a tree sequentially by repeatedly splitting the dataset into two distinct partitions (Yuan and Pollard, 2014). A split in the data minimizes the deviance in the fitted relationship between the two parameters for each partition. This process of splitting the data continues repeatedly using predefined constraints. The model produced a complexity parameter which in this study was an approximation for the R^2. The constraints in this study were: 1) for a

split to occur, the complexity parameter had to increase by 0.05 and 2) each bucket must contain at least seven observations. Pruning the tree is necessary to reduce the possibility of overfitting the model to the data (Wu and Kumar, 2010). The CART computations were done in RStudio Desktop (version 2021.09.2+382), using the library *RPART* (https://cran.r-project.org/web /packages/rpart/index.html).

Results

Temporal Variation in Water Quality and Total Microcystin

The magnitude of total microcystin concentrations varies between and within a calendar year, while the temporal trends were not so different (Figure 2). The high concentrations of total microcystin (11 and 15 µg/L) were observed in 2019, while total microcystin had prolonged elevated concentrations in 2020 but lesser concentrations in 2021. When looking across months, total microcystin was low (<0.25 µg/L) from December to March (Figure 3; Wilcoxon Rank Sum, $p < 0.05$). Then in April, total microcystin concentrations increased in magnitude and variability. Total microcystin continued to increase in May, reaching over 1 µg/L at times. The greatest variability in total microcystin concentrations occurred in June and July, while the greatest median was in August (0.7 µg/L). Median total microcystin slightly dipped in September (<0.5 µg/L) only to increase again in October (0.7 µg/L). Overall, total microcystin was greatest in summer, next greatest in fall, and least in winter and spring (Figure 4; Wilcoxon Rank Sum, $p < 0.05$). The off-season had total microcystin concentrations less than 0.5 µg/L, whereas the greater concentrations occurred during the growing seasons (Wilcoxon Rank Sum, $p < 0.05$).

This variability in total microcystin was likely driven by physico-chemical properties that varied temporally (Figure 2). Water temperature can be a main driver of algal growth since the parameter varies throughout a year. In Lake Fayetteville, the water temperature in January and February is low (<10°C) and then increased in magnitude each month from March (15°C) to June (28°C). July and August had elevated median water temperatures (30°C). After August, temperatures decreased monthly, but October had the greatest variability with temperatures reaching summertime conditions at times. Overall, water temperature trends follow total microcystin in that summer had the greatest water temperatures, the next greatest in fall, then

spring, and the least were in winter. The off-season had median water temperatures of 14℃ which increased to 27℃ during the growing season.

Unlike water temperature, TN followed a relatively consistent pattern throughout the three years except for a peak in 2021 reaching a maximum of 3.0 mg/L (Figure 2). Moreover, TN was elevated during winter and spring with the greatest median concentrations in January and May (1.8 mg/L) (Figure 3; Wilcoxon Rank Sum, $p < 0.05$). After January, TN decreased in magnitude each month until April (1.3 mg/L), where February showed the least variability (1.68 to 1.72 mg/L). In May, TN increased in magnitude and variability (1.7 mg/L) and then decreased in June only to remain low (≤1.0 mg/L) until August. Finally, TN elevated in magnitude from September (1.1 mg/L) to December (1.5 mg/L). Similar to February, TN did not vary greatly in November. Overall, median TN was greatest in the winter (1.7 mg/L) and spring (1.6 mg/L), and second greatest in the fall (1.2 mg/L), and least in the summer (1.1 mg/L) (Figure 4; Wilcoxon Rank Sum, $p < 0.05$). The off-season median was increased in magnitude (1.6 mg/L) and less variable than in the growing season (1.1 mg/L) (Wilcoxon Rank Sum, $p < 0.05$).

Nitrate followed a similar pattern to TN, but in 2020 no nitrate was present during total microcystin production which differed from 2019 and 2021 (Figure 2). In winter, NO_3-N was the greatest fraction of TN, and median NO_3-N was greatest in January (1.4 mg/L) and March (1.1 mg/L) (Figure 3; Wilcoxon Rank Sum, $p <0.05$). Then NO_3-N decreased every month until June (0.1 mg/L). From July to September, there was practically no measurable NO_3-N in the surface water. In October, NO_3-N magnitude (0.2 mg/L) and variability increased; NO_3-N continued to elevate until December (0.8 mg/L). Median NO_3-N concentrations varied between each season where NO_3-N was greatest in the winter, second greatest in the spring, third greatest in the fall and least in the summer (Figure 4; Wilcoxon Rank Sum, $p < 0.05$). The median NO_3-N concentrations

in the off and main season were significantly different (Wilcoxon Rank Sum, $p < 0.05$) at 1.1 and < 0.1 mg/L, respectively.

Similar to NO_3-N, ammonia was not present during toxin production in 2020, but was present in 2019 and 2021 (Figure 2). At the end of 2021, NH_4-N concentrations were at the greatest concentrations out of all three years. NH_4-N supply in the water was almost an order of magnitude less than NO_3-N through all three years (Figure 2). Median NH_4-N decreased from January (0.13 mg/L) to February (0.01 mg/L) (Figure 3; Wilcoxon Rank Sum, $p <0.05$). In March and April, median NH_4-N stayed around 0.07 mg/L before decreasing monthly and remaining low (<0.05 mg/L) through September. Then, NH_4-N concentrations increased in magnitude from October (0.08 mg/L) to November (0.13 mg/L) and December (0.16 mg/L), and variability was also greatest during these three months. Seasonally, median NH_4-N was greatest in winter (0.14 mg/L) and fall (0.07 mg/L), and least in summer (0.02 mg/L) (Figure 4; Wilcoxon Rank Sum, $p < 0.05$). The off-season had greater median NH_4-N (0.12 mg/L), whereas during the growing season NH_4-N concentrations were generally less than 0.05 mg/L (Wilcoxon Rank Sum, $p < 0.05$).

In Lake Fayetteville, median TP concentrations were generally less than <0.1 mg/L throughout the year, but the medians showed an interesting bimodal pattern (Figure 2). Median TP in January and February was around 0.05 mg/L and slightly decreased in March and April (0.03 mg/L) (Figure 3; Wilcoxon Rank Sum, $p < 0.05$). Then in May and June, the median TP magnitude (0.5 to 0.9 mg/L) and variability increased. TP decreased in July and remained low (0.03 mg/L) until October. In November, median TP increased (0.06 mg/L) in magnitude only to decrease (0.04 mg/L) again in December (Figure 4; Wilcoxon Rank Sum, $p < 0.05$). Overall, median TP did not different between winter, spring, summer, or fall and was not different between off and growing seasons ($p > 0.05$).

Soluble reactive phosphorus was generally low in Lake Fayetteville throughout the study except for October to December 2019 where a peak occurred (Figure 2). For ten months of the year, median SRP concentrations were below the method detection limit (<0.005 mg/L) (Figure 3). January and November were the only two months where median SRP magnitudes were greater than the method detection limit. No statistical differences for median SRP concentrations were observed across months (Wilcoxon Rank Sum, $p > 0.05$). Overall, median SRP concentrations did not differ much between seasons (0.002 to 0.003 mg/L), but summer was less than winter and fall (Figure 4; Wilcoxon Rank Sum, $p < 0.05$). Furthermore, the off-season median SRP was different from the growing season ($p < 0.05$).

An indicator of algal productivity and biomass in Lake Fayetteville was chl-*a*, and in the summers of 2019 and 2020, chl-*a* peaked around 90 µg/L but in 2021 chl-*a* peaked around 150 µg/L (Figure 2). From January until April, median chl-*a* remain low (10 to 20 µg/L) (Figure 3; Wilcoxon Rank Sum, $p < 0.05$). In May, median chl-*a* increased in magnitude (58 µg/L) and variability. After May, the median chl-*a* magnitude decreased monthly from June (40 µg/L) to July (29 µg/L). From August to September, median concentrations were between 25 and 30 µg/L. Median chl-*a* decreased further in November and December to the lowest magnitude (5 µg/L). The seasonal differences were median chl-*a* was greatest in summer and spring, and least in winter (Figure 4; Wilcoxon Rank Sum, $p < 0.05$). The growing season had a greater median chl-*a* (30 µg/L), whereas during the off-season chl-*a* concentrations were less (13 µg/L) ($p < 0.05$).

Chlorophyll RFUs did not follow the same pattern in the lake as chl-*a*. From January to April, the median CHL ranged from 2,700 to 10,000 RFUs (Figure 3; Wilcoxon Rank Sum, $p < 0.05$). Median CHL RFUs decreased in May (2,700 RFUs) only to increase in variability in June. The CHL values increased in July (6,200 RFUs) only to remain lower in August and September

(4,200 RFUs). In October, the CHL magnitude increased once again (7,800 RFUs). Then, median CHL decreased in November (5,200 RFUs) and again in December (1,500 RFUs). November had the widest variability of CHL values. Overall, the greatest difference in median CHL RFUs were in the fall (5,400 RFUs) and spring (2,900 RFUs) (Figure 4; Wilcoxon Rank Sum, $p < 0.05$). The medians of the growing season and the off-season were not significantly difference ($p > 0.05$).

In addition to CHL, PC RFUs were also measured throughout this study. From January to March, median PC values were low (700 to 1,200 RFUs) with low variability (Figure 3; Wilcoxon Rank Sum, $p < 0.05$). Median PC magnitude increased in April (2,500 RFUs) and hit a peak in May (13,000 RFUs). This peak occurred during the same month as the peak for chl-*a* and the month before a peak in total microcystin concentrations. The median PC magnitude remained around 5,500 to 7,000 RFUs from June until September before decreasing monthly until December (300 RFUs). The greatest median PC values were in spring (2,200 RFUs), summer (5,900 RFUs), and fall (4,700 RFUs) which were not different from each other but significantly greater than winter (780 RFUs) (Figure 4; Wilcoxon Rank Sum, $p < 0.05$). The growing season had a greater median PC concentration (5,400 RFUs) compared to the off-season median (1,500 RFUs) ($p < 0.05$).

The PC:CHL ratio followed the same type of pattern throughout the three years, but in 2020 the ratio remained elevated (4 to 6) longer compared to 2021 where after the initial peak the ratio remained decreased (2 to 3) (Figure 2). Furthermore, the PC:CHL ratio in Lake Fayetteville followed a bimodal pattern where peaks in PC:CHL occurred a month before peaks in total microcystin (Figure 3). From January to March, the median ratio is low (<0.2) suggesting that the algal community was not dominated by cyanobacteria (Wilcoxon Rank Sum, $p <0.05$). The ratio elevated in magnitude during April (0.9) and May (4.1), where both months experienced ratios

reaching over 1 suggesting that the algal community was dominated by cyanobacteria. After the peak in May, median PC:CHL decreased in June (1.8) and July (0.8), only to increase again in August (1.1) and September (1.5). In October, the median PC:CHL ratio decreased from 0.6 to 0.2 in December. Overall, median PC:CHL was greatest in the summer and spring, second greatest in fall, and least in the winter (Figure 4; Wilcoxon Rank Sum, $p < 0.05$). The median PC:CHL was greater for the growing season (1.2) compared to the off-season (<0.2) ($p < 0.05$).

Quantifying Parameter Thresholds

Many parameters showed significant ($p < 0.05$) non-linear response to total microcystin (Table 2), and the thresholds explained more variation in total microcystin than corresponding linear correlations. For water temperature, a significant threshold with total microcystin was observed for all years and for each individual year ($p \leq 0.05$). The thresholds varied from 27°C for all years and 2020, 18°C in 2019, and 13°C in 2021. With each threshold regardless of the variation, mean total microcystin increased when temperature increased. For all years and 2020, mean total microcystin almost doubled when temperature was greater than 27°C.

The dissolved nutrient supply had several thresholds with total microcystin in this study. NO_3-N thresholds with total microcystin were significant ($p < 0.01$) for all years and for each individual year, varying between 0.3 and 0.7 mg/L. When NO_3-N concentrations were below the threshold, mean total microcystin increased. Unlike NO_3-N, dissolved NH_4-N thresholds with total microcystin were only significant ($p \leq 0.02$) for all years and in 2020. When NH_4-N was less than the threshold (0.005 mg/L), mean total microcystin concentrations increased. Since the main driver of DIN is NO_3-N ($r = 0.97$; $p < 0.01$), the thresholds with total microcystin were significant for all four timeframes, ranging from 0.5 to 1.1 mg/L; mean total microcystin increased as DIN was less than from the threshold. SRP thresholds with total microcystin were also significant ($p \leq 0.05$) for

all years and for each individual year with the thresholds ranging 0.004 to 0.006 mg/L. When SRP supply in the lake water was least, mean total microcystin was greatest.

As for total nutrients and ratios, several thresholds with total microcystin were significant ($p \leq 0.04$) in all years, 2019 and 2020. The thresholds for all years and 2020 were both around 1.3 mg/L whereas the threshold for 2019 was 2.0 mg/L. One interesting microcystin response with TN is that for 2019, mean total microcystin was greatest when TN exceeded the threshold, but for all years and 2020, mean total microcystin was less when TN exceeded the threshold. For TP, the threshold with total microcystin was significant ($p = 0.04$) only in 2020, where mean total microcystin increased from 0.8 µg/L to 1.0 µg/L when TP was less than the threshold. Moreover, in all years, 2019, and 2021, TP was not significantly ($p > 0.05$) linearly correlated with total microcystin concentrations. The molar TN:TP ratio threshold with total microcystin was only significant ($p < 0.05$) in all years, 2020, and 2021, but not in 2019; the thresholds ranged from 25 to 33 and mean total microcystin was greater when TN:TP ratios were less than the thresholds.

In this study, many algal growth parameters also had significant thresholds with total microcystin. The chl-*a* thresholds with total microcystin were significant ($p \leq 0.02$) across all years and each individual year. The threshold for all years and 2020 was 23 µg/L which is less than the thresholds for 2019 and 2021 (57 and 59 µg/L, respectively). When chl-*a* concentrations were greater than the thresholds, mean total microcystin increased.

As for the chlorophyll raw fluorescence units, the thresholds with total microcystin were significant ($p < 0.01$) for all years, 2019, and 2020, but not in 2021. The threshold for all years (4,200 RFUs) and 2020 (4,400 RFUs) were more similar when compared to 2019 (2,300 RFUs). Furthermore, mean total microcystin increased when CHL was greater than the thresholds.

Phycocyanin raw fluorescence unit thresholds with total microcystin were significant ($p < 0.01$) for all years and each individual year. The PC thresholds for all years and 2020 (4,500 RFUs) were similar to the CHL thresholds (4,200 to 4,400 RFUs). Furthermore, the 2019 PC threshold (2,000 RFUs) was also similar to the CHL threshold (2,300 RFUs). The threshold (6,900 RFUs) for 2021 was greater than the other years, and the threshold for CHL was not significant. Mean total microcystin increased when PC was greater than the thresholds for each timeframe.

The pH thresholds with total microcystin were also significant ($p \leq 0.03$) for all years and for each individual year, ranging from 8.2 to 9.1. In 2021, the mean total microcystin doubled from the yearly average of 0.2 µg/L to 0.4 µg/L when pH was greater than 9.1. Furthermore, mean total microcystin for all years, 2019, and 2021 also increased when pH exceeded the threshold.

Precipitation was another parameter with significant thresholds with total microcystin. Unlike the other parameters, the thresholds for precipitation at 24 hours, 72 hours, and 1 week increments prior to water samples being collected, with total microcystin were all significant ($p \leq 0.02$) for only 2021. The thresholds were 0.2 cm for 24 hours, 1.9 cm for 72 hours, and 1.2 cm for 1 week. While mean total microcystin increased to the right of the thresholds, the 72-hour parameter had the greatest difference in mean total microcystin concentrations when precipitation was less (0.1 µg/L) and greater (0.5 µg/L) than the threshold in 2021.

Classification and Regression Trees

When using all data (n = 365; 2019-2021), we observed five splits in the tree (Figure 5). The first split was with water temperature, when temperature was less than 27°C total microcystin had a mean concentration of 0.32 µg/L. The next split occurred in the data with water temperature greater than 27°C; this split separated 2020 from 2019 and 2021. If the year was 2019 or 2021, this

led to a third split in the data with TN. The mean total microcystin was 2.22 µg/L when TN was greater than or equal to 1.7 mg/L or 0.5 µg/L when TN was less than 1.7 mg/L. If the year was 2020, a fourth split occurred with the data relating to CHL. If CHL, was less than 3,570 RFUs the mean total microcystin was 0.42 µg/L. When CHL was greater than the threshold, a fifth split popped out relating to TN:TP (molar). When the molar TN:TP ratio was lower than 59, mean total microcystin concentration was 3.48 µg/L. If TN:TP was greater than the threshold, mean total microcystin concentration was 1.90 µg/L. Overall, this model explained 60% of the variability in total microcystin concentrations across all three study years.

The 2019 model (n = 105) was the simplest in the study with only two splits (Figure 6). The first split was with pheophytin, which relates to algal growth and is essentially chlorophyll without central magnesium (Mg^{2+}) ions. When pheophytin was greater than 5.2 µg/L, mean total microcystin was greatest at 1.43 µg/L. If pheophytin was less than the threshold, a second split occurred with DIN at 0.68 mg/L. If DIN was less than the threshold, mean total microcystin was 0.73 µg/L, and when DIN was greater than or equal to the threshold the mean total microcystin was 0.18 µg/L. Overall, this model explained 32% of the variability in total microcystin concentrations in 2019.

For the 2020 model (n = 129), the parameters for three out of four splits overlapped with the all years model (Figure 7). The first split was with water temperature and the threshold (27°C) was the same as the all years model. If water temperature was less than the threshold, a second split occurred with season. The split for season was the only split that was different than the all years model. If the season was winter and spring, mean total microcystin was 0.08 µg/L, and if the season was summer and fall mean total microcystin was 0.83 µg/L. If water temperature was greater than or equal to 27°C, a third split popped out with CHL at 3,570 RFUs. If CHL was less

than the threshold, mean total microcystin was 0.42 µg/L and if higher, a fourth split occurred with molar TN:TP. The threshold was slightly lower in 2020 (51) compared to the all years model (59), but mean total microcystin concentration to the left (3.48 µg/L) and right (1.90 µg/L) were the same between the two models. Overall, this model explained 76% of the variability in total microcystin concentrations in 2020.

The 2021 model (n = 128) was the second simplest model with three splits (Figure 8). The first split was with precipitation received within the last 72 hours and the threshold was at 0.75 cm. When 72-hour precipitation was greater than 0.75 cm, mean total microcystin was greatest at 0.45 µg/L. If precipitation was less than the threshold, a second split occurred with chl-*a* at 62 µg/L. To the right of the threshold, mean total microcystin was 0.42 µg/L and if chl-*a* was less than the threshold, a third split occurred with NH_4-N. When NH_4-N was greater or equal to 0.32 mg/L, mean total microcystin was 0.27 µg/L, and if NH_4-N was less than the threshold, mean total microcystin was lowest at 0.09 µg/L. Overall, this model explained 45% of the variability in total microcystin concentrations in 2021.

Discussion

Microcystin concentrations and physicochemical processes in lakes are dynamic, changing within one year, between years, and spatially (Kotak et al., 1993; Jacoby et al., 2000). At Lake Fayetteville, we observed varying peak mean microcystin concentrations (1.9 to 6.2 μg/L) and longevity (consecutive days with mean concentrations >2 μg/L according to WHO guidelines for waterbodies with recreational uses (WHO, 2003; 0 to 68 days) which changed from year to year. The greatest peak in mean concentration was in 2019, but the longest longevity of mean toxin concentrations was in 2020. While mean concentrations at Lake Fayetteville did not violate the EPA guidelines for total microcystin in recreational waters (USEPA, 2019; 8 μg/L), two individual samples (11 and 15 μg/L) did in 2019 (Wagner et al., 2021). While some studies observed that microcystin concentration is directly related to cyanobacterial biomass, other studies suggested that maximum peaks in microcystin concentrations do not correlate to biomass of toxin producing genera (Kotak et al., 1995; Watanabe et al., 1994). In this study, a myriad of factors like nutrients and environmental conditions showed relationships with total microcystin concentrations. Moreover, the factors which may increase toxin production and concentration for one year may change in the next year (Jacoby et al., 2000). When looking at all three years of data, the main factors associated with increased microcystin concentrations at Lake Fayetteville were increased water surface temperatures, elevated algal biomass, low inorganic N concentrations, and molar TN:TP ratio.

An increase in cyanobacteria growth from increased surface water temperatures (>15°C) has been documented extensively (McQueen and Lean, 1987; Paerl et al., 2001; O'Neil et al., 2012; Giannuzzi, 2018). In fact, the dominant cyanobacteria genera can shift across temperature changes between *Anabaena, Aphanizomenon,* and *Microcystis* (Robarts and Zohary, 1987; Paerl

et al., 2001). All three of these toxin-producing genera have been observed at Lake Fayetteville in the spring where temperatures range from 14 to 28℃ (Meyer, 1971). When below the surface water temperatures at Lake Fayetteville exceeded the thresholds (13 to 27℃), total microcystin concentration increased. Furthermore, each yearly peak in mean total microcystin occurred in June when water temperatures exceeded 27℃, except in 2021 when water temperature was only 21℃. Previous studies have showed that microcystin concentrations increase with warmer water temperatures, with a maximum around 20 to 30℃ (Davis et al., 2008; Walls et al., 2018). While water temperature is an important factor in cyanobacteria growth toxin concentrations, many other factors can also influence cyanobacteria growth and toxin production.

In many lakes throughout the United States, chl-*a* is often one of the best variables to predict microcystin concentrations (Yuan et al., 2014; Lee et al., 2000; Gram et al., 2006; Yoshida et al., 2007). When chl-*a* concentrations at Lake Fayetteville were greater than the thresholds (23 to 59 μg/L), total microcystin increased. A previous study on six reservoirs in Brazil calculated a chl-*a* threshold of 40 μg/L for when microcystin was >1 μg/L, which was similar to the thresholds observed in this study (Cunha et al., 2018). Furthermore, in many lakes in the United States had a 50% change of microcystin values >1 μg/L when a chl-*a* was greater than 67 μg/L (Hollister and Kreakie, 2016). While this threshold was increased compared to the thresholds in our study, chl-*a* concentrations above 67 μg/L were observed in Lake Fayetteville. Overall, chl-*a* concentrations can be a helpful tool in understanding algal biomass, but since cyanobacteria have other pigments like PC, the correlation between chl-*a* and cyanobacteria may vary (Cunha et al., 2018).

When predicting cyanobacteria dominance and growth, PC RFUs may be easier and more accessible for lake managers than chl-*a* pigment concentrations. The CyanoFluor is a rapid and relatively inexpensive field monitoring tool, which analyzes for PC RFUs (Turner Designs, 2017).

Furthermore, strong linear relationships between PC RFUs and cyanobacteria biomass have been observed (Bertone et al., 2018; Thomson-Laing et al., 2020). At Lake Fayetteville total microcystin concentrations increased when PC RFUs were greater than the thresholds (2,000 to 6,900 RFUs). The CyanoFluor also provides a PC:CHL ratio to understand the proportion of the community that is dominated by cyanobacteria. In Lake Fayetteville, when the PC:CHL ratio exceeded 0.4, microcystin concentrations increased. While cyanobacteria growth is essential to monitor, the conditions that support growth may be different than the conditions that affect toxin production (Sinang et al., 2013).

One nutrient prominent in anthropogenic eutrophication is N, which is essential for cyanobacterial growth and toxin production (Horst et al., 2014). Many cyanobacteria can use both DIN (including NO_3-N and NH_4-N) and dissolved organic forms like urea (Donald et al., 2011; Paerl, 1988). Previous research has focused on NO_3-N and NH_4-N inputs because of the prominent use of inorganic fertilizers, although inputs from urea fertilizers and animal wastes are becoming more common (Gobler et al., 2016; Paerl et al., 2016). Furthermore, urea has the potential to become a source of DIN in lakes, because it can hydrolyze into two NH_4-N ions. DIN was more likely to stimulate the growth of toxic strains of *Microcystis* compared to dissolved organic forms which stimulated the growth of non-toxic strains (Davis et al., 2010). While some studies suggest that N form affects cyanobacteria growth (e.g., O'Neil et al., 2012; Ha et al., 2009), other studies suggest that the magnitude of the N supply matters more (Wagner et al., 2021).

In this study, we measured two different types of inorganic N, NO_3-N and NH_4-N. Our thresholds for NO_3-N were 0.3 to 0.7 mg/L and total microcystin concentrations were greater when NO_3-N supplies were low (< 0.3 mg/L). However, the timing of the microcystin peak relative to NO_3-N being < 0.3 mg/L varied between years. Previous research has mixed results on the effect

of NO_3-N on microcystin concentrations. In one study looking at blooms on four rivers in China, NO_3-N was lower in stagnant areas that showed increased algal growth and microcystin concentrations (He et al., 2021). In another study in Brazil, NO_3-N concentration did not significantly affect microcystin concentrations (Cunha, et al., 2018).

Ammonia is the most energetically favored form of DIN since the ion is not converted into another form for diffusion into the cell (Takamura et al., 1987; Yoshida et al., 2007). Previous studies have suggested that low concentrations of NH_4-N (0.007 to 0.025 mg/L) are sufficient to support *Microcystis* blooms (Jacoby et al., 2000; Chaffin et al., 2011). As for NH_4-N in Lake Fayetteville, nCPA thresholds (0.005 mg/L) were only significant for the all years and 2020, and if NH_4-N was less than the threshold, total microcystin concentrations increased. This negative correlation with NH_4-N and microcystin concentration was previously observed on Lake Erie (Rinta-Kanto et al., 2009). This is different than the RT split for NH_4-N in the 2021 where mean total microcystin increased from 0.09 to 0.27 μg/L when NH_4-N exceeded the threshold (0.32 mg/L). However, the RT split is not directly comparable to the thresholds since the data is constrained by previous splits.

Another way lakes receive N inputs is through wet deposition during precipitation events. A previous study calculated a total N input of 1.3 $g/m^2\ yr^{-1}$ from wet deposition into Lake Fayetteville, which was considerably less than the calculated watershed inputs (29 $g/m^2\ yr^{-1}$) (Grantz et al., 2014). Typically, precipitation events occur in fall and spring and increase inorganic N concentrations. An excess of DIN in the water favors the growth of non-nitrogen fixing cyanobacteria (nondiazotrophs) that are more likely to produce toxins (Carmichael, 1994; Schindler et al., 2008; Paerl et al., 2001). Nondiazatrophs will then reduce DIN supplies through uptake for metabolic processes like growth or toxin production. After spring and into summer

precipitations decreases leading to a decrease in N inputs. A change in cyanobacteria community to nitrogen fixing cyanobacteria (diazotrophs) may occur during the summer since DIN in the system is depleted (Paerl, 1988; Flett et al., 1976). Since some diazotrophs are unable to produce toxins, a dominance of diazotrophs in the algal community could lead to a decrease in total microcystin in the water (Paerl et al., 2001).

The other nutrient observed in excess due to eutrophication is P. In previous studies, SRP was positively correlated with cyanobacteria growth rates and microcystin concentrations (Jacoby et al., 2000; Davis et al., 2009; Rinta-Kanto et al., 2009). At Lake Fayetteville, when SRP concentrations were lower than the thresholds (0.003 to 0.006 mg/L), total microcystin increased. With consistently low SRP concentrations (< 0.02 mg/L) throughout the year, but an increase in algal growth during the summer, the algae may immobilize P as soon as it becomes available reducing measurable SRP in the water (Austin et al., 2020).

TN also showed mixed results with total microcystin concentrations. Some previous studies observed a positive correlation between TN and microcystin (Yuan et al., 2014; Rolland et al., 2005; Giani et al., 2005), while others observe a negative correlation (Rinta-Kanto et al., 2009; Conradie and Barnard, 2012) or no correlation (Jacoby et al., 2015; Cunha et al., 2018). In Lake Fayetteville, the microcystin response to TN concentrations varied depending on the year. In 2019, if TN was greater than the threshold (2.0 mg/L), total microcystin increased. If the all years and 2020 thresholds (1.2 to 1.4 mg/L) were not exceed, total microcystin increased. The Lake Fayetteville thresholds (1.2 to 2.0 mg/L) were similar to other calculated TN thresholds of 1.6 to 1.8 mg/L (Cunha et al., 2018; Shan et al., 2020). While worldwide thresholds can be beneficial in understanding general trends with microcystin concentrations, the response from cyanobacteria varies across waterbodies and years.

Many studies have not observed a correlation between TP and microcystin concentrations (Shang et al., 2015; Jacoby et al., 2015; Conradie and Barnard, 2012). In this study, the TP threshold with microcystin was only significant 2020, and if TP was lower than the threshold (0.06 mg/L), total microcystin increased. A lack of response in microcystin concentration with TP may be because TP includes immobilized organic P which not readily available for uptake by algae.

The effect of TN:TP ratios on cyanobacteria growth has also been studied intensively, and typically when the TN:TP ratio decreases, microcystin concentrations increased (Yuan et al, 2014; Jacoby et al., 2015). At Lake Fayetteville, when TN:TP were less than the thresholds (54 to 72), the total microcystin concentrations increased. Furthermore, microcystin concentrations were elevated at molar TN:TP ratios of 40 to 70. The low end of our molar TN:TP ratios with elevated microcystin concentrations overlapped with a meta-analysis where the greatest microcystin concentrations occurred around molar TN:TP ratios of 33 to 44 (Scott et al., 2013). Furthermore, another study calculated a molar TN:TP threshold of 121, where a ratio lower than 121 had a greater probability of producing a microcystin concentration ≥ 1 μg/L (Cunha et al., 2018).

Overall, many physico-chemical parameters showed significant thresholds at Lake Fayetteville, and lake managers could use certain thresholds in this study to monitor the lake and create a HAB warning system when those thresholds were breached. The most rapid and relatively inexpensive parameters are water temperature, with a threshold of 13℃ and paying special attention when temperatures are > 27℃, and PC:CHL ratios using a CyanoFluor, where a ratio greater than 0.4 which suggests the community is consists primarily of cyanobacteria. If the managers had access to a water quality lab, information regarding the concentrations the two forms of DIN, when DIN is low (< 0.3 mg/L), and if molar TN:TP ratios are within 40 to 70 would also help in predicting HABs and the conditions necessary for increased toxin production.

Conclusion

Eutrophication and global climate change will continue to exacerbate the negative effects of harmful algal blooms, primarily toxin production, which is a human health risk. At Lake Fayetteville, peak mean total microcystin concentrations and toxin production longevity both varied between years. Changes in microcystin concentrations between years could suggest a shift in the algal community and future research is necessary to understand the cyanobacteria genera present before, during, and after peak toxin production.

During this study, we analyzed a multi-year database of various physicochemical water quality parameters to propose thresholds with total microcystin. While some factors (water temperature and TN:TP ratio) aligned with previous research, other factors showed mixed results (SRP and TN). In our dataset, both nutrient and environmental conditions showed thresholds with total microcystin suggesting that the factors influencing HAB growth and toxin production are complex. Overall, two easy to measure parameters (water temperature and PC:CHL ratio) produced significant thresholds suggesting lake managers could monitor the parameters to help predict a HAB capable of increased toxin production.

The regression tree models for Lake Fayetteville also varied in the number (2 to 5) of splits, parameters related to total microcystin, and in the variation (32 to 76%) in total microcystin the models explained. To have an accurate regression tree model for all years, further data is needed to accurately assess the water quality parameters that influence microcystin concentrations over many years or if they always vary between years. An understanding of the internal factors that could be controlled and the external factors that could help predict elevated toxin production would ultimately help preserve and protect human health.

Table 1. Analytes, abbreviations, lab methods, equipment, and method detection limits (MDLs) for water samples collected from Lake Fayetteville and delivered to the water quality lab within the Arkansas Water Resources Center; more information can be found at https://awrc.uada.edu/water-quality-lab/certification-and-quality-assurance/.

Analyte	Abbreviation	Method	Equipment	MDLs
Ammonium-N	NH_4-N	EPA 351.2	Skalar Sans ++ Wet Chemistry Analyzer	0.017 mg/L
Nitrate+Nitrite-N	NO_3-N	EPA 353.2	Skalar Sans ++ Wet Chemistry Analyzer	0.014 mg/L
Soluble Reactive Phosphorus	SRP	EPA 365.1	Skalar Sans ++ Wet Chemistry Analyzer	0.005 mg/L
Total Nitrogen	TN	APHA 4500-P J; EPA 353.2	Skalar Sans ++ Wet Chemistry Analyzer	0.01 mg/L
Total Phosphorus	TP	APHA 4500-P J; EPA 365.1	Skalar Sans ++ Wet Chemistry Analyzer	0.010 mg/L
Chlorophyll-*a*	Chl-*a*	APHA 10200 H3	Turner Design Fluorometer	0.5 µg/L
Pheophytin		APHA 10200 H3	Turner Design Fluorometer	
Chlorophyll	CHL		CyanoFluor Handheld HAB Indicator	N/A RFU
Phycocyanin	PC		CyanoFluor Handheld HAB Indicator	N/A RFU
Total Microcystin		EPA 546	Abraxis Plate Reader	0.06 µg/L

RFU = Raw Fluorescence Unit

Table 2. Top ten (based on R^2) thresholds using nonparametric change point analysis (nCPA) between select physico-chemical and algal parameters with total microcystin (MC) concentrations at Lake Fayetteville across all data and individual calendar years (2019, 2020, and 2021).

Year	Variable	Units	Threshold	Median	*P*	R^2	Mean MC < Threshold (µg/L)	Mean MC ≥ Threshold (µg/L)
All Years	PC*	RFU	4530	4530	0.001	0.177	0.234	0.916
	Temp	℃	27.0	27.0	0.001	0.172	0.315	1.062
	DIN	mg/L	0.53	0.53	0.001	0.169	0.883	0.182
	DIN%		27.7	27.7	0.001	0.160	0.908	0.230
	NO_3-N	mg/L	0.50	0.50	0.001	0.157	0.844	0.162
	CHL-*a*	µg/L	23.4	23.4	0.001	0.112	0.252	0.821
	PC:CHL*		0.37	0.37	0.002	0.106	0.154	0.704
	CHL*	RFU	4426	4477	0.001	0.105	0.281	0.801
	pH		8.58	8.58	0.001	0.087	0.314	0.816
	Conductivity	µS/cm	158	159	0.001	0.086	0.917	0.389
2019	PC*	RFU	1989	2348	0.001	0.413	0.176	0.684
	CHL*	RFU	2272	2278	0.001	0.337	0.155	0.649
	PC:CHL*		0.39	0.39	0.001	0.235	0.264	0.647
	PHEO	µg/L	5.18	5.18	0.001	0.208	0.559	1.425
	DIN%		49.5	49.5	0.003	0.202	0.916	0.218
	DIN	mg/L	0.71	0.71	0.003	0.191	0.892	0.187
	NO_3-N	mg/L	0.69	0.69	0.004	0.191	0.892	0.187
	TN	mg/L	1.95	2.00	0.005	0.177	0.583	1.505
	CHL-*a*	µg/L	57.3	36.9	0.003	0.168	0.615	1.479
	pH		8.20	8.20	0.008	0.150	0.303	0.902
2020	Temp	℃	27.0	27.0	0.001	0.395	0.354	1.930
	NO_3-N	mg/L	0.32	0.27	0.001	0.287	1.415	0.179
	DIN	mg/L	0.45	0.44	0.001	0.281	1.391	0.166
	DIN%		28.1	25.2	0.001	0.261	1.414	0.238
	TN	mg/L	1.37	1.31	0.001	0.259	1.312	0.118
	Supply N:P		84.1	84.1	0.001	0.213	1.406	0.229
	PC	RFU	4524	4524	0.001	0.179	0.265	1.251
	PN:PP		41.5	41.3	0.002	0.165	0.367	1.302
	Conductivity	µS/cm	159	159	0.001	0.145	1.316	0.434
	PC:CHL		0.37	0.43	0.001	0.138	0.129	1.092
2021	72 HR Precip	cm	1.91	1.91	0.001	0.271	0.140	0.452
	PC:CHL		2.39	2.15	0.001	0.237	0.155	0.533
	CHL-*a*	µg/L	59.1	59.1	0.002	0.214	0.155	0.498
	NO_3-N	mg/L	0.71	0.71	0.003	0.173	0.242	0.056
	PC	RFU	6909	6719	0.003	0.169	0.161	0.496
	DIN%		66.6	66.6	0.007	0.134	0.236	0.073
	24 HR Precip	cm	0.09	0.09	0.005	0.114	0.152	0.338
	DIN	mg/L	1.14	1.12	0.010	0.110	0.219	0.047
	pH		9.12	9.12	0.021	0.098	0.159	0.351
	TN:TP	mass	24.6	24.7	0.032	0.086	0.294	0.150

*These parameters with 2019 and all years had fewer observations because the CyanoFluor was not used consistently until June 25, 2019.

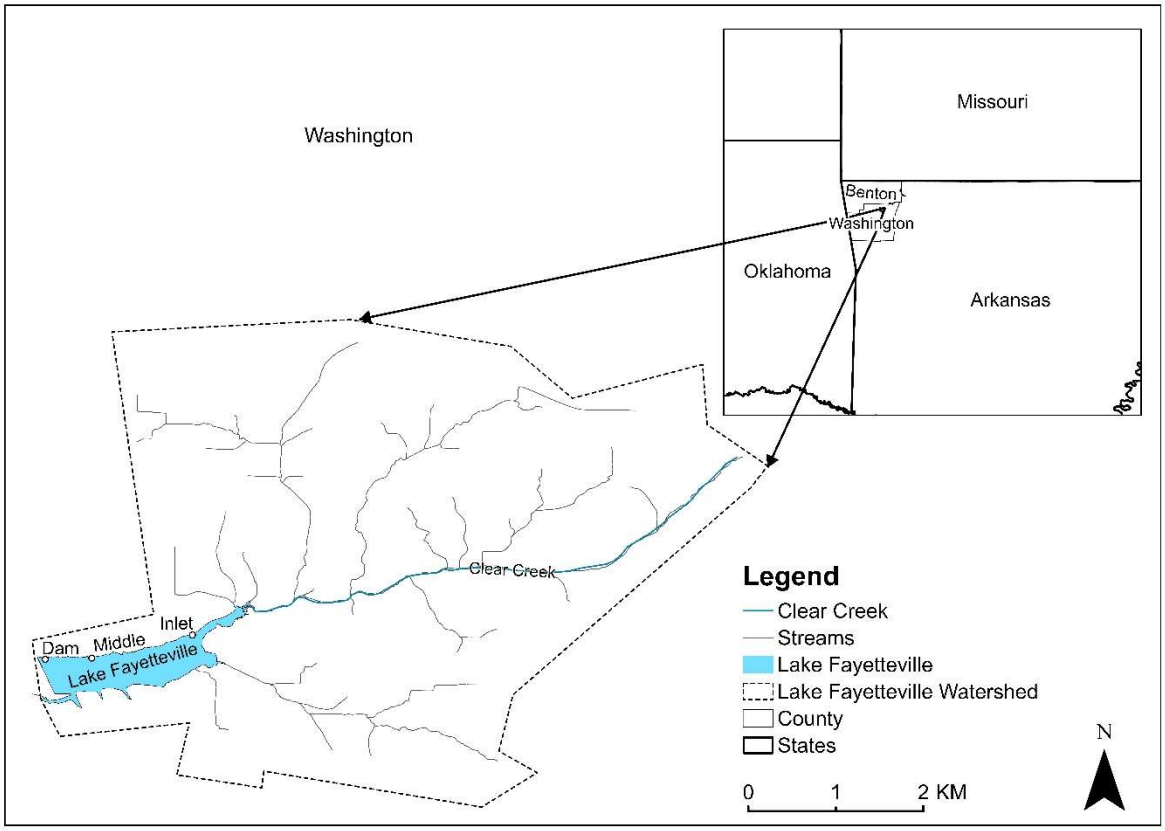

Figure 1. Lake Fayetteville and the Lake Fayetteville watershed in Arkansas; the dam, middle, and inlet were sampling sites from 2019 to 2021.

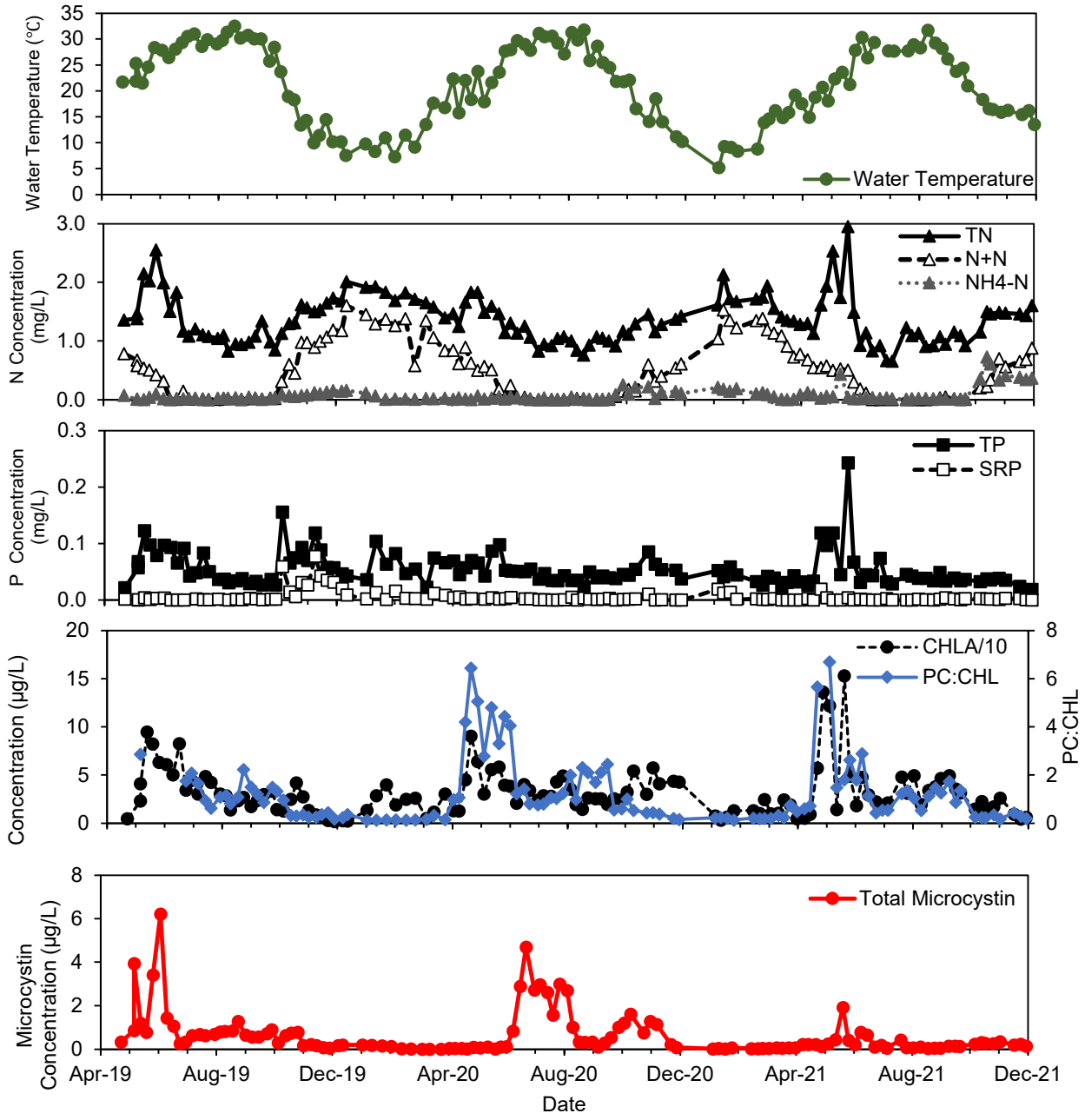

Figure 2. Select physicochemical water quality parameters averaged from three sampling sites at Lake Fayetteville, Arkansas graphed against time (April 2019 to December 2021).

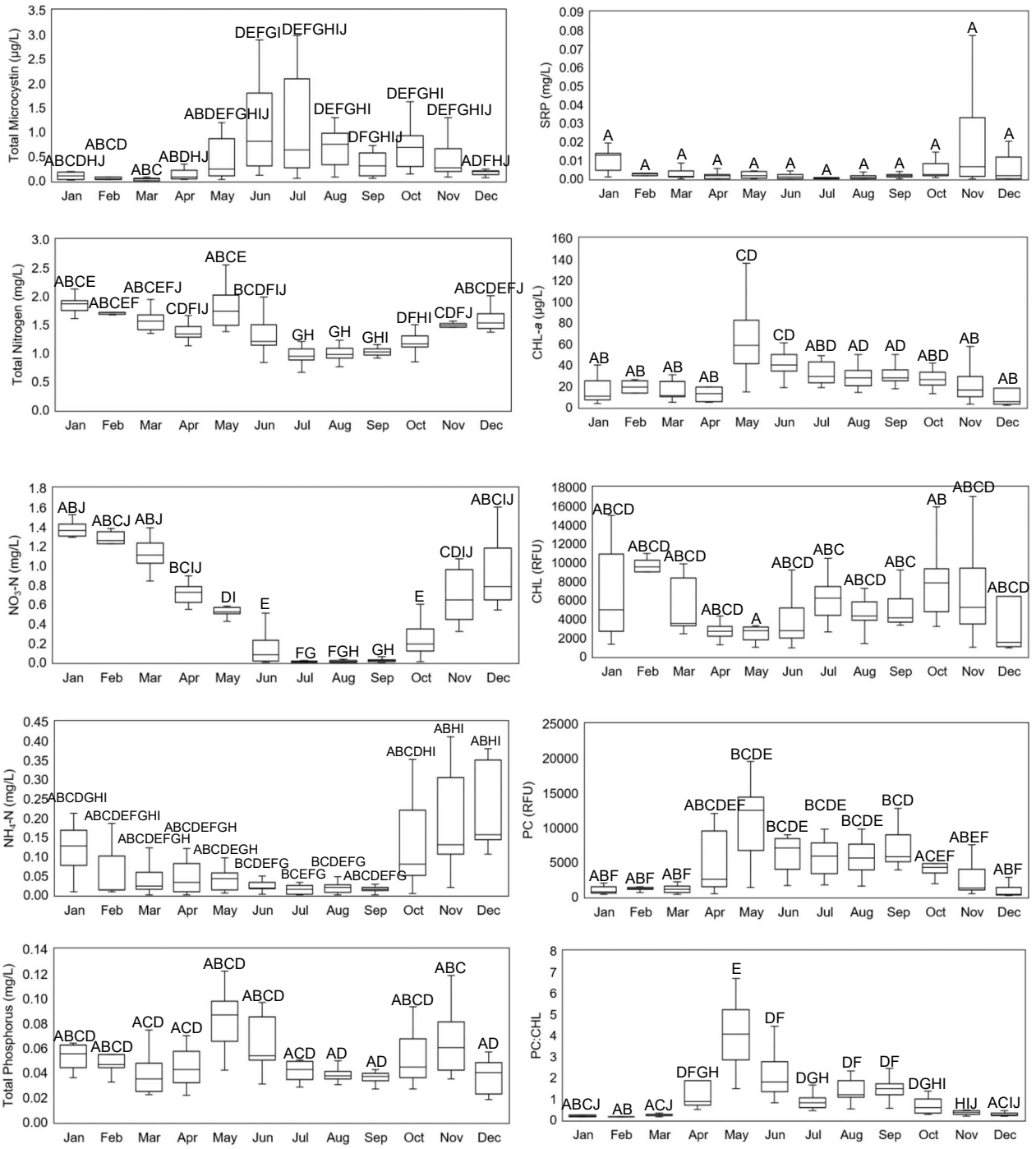

Figure 3. Box and whisker plots selected water quality parameters from 2019 to 2021 at Lake Fayetteville plotted in monthly increments; different letters across months show significant ($p < 0.05$) differences in monthly medians.

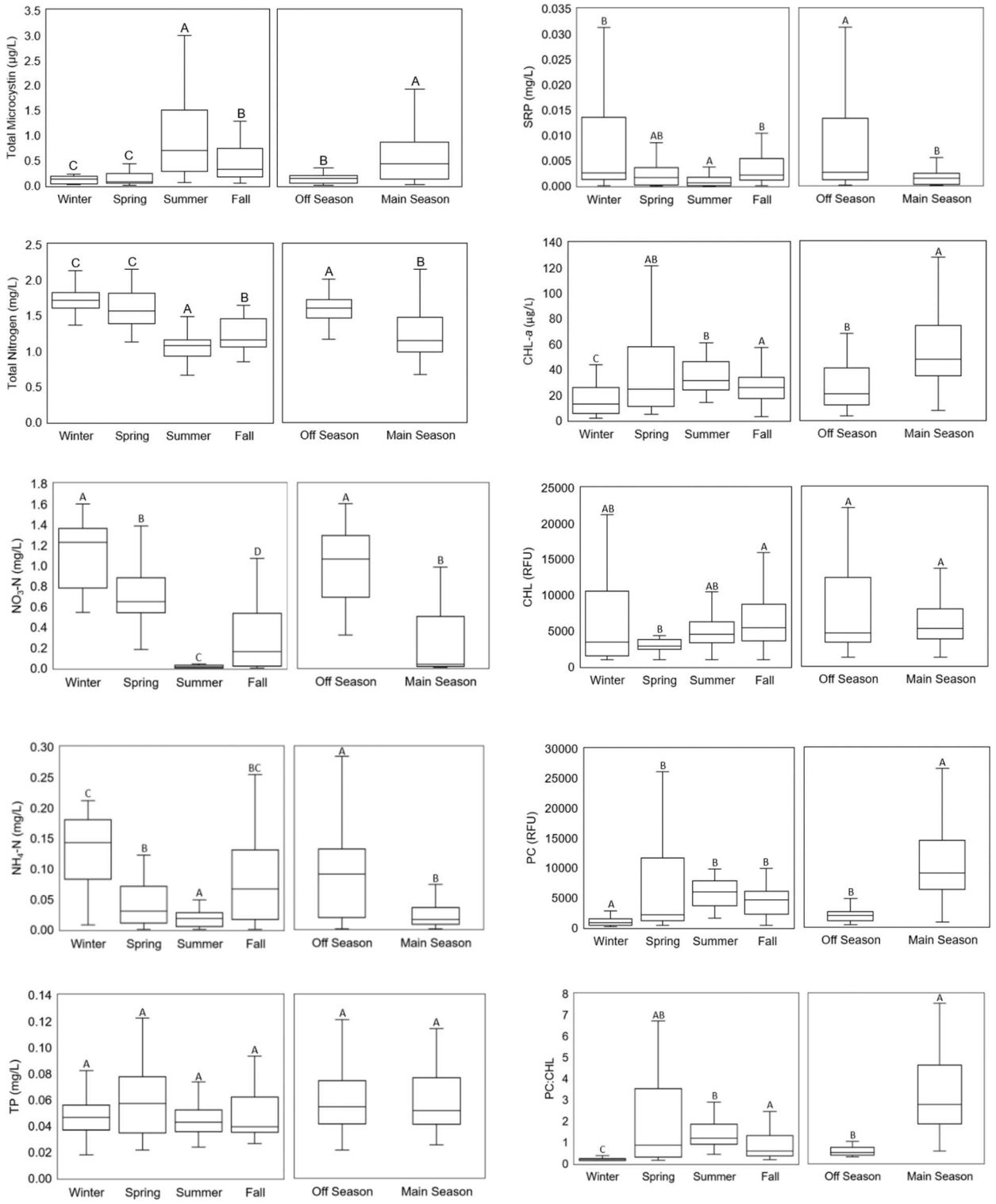

Figure 4. Box and whisker plots selected water quality parameters from 2019 to 2021 at Lake Fayetteville plotted in meteorological and algal growing seasons increments; different letters across seasons show significant ($p < 0.05$) differences in season medians.

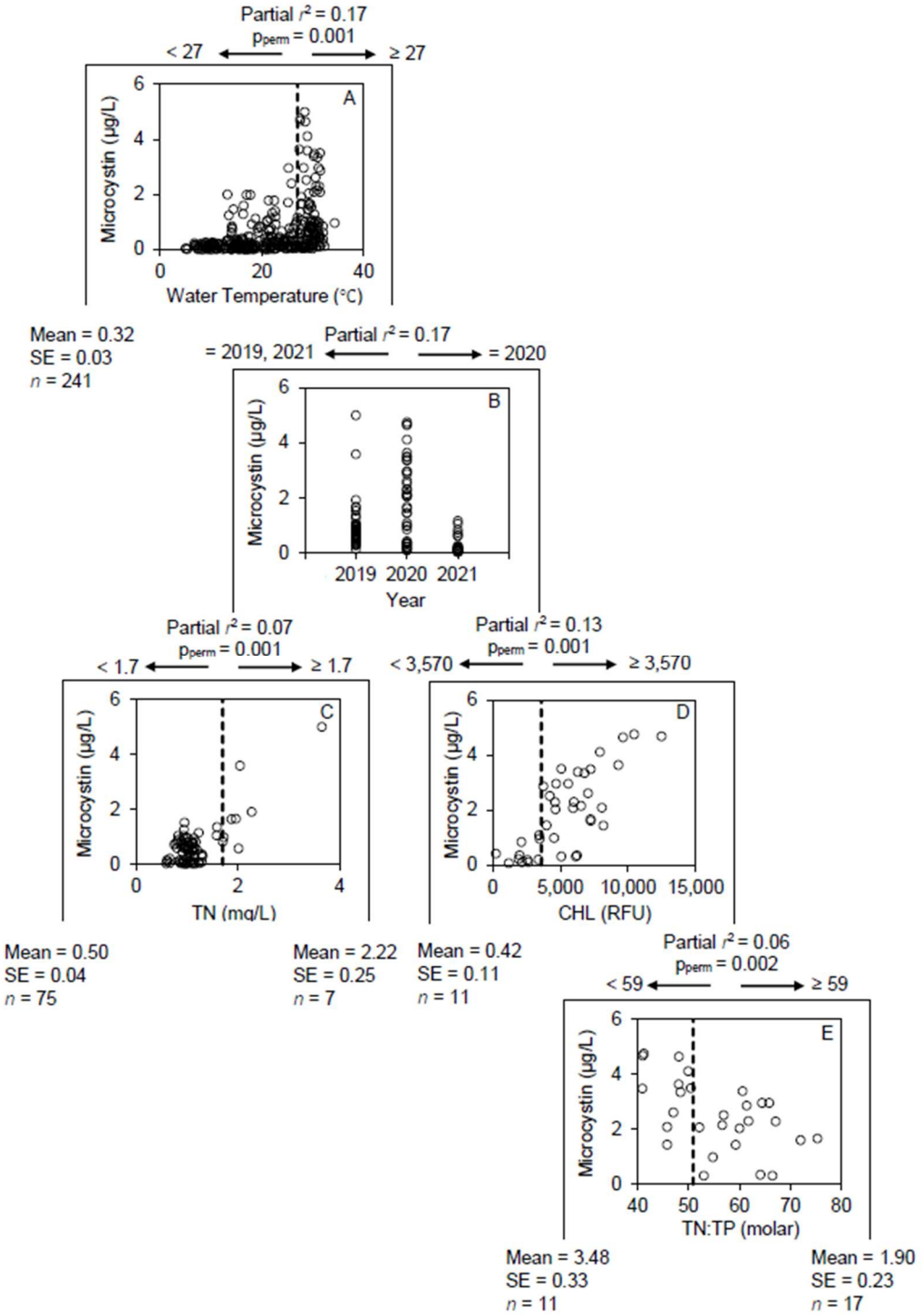

Figure 5. Classification and regression tree model for all years (2019 to 2021) at Lake Fayetteville showing major change points (dashed vertical lines) related to water temperature (°C) just below surface, year, total nitrogen (mg/L), chlorophyll raw fluorescence units (CHL RFUs), and total nitrogen to total phosphorus ratio (molar TN:TP).

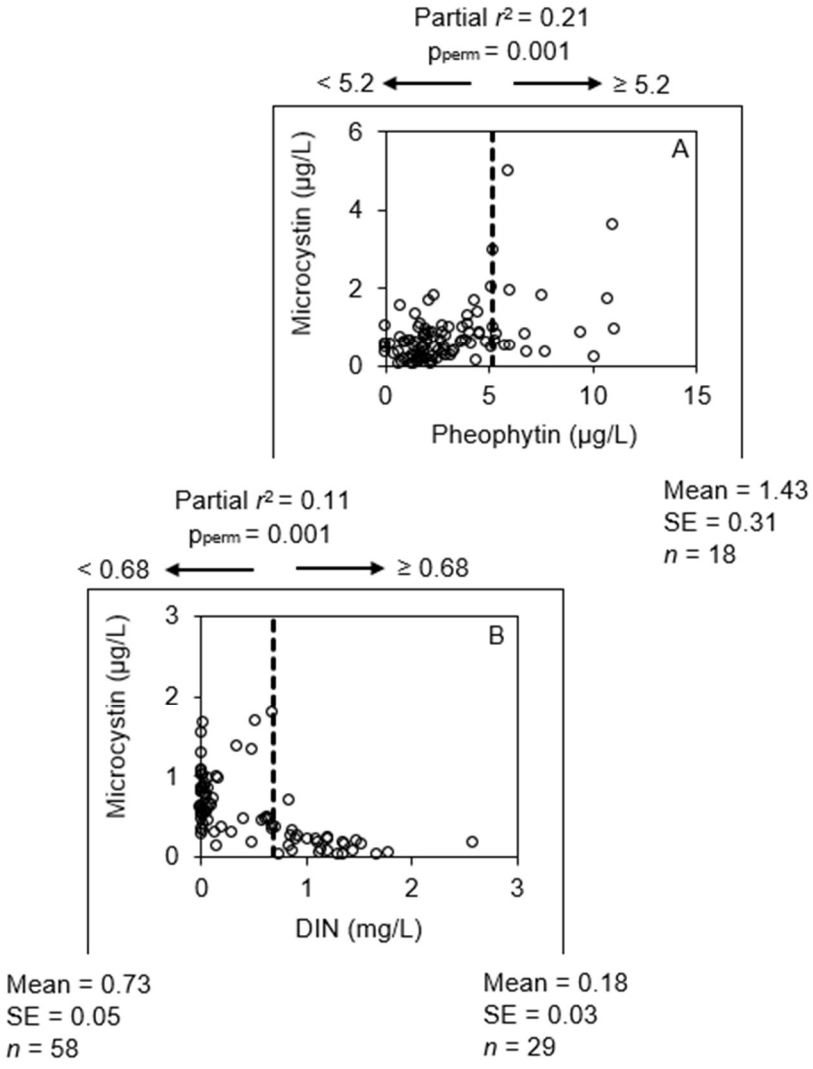

Figure 6. Classification and regression tree model for 2019 at Lake Fayetteville showing major change points (dashed vertical lines) related to pheophytin (µg/L) and dissolved inorganic nitrogen (DIN, mg/L).

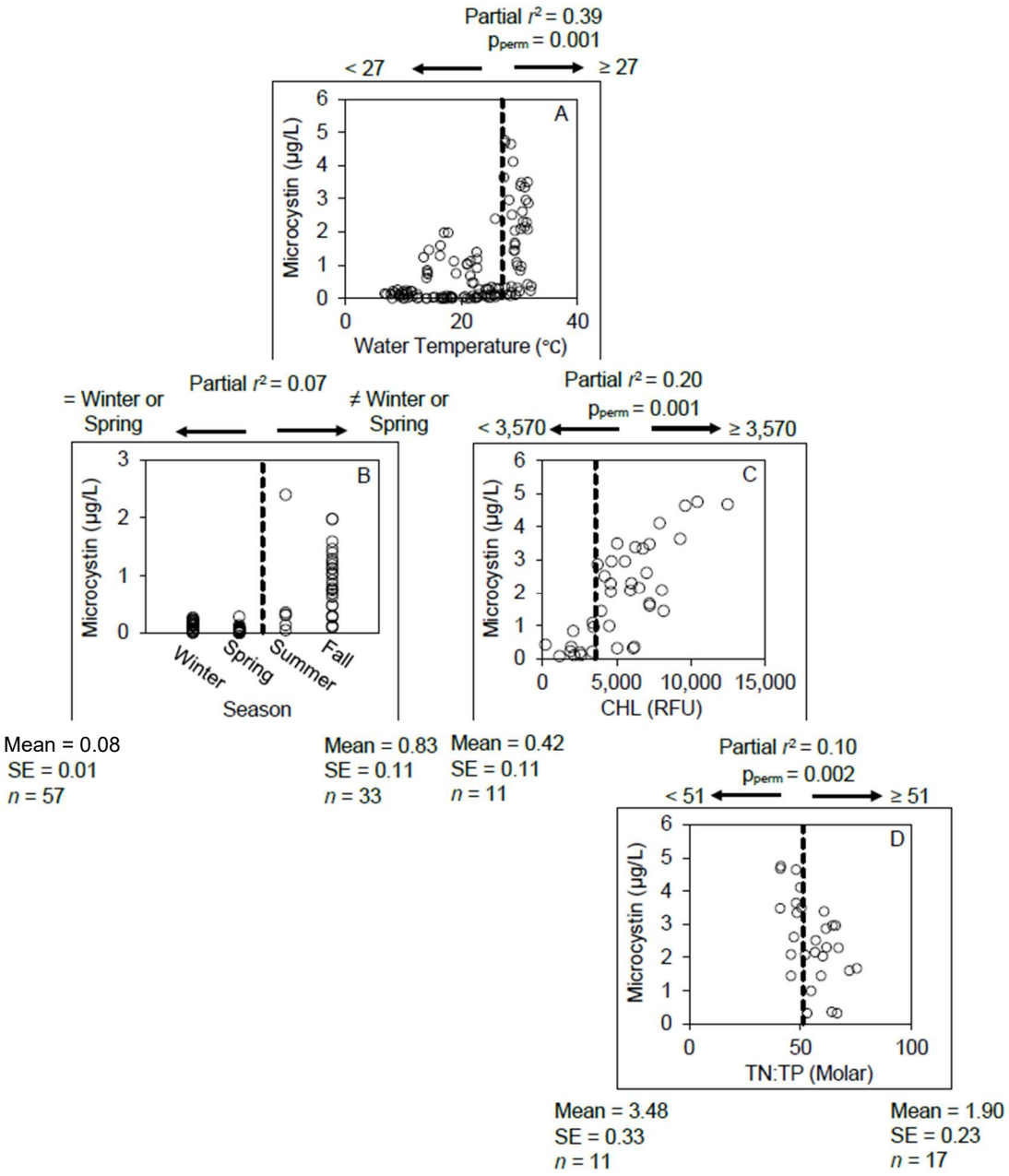

Figure 7. Classification and regression tree model for 2020 at Lake Fayetteville showing major change points (dashed vertical lines) related to water temperature (°C) just below surface, season (winter: December 1 to February 28; spring: March 1 to May 31; summer: June 1 to August 31; fall: September 1 to November 30), chlorophyll raw fluorescence units (CHL RFUs), and total nitrogen to total phosphorus ratio (molar TN:TP).

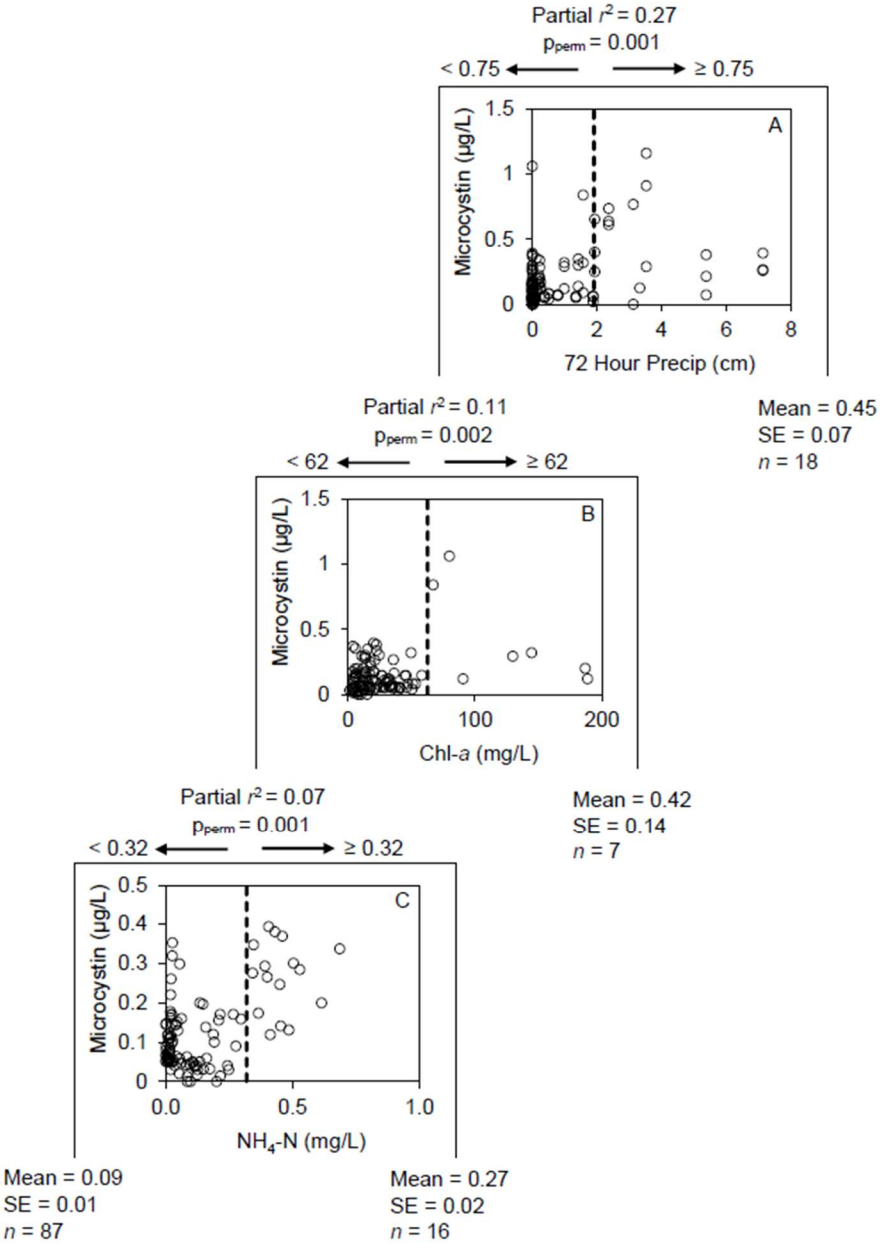

Figure 8. Classification and regression tree model for 2021 at Lake Fayetteville showing major change points (dashed vertical lines) related to precipitation within the previous 72 hours (72-hour precip, cm), chlorophyll-*a* (chl-*a*, µg/L), and dissolved ammonia-N (NH_4-N, mg/L).

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