

TRACKING AND MANAGING NON-POINT SOURCE POLLUTION AT LAKE HERRICK
WATERSHED, ATHENS, GEORGIA

by

ASHWINI KANNAN

(Under the Direction of David Elliot Radcliffe)

ABSTRACT

The Lake Allyn M. Herrick watershed is about 131 ha and covers portions of the University of Georgia's East campus, the Oconee Forest and residential and commercial land use. Lake Herrick, a 6-ha water body established in 1982 on the University of Georgia campus, was closed in 2002 for recreation due to fecal contamination. Subsequent monitoring confirmed cyanobacterium blooms on the surface of the lake and elevated nutrient concentrations, especially phosphorus. Previous studies showed that phosphorus and fecal coliform were the main contaminants. In our study, two inflow tributaries (sites Birdsong and Armadillo) and the outlet stream (site Below Dam) were monitored for discharge, *E. coli*, forms of nitrogen and phosphorus, and other water quality parameters during baseflow and storm conditions from February 2016 to October 2017. Our results showed that total phosphorus was significantly higher during stormflow compared to baseflow and total nitrogen remained the same. *E. coli* results indicated that most of the bacteria entered the lake through the tributaries during stormflow. Microbial source tracking methods were used to detect the bacterial source in the samples specific to a dog, ruminant or human host. We found that dogs are a more likely source of this bacteria than humans or deer. The fact that human sources were uncommon in the Lake

Herrick watershed indicated that there was reduced risk for human source contamination. Lake Herrick was reopened for limited recreation in October 2018.

INDEX WORDS: water quality, nutrients, eutrophication, urban streams, fecal contamination, microbial source tracking

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B.E, College of Engineering, Guindy, 2015

A Thesis Submitted to the Graduate Faculty of The University of Georgia in Partial Fulfillment
of the Requirements for the Degree

MASTER OF SCIENCE

ATHENS, GEORGIA

2018

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December 2018

DEDICATION

I would like to dedicate this thesis to my mother Meena and father Kannan who have guided me through every little thing in my life. This wouldn't have been possible without your continued support and love.

ACKNOWLEDGEMENTS

I would like to thank my co-major professor and mentor Dr. Radcliffe for his guidance throughout my time at UGA. I am very thankful to him for introducing me to various research projects and helping me gain the exposure in this field. Special thanks to my major professor Dr. Das for his timely input regarding academics and coursework. I would also like to thank Dr. Bledsoe and Dr. Hawkins for serving on my committee and providing valuable suggestions though this research project.

Special mention to Thalika Saintil who has been the best research partner. Thanks for being always there right from walking to collect the samples to processing it in the lab and even later to work on homework's. Thanks to Professor Rasmussen for helping us with the equipment, field work and valuable suggestions for the past two years. I would like to mention the members of the EPA ORD laboratory for agreeing to run our microbial source tracking samples at their lab. Thanks to River Basin Centre and UGA Office of Sustainability for funding this research.

Thanks to my sisters, Meenu and Anu, grandparents, roommate Divya and all my other friends and family for keeping me positive and helping me move forward through all the dull moments.

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CHAPTER 1

INTRODUCTION AND LITERATURE REVIEW

Lake Herrick is a recreational water body within the campus of University of Georgia. The watershed is a small urban catchment (131 ha) with 72% developed (NLCD Classification) land use. After 20 years, the lake was closed for recreation around 2002 which prohibited swimming and boating but allowed fishing.

Pollution of surface waters and eutrophication is a world-wide problem and Lake Herrick is no exception. Currently many classes use the lake for research projects and experiential learning. The trails and dog park at the Oconee Forest and the Intramural Fields adjacent to the lake are widely used by students. An Ad Hoc Lake Herrick watershed restoration committee was formed in February 2016 to help prioritize the management and improvements of the lake. The report by the Ad Hoc Committee has details of the other subcommittee and management efforts that were planned during the spring of 2016 (Report, 2016).

Phase 1 of the restoration project completed during the summer of 2018 removed accumulated sediments at the Oconee Forest pond. Invasive plants near the pond were removed and replaced by natural flora and new stormwater control measures were installed. The lake was reopened for recreation in October 2018 with the old beach area being renovated to a pavilion area and launch deck for canoes and kayaks. Future work includes the installation of stormwater control measures at the Rogers Road Housing Complex, the Rec Sports Complex and the two inlet tributaries of the lake. The water quality data collected in this study will provide a better understanding of the source and transport of contaminants in the watershed. It could help in the

modeling efforts of green infrastructure within the watershed, which is one of the proposed future goals.

Impact of Water Quality in Watersheds

Urban land uses within a watershed directly impair the quality of water and aquatic life (Ann Cunningham et al., 2010; Grimm et al., 2000; Paul et al., 2001). GAEPD has standards for recreational waters with daily average DO of 6.00 mg/L and pH within a range of 6.00 to 8.50, temperature not exceeding 90° F. Lake Herrick meets all the recreational water quality criteria for DO, pH and temperature. Turbidity is the clarity of water and it is an indication of the suspended sediments in the water. High turbidity levels cause problem for the macroinvertebrate communities, suppression of light and alteration of BOD levels. Turbidity generally follows the first flush in discharge during stormflows (Lawler et al., 2006).

Higher conductivity and pH are an indication of pollution and it is directly associated with the land uses in the watershed (Conway, 2007; Kaushal et al., 2005). Some studies have found that land use had little effect on pollutant loads (Wickham James et al., 2002). On the other hand, a study in a southern Ontario watershed of Canada found that catchment land use had greater influence on water quality than riparian buffers (Sliva et al., 2001). Hence, awareness by the watershed residents and stakeholders along with the maintenance of buffers could filter nutrients and other pollutants before they reach the stream.

The other two contaminants of concern are algae and nutrients. Excessive nutrient input into the stream causes over production of organic matter. Not all algae are harmful, but some cyanobacterium can cause blooms that produce toxins called *Microcystis* (Paerl et al., 2016). Bacteria are naturally present in the environment and total coliform is a measure of all the bacteria in the environment. Fecal Coliform is very specific to fecal *bacteria* and it is used as an indicator

of water quality. *E. coli* is a sub group within fecal coliform and it is used as a indicator because of its linear association to GI associated illness. These contaminants are further discussed in the upcoming sections.

Nutrient Impairment

Anthropogenic influences in the ecosystem increases the nitrogen and phosphorus inputs into the streams that raises the concern of eutrophication. Eutrophication interferes with the aesthetics, recreation and causes discoloration, pH changes, low dissolved oxygen, excessive growth of suspended, filamentous, and benthic algae. Shift in the trophic state of the water body and dominance of a single species occurs due availability of nutrient in excess. There have been three occurrences of blue-green algal blooms of the genus *Microcystis* in the summer of 2002 and 2005 and winter of 2015 recorded at Lake Herrick (Christopher, 2015). Previous research and studies have suggested phosphorus, and to a lesser extent, nitrogen entering the lake from the tributaries during baseflow and during storms are potential triggers for the undesirable blooms.

Walker (1987) reported that “cause-effect relationships linking urban development to lake and reservoir eutrophication are well established,” and that “urban watersheds typically export 5 to 20 times as much phosphorus per unit per year, as compared to undeveloped watersheds in each given region.” In shallow lakes like Lake Herrick it is important to reduce and manage the external nutrient loading as well as the internal nutrient loading.

Total phosphorus (P) consists of dissolved and particulate forms, and the dissolved form can be inorganic and organic. Dissolved inorganic P is the most biologically available phosphorus in the system, but dissolved organic P is also important. In general, total dissolved P concentrations greater than about 10-50 µg/L have been found to trigger algal blooms (McDowell et al., 2004). Particulate P is important in that it represents additions to lake bed sediment P.

Various studies have shown that particulate P is one of the dominant forms in runoff and it usually accumulates on the finer sediments of 1-25 microns and is washed off during runoff or irrigation events (Bennett et al., 2001; Vaze et al., 2002). Higher total P concentrations during the storm events are also attributed to the particulate P being washed from impervious surfaces.

The biological availability of nitrogen (N) and P varies with respect to their chemical composition, but the ratio of N:P can be used as a proxy to draw inferences on nutrient limitations in the system. Historically it has been thought that most lakes are P limited and oceans are N limited. Streams behave like lakes except for the differences in residence time and stratification. Research studies regarding eutrophication in streams (Dodds et al., 2016) have found total N and P were highly correlated to benthic algae biomass with nutrients jointly explaining more variations.

Most water bodies should be limited by one or the other nutrient, and the phase where it shifts from N to P limitation or vice versa is where production is most likely to occur (Sterner, 2008). Hence, for the management strategies judicious control of N and P is required.

Gardening activities, washing of hardscapes, plant debris, ammonia fertilizer, and compost are reported as sources of N during urban run (Groffman et al., 2004; McPherson Timothy et al., 2007). A study found that human activity altered the stream nitrate (NO_3^-) concentrations in both agricultural and residential catchments to the same magnitude. They also showed higher concentration pattern during summer and spring and dilution during fall and winter (Poor et al., 2007). Hence there is a need to identify the patterns and the dominant form of N in baseflow and stormflow. Nitrogen forms consist of ammonium, nitrate, and organic N.

Fecal Coliform and E. Coli

Total coliform bacteria are widespread in nature and are found in manure, soil, water, submerged wood, animal, and human feces. The most commonly used indicator bacteria for fecal

contamination are Total Coliform, Fecal Coliform, *E. coli*, Streptococci and Enterococci. Fecal Coliform and *E. coli* are subgroups within the total coliform group. These groups indicate possible contamination by sewage.

Fecal coliforms are known to contain genes that have both fecal and non-fecal origin. There are studies that show they are poor indicators of gastrointestinal diseases caused by recreating in water. Various studies at EPA show that *E. coli* and Enterococci are most correlated with gastrointestinal diseases and the agency recommends using them as indicators for cautioning beach users (Adopt-A-Stream). While Enterococci are more generally recommended for salt water, *E. coli* is a better indicator for fresh water and it is one of the prominent groups of fecal coliform bacteria.

The US EPA recommends a conversion factor of 0.63 for *E. coli* to Fecal Coliform bacteria. The US EPA recommends a limit of 126 MPN/ 100 ml geometric mean or STV (Statistical Threshold Value) of 410 MPN/100 mL over a period of 30 days for *E. coli*. The STV was recommend as a new standard in 2012 for recreation water quality criteria. The STV should not exceed 410 MPN/ 100 mL in more than 10% of the samples collected. This is associated with an estimated illness rate of 30 persons per 1000 who come in primary contact with recreation water (USEPA, 2012).

Source Tracking

Testing of waters for every possible disease-causing microorganism is difficult, especially for pathogens. Fecal Indicator Bacteria that are easy to detect and have their origin from the same source as the disease-causing pathogens are used as indicators. These indicator bacteria live in the gastrointestinal tract of warm-blooded animals and once detected in water samples their source is not identified using regular culture-based methods. Microbial Source Tracking (MST) is a

technique to identify genes associated with the target. Several qPCR assays have been developed for host specific source-tracking markers and this makes the entire approach quick.

The qPCR method is rapid, and the source could be identified within 4 to 6 hours of sample collection. Also, non-human contamination poses a lower level of risks for recreational waters. Essentially, the best management practice would differ substantially if the source was identified to be wildlife rather than human (J. A. Soller et al., 2010).

Principal Component Analysis (PCA)

Principal Component Analysis (PCA) is normally used to identify the relationship between different variables using the linear association matrix. This type of analysis is used when there are different explanatory variables related to one another. Since it shows the association between the variables it reduces the dimensionality of the dataset. Eigen vectors and eigen values are extracted from the covariance matrix obtained from the original variables. Identifying the principal component of the database that maximizes the variance is another aspect of this analysis.

The principal components are obtained by multiplying the uncorrelated variables with the eigen vectors, and the resulting variable is an uncorrelated (orthogonal) variables (Filik Iscen et al., 2008). The eigen values of principal components are a measure of their variation. In this study PCA was performed on in-situ parameters, *E. coli*, forms of N and P, conductivity, and turbidity to understand the seasonal pattern and linear association of variables in the dataset for all three sites (Singh et al., 2005).

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2. TRACKING AND MANAGING NON-POINT SOURCE POLLUTION IN THE
LAKE HERRICK WATERSHED, ATHENS, GEORGIA ¹

¹ Kannan, Ashwini, D.E. Radcliffe, T.A.P. Saintil, T. Rasmussen, M. Molina To be Submitted to the Journal of Environmental Quality.

CHAPTER 2

**TRACKING AND MANAGING NON-POINT SOURCE POLLUTION IN THE
LAKE HERRICK WATERSHED, ATHENS, GEORGIA**

ABSTRACT

The Lake Allyn M. Herrick watershed is about 131 ha and covers portions of the University of Georgia's East campus, the Oconee Forest, residential and commercial land use. Lake Herrick, a 6 ha water body established in 1982 at the University of Georgia campus was closed in 2002 for recreation due to fecal contamination. Subsequent monitoring confirmed cyanobacterium blooms on the surface of the lake and elevated nutrient concentrations, especially phosphorus. Previous studies showed that phosphorus and fecal coliform were the main contaminants. Two inflow tributaries (Birdsong and Armadillo) and the outlet stream (Below Dam) were monitored for discharge, *E. coli*, forms of nitrogen and phosphorus and other water quality parameters during baseflow and storm conditions from February 2016 to October 2017. Our results showed that total phosphorus was significantly higher during stormflow compared to baseflow, but total nitrogen remained the same. *E. coli* results indicated that most of the bacteria entered the lake through the tributaries during stormflow. Microbial source tracking methods were used to detect the bacterial source in the samples specific to a dog, ruminant or human host. We found that dogs were a more likely source of this bacteria than humans or deer. The fact that human sources were uncommon in the Lake Herrick watershed indicated that there was reduced risk for human source contamination. Lake Herrick was reopened for limited recreation in October 2018.

INTRODUCTION

Background

Change of land use from forest and agriculture to suburban and urban has been progressing at a rapid rate with a majority of the population residing in the urban areas (O'Driscoll et al., 2010). Urbanization is accompanied by an increase of impervious surface and affects the quantity and quality of water both above and below the ground. Anthropogenic influences have led to compromised water quality due to the addition of large quantities of pollutants otherwise not found in the natural environment.

Point source pollution includes industrial and municipal effluent which can be tracked under with the Clean Water Act (CWA) permits for point sources. However, non-point source pollution depends on land use and is difficult to track because of the variability of the pollutants and its diffuse nature. Hence, it is important to understand the role of different land uses and how they affect water quality of aquatic systems. Higher concentrations of pollutants are typically associated with the rising limb of hydrographs during storm events (Chang et al., 2005), but change in the forms of the pollutants cannot be explained by the first flush phenomenon because of the dynamics of the chemistry (Old et al., 2003).

The national water quality inventory report to Congress stated that 55% of the assessed rivers and streams were impaired and did not support one or more designated uses. The top causes of pollution were pathogens, sediments, and nutrients. The report also states that 70% of lakes, ponds, and reservoirs were impaired by mercury, nutrients, and Polychlorinated Biphenyl's (PCB's). About 40% of the water bodies were impaired due to phosphorus and 39% had the algal toxin *Microcystin* detected in them (USEPA, August 2017).

Urban catchments respond faster than other landuse to a storm due to higher imperviousness. The hydrographs are flashy, with the first flush usually washing more pollutants from the watershed. Watershed size is also important because it interacts with the land use and the effects of climate and weather. There is a hydrologic contrast in how the pollutants affect the water bodies like rivers, dams, lakes, and other impoundments. Rivers have lower residence time and they may flush the sediments as they enter the system. This process plays an important role in the suspension and resuspension of particulate phosphorus. Water bodies with long water residence time can aggregate sediments. The sediments can act as both a source and sink. Hence, careful monitoring is needed to estimate the pollutant loads and take relevant actions (Aalst et al., 2010).

Motivation

Lake Allyn M. Herrick is an urban shallow lake on the University of Georgia (UGA) campus. It spans about 6 ha, with 167 ML water and an average depth of 2.74 m. A small pond, Oconee Forest Pond, is in the Oconee Forest Park and feeds Lake Herrick via a tributary stream called Birdsong (BS) Creek.

Another inlet stream called Armadillo (AR) collects storm water from the Five Points residential neighborhood. The outflow from the reservoir, beginning at the Below Dam (BD) site, joins the North Oconee River (Figure 1), which is listed in the impaired stream list for violating fecal coliform standards. Lake Herrick's watershed spans about 131 ha, delineated using ArcGIS hydrology tools with the outlet point at the BD site. According to the NLCD 2011 class, 72% of the watershed is developed, 23% is undeveloped and 5% is water.

In 2002, Lake Herrick Beach, which was popular with students, was closed for recreation (swimming and fishing) due to high concentrations of fecal coliform (Christopher, 2015). Several research projects suggested that the impairment was mainly due to urban runoff and the tributaries

that washed from the sub-catchments into the lake (UGA, 2017). There is evidence of deer, dog, and Canada geese populations within the watershed. The Athens community uses the trails in the Oconee Forest Park to walk their dogs. Deer also leave droppings along streambeds increasing fecal coliform counts in the water. Most of the sewer lines in the watershed are within the Five Point neighborhood and could introduce human pathogens. In fact, one sewer line directly crosses one of the tributaries at the point of entry to the Lake. However, the Athens-Clarke County (ACC) public utilities department has not found any leaks from previous inspections. Storm water drainage pipes discharge high volumes of untreated runoff into the lake and might account for some of the non-point sources of pollution.

The US EPA regulatory limits for fecal indicator bacteria *E. coli* are 126 MPN/ 100 mL for a geometric mean of four samples collected at least a week apart and 235 MPN/100 mL for a one-time sample. Recently, the Georgia Department of Natural Resources proposed adoption of these standards (personal communication, Elizabeth Booth, Georgia Environmental Protection Division).

Sediment washed into the lake could both act as both a source and sink of pollutants, hence careful monitoring is needed to estimate the pollutant loads (Aalst et al., 2010). The lake originally had a depth of around 8 meters near the dam when it was established and later the level had diminished to 5.5 meters when surveyed by Krauss and his team in 1999 (Christopher, 2015).

There have been several blue-green algal blooms of the genus *Microcystis* in the summer of 2002 and 2005 and winter of 2015 recorded at Lake Herrick (Christopher, 2015). Phosphorus, and to a lesser extent, nitrogen entering the lake from the tributaries during baseflow and during storms are potential triggers for the undesirable blooms. The lake bed sediments may be another source of phosphorus.

In a report, Walker (1987) stated that "cause-effect relationships linking urban development to lake and reservoir eutrophication are well established", and that "urban watersheds typically export 5 to 20 times as much phosphorus per unit per year, as compared to undeveloped watersheds in each given region". In shallow lakes like Lake Herrick, it is important to reduce and manage the external nutrient loading as well as the internal nutrient loading.

Total P consists of dissolved and particulate forms, and the dissolved form can be inorganic and organic. Dissolved inorganic P is the most biologically available form of P, but dissolved organic P is also important. In general, total dissolved P concentrations greater than about 0.01 - 0.05 mg/L have been found to trigger algal blooms (McDowell et al., 2004). Particulate P is important in that it represents additions to lake bed sediment P. Nitrogen also contributes to eutrophication and forms consist of ammonium, nitrate, and organic N.

Brown and Caldwell (B&C) is an environmental consulting company that was hired by UGA to run a series of water quality tests on the streams which run through campus starting in 2006. For Lake Herrick, B&C sampled at two strategically placed locations MS4-4b, located at the outflow of the lake and MS4-4c, located closer to the Oconee Forest Pond outlet (Figure 1). MS4-4a, a little downstream of the forest pond, was removed as a sampling location in 2007 due to the creation of a beaver dam wetland, and instead MS4-4c was sampled. There were 8 sampling events each year: 4 dry and 4 wet events to determine the effects of storm and baseflow. B&C analyzed the samples for total suspended solids (TSS), fecal coliform, *E. coli*, volatile organic compounds (VOCs), total phosphorus, total nitrogen, metals (As, Cu, Pb, and Zn), oil and grease, mercury, pH, dissolved oxygen, temperature, and conductivity. The monitoring program is ongoing. After 2015, B&C added two new stations: MP21 on Armadillo Creek and MP20 on Birdsong Creek downstream of the Oconee forest pond.

The objectives of our research were:

1) To compare the *E. coli* concentrations during baseflow and stormflow and the seasonal trends of *E. coli* concentration to aid in source identification. To identify bacteria sources using genetic and culturable *E. coli*, human-associated Bacteroides markers HF183-MGB, ruminant-associated Rum-2-bac, and dog-associated Dogbac.

2) To determine the dynamics of nutrients at inlet tributaries and the outlet. To estimate the sources of various forms of phosphorus and nitrogen concentrations in the inflow and outflow and draw cause-effect relationship for the algal blooms.

MATERIALS AND METHODS

Landuse and Description

Lake Herrick's watershed is located on the southern campus of the University of Georgia, Athens. It is situated along the North Oconee River in Clarke County, in the Piedmont region of northeast Georgia. The 131-ha watershed is part of the North Oconee river basin (32,100 ha) that originates in the highly forested Blue Ridge Mountain region (Meyer et al., 2017). Athens receives an annual rainfall of 1.22 m which can fluctuate by 40% due to seasonal variations (Rasmussen, 2016). The annual rainfall for the study period of 2016 and 2017 was 1.01 and 1.15 m respectively (UGA, 2017).

Runoff in the Piedmont region is high in sediment due to the leaching of older, erodible soil and over development. However, the rivers are green because of the algae and plants that float in the water (Rasmussen, 2016). The study area lies in the Piedmont and falls under nutrient levels (iii) Ecoregion 45, and the 25th percentile values for TN, TP and Turbidity are 0.615 mg/L , 0.030 mg/L and 5.7 NTU respectively (USEPA, 2000).

A major portion of the watershed area is covered by Urban Residential Low, Medium, High and Industrial Density Units (URLD, URLD, URHD and UIDU), which constitute 72% of the watershed according to the NLCD 2011 classification. The watershed boundary and the three sampling locations are shown in Figure 1. There are 831 units in the urban areas, according to the address point shape file obtained from Athens Clarke County. These units consist of apartments, condos, duplexes, residential, non residential and parcel addresses. The UIDU and URHD classes constitute about 6% of the watershed area and include the high density parking decks in the southern part of watershed close to the lake. The low and medium density areas are located between South Milledge Ave and East Campus Road with Graduate and Family Housing units near East Campus Road and the residential neighborhood adjacent to South Milledge Ave. The Intramural Fields, railway tracks, baseball and tennis courts are known impervious surfaces located close to Birdsong Creek and the lake.

The rest of the watershed is the Oconee Forest Park/Natural Area and it accounts for about 23% of the area, with mixed successional vegetation such as oak, hickory, tulip poplar, beech, and pine. Understory plants include dogwood, assorted native shrubs, a herbaceous layer, and a leaf litter floor (Morphis et al., 2014). Oconee Forest Pond (0.52 ha) is in the Oconee Forest Park upstream of Lake Herrick on Birdsong Creek. The remaining 5% of the watershed is covered by water, which consists for the most part (about 6.31 ha) of lake surface area. There are only two septic systems within the catchment in the Five Points neighbourhood and four along the watershed boundary parallel to the S. Milledge Avenue (Figure 1).

The total impervious surface area in the watershed is approximately 21.5%, calculated using the address point shapefile from the Athens Clarke County using GIS. This included all parking deck, alleys, driveways, mobile homes, paved roads, unpaved roads, parking areas, patios,

public and private sidewalks, sheds, garages, pools, unpaved driveways, unpaved parking lots and unpaved roads. However, impervious surface is total impervious area (TIA) rather than the directly connected impervious area (DCIA), which may be a better predictor of stream ecosystem alteration but is more difficult to determine. Although there are empirical relationships to determine the DCIA from TIA, on site assessments are necessary to determine the connected impervious surface (Roy et al., 2009). Stream health is characterized as ‘protected’ if the impervious surface is less than 10% of the total watershed area, ‘impacted’ for 10-30% and ‘degraded’ for over 30% (Arnold et al., 1996). With regards to the ownership, 63% of the watershed is owned by UGA (UGA Ground Department GIS data).

History

Lake Herrick was impounded in June 1982, but the land was cleared of trees by 1938. The history of Lake Herrick is contained in two reports (Christopher, 2015; Morphis et al., 2014). Lake Herrick, Oconee Forest Park and the Intramural Fields are managed by the UGA School of Forestry and Natural Resources and the UGA Recreation Sports and Facilities Division. After two years, the lake was opened for fishing in 1984. Filamentous algae were recognized in the lake in 1985 and 1986. To reduce algal growth, the university decided to treat the lake with 91 kg of copper sulphate crystals the first year, followed by 16 kg the next year.

A report drafted by UGA law students discussed the possible pathways through which contaminants could enter the lake. Runoff could carry fertilizer, insecticide and fungicide from the intramural fields maintained by the university ground crews. Birdsong Creek used to convey runoff from a pond on the far side of the bypass and probably from the bypass too. Armadillo Creek conveyed runoff from the residential area. The depth of the lake near the foot bridge was reduced

by 0.15 m over a period of 12 years from 1985. The height at the dam was reduced by 2.54 m over 17 years from the time it was built because of sedimentation and erosion events in the past.

Discolorations were observed in July 2002 due to suspended matter, and Armadillo Creek was thought to be the reason for the discoloration. Professor Marshall Darley sampled phytoplankton in the lake during the same period and observed high cyanobacterial colonies. They belonged to the genus *Microcystis*, known to produce toxins unsafe for human and animal consumption. They had been appearing in the lake for at least a year, but by late August the bloom hit a peak. Lake users were prohibited from recreating and swimming sometime between 2002 and 2005 (Morphis et al., 2014).

Lake Herrick's watershed restoration is a multi-phased project aimed at restoring the lake to promote research, learning and recreational activities. The first phase of the project began in the fall of 2017 and was completed in the summer of 2018. Phase I of the project was designed to rehabilitate the Oconee Forest pond through the removal of sediments accumulated over the past 50 years and the replacement of invasive plants with native flora. Phase II aimed at shoreline improvements in the beach area, including a dock for launching boats. Future work will include the implementation of storm water control measures within the watershed, a lake-side walking and running trail, and planting of native riparian vegetation (UGA, 2017). It has been common for pet owners to bring their dogs to Oconee Forest Park where there are trails and a fenced dog park upstream of the pond. The dog park was closed in summer of 2017, dogs were required to be on leash, and signs were erected along the trails encouraging dog owners to pick up their pet's waste using bags provided at the sign (Linthicum, 2017). The area remains popular with dog owners.

Sampling Sites

Based on the recommendations from previous studies at Lake Herrick, three monitoring locations were chosen: one at each inlet tributary (Birdsong: BS and Armadillo: AR) and the third one just below the dam (BD) (Figure 1). Samples were collected using automated ISCO samplers installed at each location. Grab samples were also collected twice per month for less intensive sampling and four times per month for intensive sampling months under baseflow conditions. The automated ISCO samplers were triggered when stage increased for events greater than 0.25 cm. Each sampling station responded differently to storm events, hence the stage for each site was set individually.

Sample Collection

The inlet and outlet streams were monitored from February 2016 until November 2017 for stage, *E. coli*, conductivity, turbidity, nutrients, temperature, pH, specific conductance and DO. The samples were analyzed for microbial source tracking from January 2017 until October 2017. A Quanta multimeter probe was used to measure in-situ parameters of specific conductance, pH, temperature, and dissolved oxygen (DO) during baseflow. The samples were divided into duplicates. One set of samples were analyzed for *E. coli*, turbidity, conductivity, and nutrients. The other set of samples were transported on ice to the EPA Office of Research and Development (ORD) laboratory facilities in Athens, GA.

Additional water level sensors were placed at all the sites close to the ISCO transducers to measure the water level in the stream. The ISCO samplers were powered by solar panel and the additional water level sensor data were used as back up in case of ISCO power failures. The ISCO automated storm sampler was calibrated to take fourteen 950-mL samples every 30 minutes and then the samples were composited by event in the lab. Samples were collected over a period of seven hours and they were removed from the refrigerated sampler within four hours.

Stream water level at the Armadillo site was monitored with an Odyssey water level logger placed at the side of a rectangular weir at the entrance to a highway culvert before the creek reaches the lake. The discharge measurements were taken on the downstream side of the tunnel and stage values (10-minute intervals) data were extrapolated for daily and sub daily discharge. A 0.46-m H-flume was installed at the Birdsong site and a Druck water level sensor was placed at the mouth of the flume. An Odyssey water level sensor was placed at the bridge in the lake to measure water level fluctuations and this was used as proxy for stream stage at the outlet of the dam.

UGA Isotope laboratory

The nutrient analysis was done at the UGA Isotope Laboratory. The samples were processed after collecting the samples from ISCO automated samplers. Samples were kept frozen until analysis. The sample preparation was performed at the UGA Soil Physics Laboratory. The Isotope lab used a cadmium reduction method for nitrate analysis recommended by the United States Department of Environmental Protection Agency (U.S. EPA). A gas diffusion-conductivity technique using TL-2800 Ammonia/Nitrate Analyzer system was used.

Analytes

A total of 103 samples from February 2016 to July 2017 were analyzed for all seven analytes. The samples were analyzed for dissolved inorganic phosphorus (DIP), total dissolved phosphorus (TDP) and total phosphorus (TP). We calculated dissolved organic phosphorus (DOP), total particulate phosphorus (TPP) as follows:

$$\text{DOP} = \text{TDP} - \text{DIP}$$

$$\text{TPP} = \text{TP} - \text{DIP} - \text{DOP}$$

$$\text{TDP} = \text{DIP} + \text{DOP}$$

The samples were also analyzed for ammonia nitrogen ($\text{NH}_4^+\text{-N}$), nitrate nitrogen ($\text{NO}_3^-\text{-N}$) total dissolved N (TDN), and total nitrogen (TN). Dissolved organic nitrogen (DON) and total particulate nitrogen (TPN) were calculated as follows from the measured fractions:

$$\text{TPN} = \text{TN} - \text{TDN}$$

$$\text{DON} = \text{TDN} - \text{NH}_4^+\text{-N} - \text{NO}_3^-\text{-N}$$

E. coli

We used the EPA standard and proposed Georgia standard for *E. coli* of a single sample value of 235 MPN/100 mL as a standard of water quality in our study. The 235 MPN/100 mL value corresponds to 36 out of 1000 people getting sick who come in contact with the water. In this study the *E. coli* concentrations were determined using the IDEXX Colliert-18 kits at the UGA Soil Physics Laboratory within four hours after sample collection following the standard method (APHA, 1998). The concentration of *E. coli* is reported in MPN/100 mL. Duplicate analysis was performed on samples with 10% dilution for baseflow samples and 100% for storm samples since the counts were usually higher during storms.

Source Tracking

For the source tracking analysis, we collected a total of 30 baseflow and 31 storm samples from January 2017 to October 2017 to be analyzed for microbial source tracking. The baseflow samples were collected in a synoptic manner with weekly intensive sampling schemes. The storm samples were collected using an ISCO- automated sampler at 30-min intervals as described above. The samples were taken to the EPA ORD Laboratory for further analysis. One hundred mL of water were filtered using 0.47- μm Iso-pore polycarbonate filters to concentrate *E. coli* cells for DNA extraction using MoBio Power-Soil DNA Isolation Kits (MoBio Laboratories, Carlsbad, CA).

Filters were stored at -80 degrees Celsius until extraction. The DNA from the filters were extracted through Mobio Power Lyzer Power Soil DNA extraction kits.

After the extraction, we ran a quantitative polymerase chain reaction (qPCR) using sets of standards depending on the assay. All the qPCR analysis was conducted using Quant Studio 3 Real-Time PCR System (Thermo Fisher Scientific, Waltham, MA). All the methods for this procedure followed rigorous EPA guidelines including Quality Assurance (QA) and Quality Control (QC) practices. All the reactions were performed in duplicate and the third set was saved as a backup. All reactions with duplicate samples were run for 40 cycles on a 96-well plate with eight triplicate standards, 2 controls, and the extracted samples. Several reagents were used including Fast TaqMan (Taq), primers and probes (PP), and bovine serum albumin (BSA) to prepare the master mix that was used as the standard. All quantification methods were followed as described (Oladeinde et al., 2014) in the study. Markers were used for detection of dog (Sinigalliano et al., 2010), human (Haugland et al., 2010), ruminant (Mieszkin et al., 2010) and genetic *E. coli* (Chern et al., 2011).

The threshold cycle (C_T) was exported from the Quant Studio 3 software to an Excel sheet. Four values were obtained in TSC (Target Sequence Copies)/4 μ L or Gene Copies (GC)/4 μ L for each sample set and the samples which were 1.5 times greater than the mean C_T were flagged to re-run. The values were converted and averaged to TSC/100 mL using the formula sheet provided at the EPA. Concentrations of MST markers were transformed to base-10 logarithm. Markers were 'undetected' if no C_T values were reported within 40 cycles and were given a log transformed value near 0. The Limit of Quantification (LOQ) was 10 TSC/4 μ L and any sample quantity value below 10 TSC/4 μ L was assigned a log transformed value of 0.7.

Principal Component Analysis of Water Quality Variables

Principal Component Analysis (PCA) was performed separately on the baseflow and stormflow data for the in-situ parameters, *E. coli*, forms of N and P, conductivity, and turbidity to understand the seasonal patterns, differences between baseflow and stormflow and correlation between variables in the dataset for all three sites (Singh et al., 2005).

Residence Time

Average hydraulic residence time is a measure of the time water spends in a lake from the time it enters the lake to the time it exits. Longer residence time can lead to significant effects on the water quality and more interaction with the bottom sediments. The average residence time for Lake Herrick was calculated for baseflow assuming uniform mixing and stratification using the equation shown below:

$$\begin{aligned}\text{Average Residence Time} &= \text{Average Volume of Lake} / \text{Average Baseflow Rate} \\ &= (60000 \text{ m}^2 * 5.5 \text{ m}) / (0.01 \text{ m}^3/\text{s}) \\ &= 381 \text{ days}\end{aligned}$$

During high flows in storms with flow rates such as 1 m³/s, the residence time was about four days. Hence, the pollutants were flushed out of the lake faster during periods of high flow.

Statistics

One-way ANOVA was run to find differences in *E. coli* concentrations between sites for base and stormflow separately. If there was a significant difference between the sites, a post-hoc Tukey test was run to compare the groups. Also, a one sample t-test was run at each site during baseflow and stormflow to test the difference in the magnitude of bacteria concentration during periods of rainfall.

RESULTS AND DISCUSSIONS

Forms of Nitrogen and Phosphorus

The biological availability of N and P varies with respect to their chemical composition, but the atomic mass ratio of N:P can be used as a proxy to draw inferences on the nutrient limitation in the system. A study by Guildford et al. (2000) plotted the ratios for 221 lakes in 14 countries and showed that a N deficiency was apparent at $N:P < 9$, P deficiency at $N:P > 22$ and both were co-limited in the intermediate range between 9 and 22 (Guildford et al., 2000). Also, a study with Canadian lakes found that highest cyanobacterial biomass occurred at the intermediate range.

We calculated the N:P ratio at the three sites in our study using the TN and TP data. The N:P ratio was plotted against TP (Figure 2a) and TN (Figure 2b). The TP did not vary between sites and the N:P ratio decreased as TP increased. N-limited samples at all the three sites combined were less than 10% of the total samples and none of the baseflow samples were N-limited. Nearly 26% of the samples were co-limited at Birdsong and Below Dam sites and 49% were co-limited at the Armadillo site. Most of the co-limited samples were stormflows. More than 50% of the combined samples were P-limited and most were baseflow samples.

This suggests that the input and output sites behave differently in terms of the N:P ratio due to different retention times and other factors. Mostly, the lake was P-limited under baseflow conditions. With stormflows bringing in runoff from the catchment, the lake shifted from P-limited to either co-limited or N-limited with higher loading of TP. Also, it is evident from Figure 2b that the TN concentration did not vary much when plotted against the N:P ratios for stormflow and baseflow samples. This shows that effective control of P transport could combat the problem of blooms in the lake.

The average flow value at the Birdsong site was $0.01 \text{ m}^3/\text{s}$ (Figure 3a). The flow after April 1st (the blue line in Figure 3a.) was noisier because the sensor shifted after a storm event on 5

April 2017. Independent t-tests showed that the TP concentrations for storm samples were significantly higher than the baseflow sample concentrations. TN concentrations were remarkably similar between baseflow and stormflow samples and t-tests showed there was no significant difference between samples. A study of four headwater streams in Gwinnett County (Hoghooghi, 2016) found similar results.

A few TP values greater than 200 $\mu\text{g/L}$ were recorded in May 2017 with 263 $\mu\text{g/L}$ and twice in June 2017 with 213 $\mu\text{g/L}$ and 308 $\mu\text{g/L}$ at the Birdsong site. All were stormflow samples. The rest of the samples were usually below 100 $\mu\text{g/L}$. Almost all the TN samples were below 1.50 mg/L. Two exceptions were a baseflow sample in July 2017 with 1.53 mg/L and a stormflow sample in August 2016 with 1.93 mg/L. Since there are no standards for nutrients in Georgia, we used the nutrient criterion recommended by EPA assuming the 25th percentile of sampled reference streams in the region represented minimally impacted streams. Athens comes under the Nutrient Ecoregion IX, level III Ecoregion 45 (USEPA, 2000). A total of 54% of the samples exceed the ecoregion criteria of 0.03 mg/L for TP and 100% of the samples exceed the value of 0.62 mg/L for TN at the Birdsong site.

The average flow rate at the Armadillo site was 0.01 m^3/s , similar to the Birdsong value. Also like the Birdsong site, t-tests showed that TP concentrations were significantly higher under stormflow conditions compared to baseflow conditions but there were no differences between stormflow and baseflow TN concentrations. Most of the samples for TP were less than 150 $\mu\text{g/L}$ with one high value recorded at 215 $\mu\text{g/L}$ (Figure 3b). Most of the TN samples were below 2.00 mg/L except three values, one during baseflow at 2.15 mg/L and two others during stormflow with 2.24 and 2.54 mg/L. A total of 71% of the samples exceeded the ecoregion criteria of 0.03 mg/L for TP and 97% of the samples exceeded the value of 0.62 mg/L for TN at the Armadillo site.

With respect to the Below Dam site, the flow was relatively constant with an average value of 0.006 m³/s (Figure 3c). Independent t-tests showed that both the TP and TN values during stormflow were significantly higher than the baseflow values. The TP samples were below 0.10 mg/L aside from two stormflow samples with concentrations of 0.130 mg/L and 0.108 mg/L. The TN values were less than 1.5 mg/L except for two values at 1.62 and 1.72 mg/L. A total of 55% of the samples exceed the ecoregion criteria of 0.03 mg/L for TP and 93% of the samples exceed the value of 0.62 mg/L for TN at the Birdsong site.

One-way ANOVA showed that the P concentrations in different forms (DIP, DOP and TPP) were not significantly different between sites (Figures 4a, 4c, and 4e), but N forms did differ between sites (Figures 4b, 4d, and 4f). ANOVA tests also revealed that the NO₃⁻ concentrations were significantly higher at Armadillo than Below Dam under both stormflow and baseflow conditions. The NO₃⁻ concentrations at Birdsong were significantly higher than Below Dam during baseflow conditions. Ammonium (NH₄⁺) was significantly higher at Below Dam than the other two sites under both flow conditions. Total particulate N (TPN) concentrations were significantly higher during baseflow at Armadillo and Birdsong, compared to Below Dam. These results imply processing of N forms within the lake, but not P forms.

The average P fractions of DIP, DOP and TPP at Birdsong during baseflow were about 8%, 35% and 57% respectively (Figure 4a). The average fractions during stormflow for DIP, DOP and TPP were about 6%, 26% and 68% respectively. A t-test showed that TP concentrations at Birdsong were significantly higher during stormflows than baseflow. Also, the organic fraction (DOP) was significantly higher during stormflow than baseflow. These results showed the importance of organic fractions in P loading to the lake. Also, the three high values recorded during May and June 2017 (263, 213 and 308 µg/L) probably indicate sediments washed from the Upper

Pond renovation work. Also, renovation work at the Red Coats band practice field happened during the same period, which drains via a storm drain to the Birdsong Creek close to the sampling location. A higher fraction of PP in these three samples also implied the source of the higher concentration was runoff with sediments.

The average N fractions during baseflow were 5% NH_4^+ , 68% NO_3^- , 9% TPN and 17% DON (Figure 4b). The TN average concentration did not change significantly during stormflows (as noted earlier) because NO_3^- concentrations, which originate in groundwater, decreased by half and TPN and DON, which originate in runoff, doubled: 4% NH_4^+ , 38% NO_3^- , 21% TPN and 35% DON. These changes were statistically significant.

The average P fractions for DIP, DOP and TPP at Armadillo during baseflow were about 12%, 31% and 57% respectively (Figure 4c), like the Birdsong results. During stormflow the DIP, DOP and TPP fractions were about 10%, 30% and 60% respectively (also like Birdsong). Likewise, TP concentrations were significantly higher at Armadillo during stormflow compared to baseflow. Also, the DOP and TPP fractions were significantly higher during stormflow than baseflow.

At Armadillo, the N fractions during baseflow were 2% NH_4^+ , 69% NO_3^- , 16% TPN and 13% DON (Figure 4d). During stormflow, the fractions were 2% NH_4^+ , 45% NO_3^- , 23% TPN and 29% DON. The t test showed the NO_3^- fraction decreased significantly during stormflow and the organic fractions (TON) increased.

At the Below Dam site, organic and particulate P were still the largest fractions with baseflow P consisting of 8% DIP, 40% DOP and 52% TPP. Stormflow fractions were 14% DIP, 38% DOP and 47% TPP. Also, TP concentrations were significantly higher during stormflow than

baseflow and DOP was significantly higher during stormflow than baseflow. These results showed little change occurred in the P fractions between the input streams and the outflow from the lake.

The N fractions at Below Dam during baseflow were 15% NH_4^+ , 29% NO_3^- , 21% TPN and 35% DON (Figure 4f). During stormflow the fractions were similar with 15% NH_4^+ , 19% NO_3^- , 26% TPN and 40% DON. The main differences between the Below Dam sites and the tributaries was the increase in NH_4^+ (perhaps due to mineralization) and decrease in NO_3^- (perhaps due to denitrification). In contrast to the P results, this indicated N transformation within the lake.

Comparison to Brown and Caldwell measurements from 2012 to 2017

This section compares the Brown and Caldwell (B&C) samples of TN and TP to samples from our study sites at the Birdsong (BS), Armadillo (AR) and Below Dam (BD) sites. Brown and Caldwell collected samples for eight events each year, four dry and four wet events beginning in 2006. However, for comparison with the samples collected in our study only the samples from 2012 to 2017 were used so sampling sites would be consistent (Figure 5a and 5b). During this period, Brown and Caldwell collected samples at only two sites, outflow of the Oconee forest pond near the dog park (MS4-4c) and outflow from the dam (MS4-4b) (Figure 1). Two additional B&C sites were added in 2017 at the same location as our Birdsong site (MP-20) and Armadillo site (MP-21).

In general, the Brown and Caldwell results showed little change in average TN from 2012 to 2017 except for a decrease perhaps at the Oconee Forest Pond (MS-4c) and Below Dam (MS-4b) sites after 2012 and a sharp increase at the Pond site in 2017. Brown and Caldwell TN concentrations from 2016 and 2017 were like the values in our study, but higher for the MS4-4c outflow of the Oconee Forest Pond with 5.70 mg/L which is upstream of the B&C site MP-20 with

2.58 mg/L. By comparison, we measured an average TN concentration at the Birdsong site of 0.80 mg/L.

Average TP concentrations measured by Brown and Caldwell were near 0.050 mg/L during all years except for high concentrations at MS4-4c during 2012 and 2017 (0.72 and 0.80 mg/L, respectively). Although our average TP concentration at Birdsong was lower than the Brown and Caldwell results, we did see a sharp increase in TP stormflow concentrations near the end of 2017 (Figure 4a). We believe these increases may have been associated with renovation work on the Oconee Forest Pond dam, Red coat band practice field and the surrounding area.

Overall, our nutrient results indicated that Lake Herrick was normally P limited (Figure 2). The TN concentrations in Birdsong and Armadillo were similar and usually less than 1.5 mg/L (Figures 4b and 4d). The NO_3^- concentrations were less than 1 mg/L during baseflow in both tributaries indicating that groundwater NO_3^- concentrations were low in the watershed. Most of the P seemed to be coming from Armadillo during stormflow and PP was the most common form (Figures 4a and 4c). This could be inorganic P sorbed to eroded sediment or scoured stream sediment. Since the Armadillo watershed includes many homes, the source could have been lawn fertilizers. Input of DOP from leaves was probably more common in Birdsong and so this did not seem to be the source of the elevated P.

E. coli

Boxplots of *E. coli* concentrations with the minimum, 25th percentile, median, 75th percentile and maximum values are shown in Figure 6. The graph is plotted in logarithmic scale.

Significantly higher concentrations of *E. coli* were found in stormflow compared to baseflow at Armadillo and Birdsong. However, concentrations at Below Dam were not significantly different between baseflow and stormflow, indicating the ameliorating effect of the

lake. The storm flow *E. coli* concentrations at Armadillo and Birdsong were roughly 30-fold higher than during baseflow.

Sixty seven percent of the baseflow samples at Birdsong and Armadillo exceeded the single sample criterion of 235 MPN/100 for the summer of 2016, whereas during winter only 20% and 40%, respectively, exceeded the criteria at these sites (Table 1). Baseflow samples for the year 2017 exceeded the standard more frequently with 83% and 17% at the Armadillo and Below Dam sites, respectively. Birdsong Creek had no samples exceeding the standard for the summer of 2017. This could be because a beaver dam caused water to pond just upstream of the sampling station. The renovation of Oconee Forest pond, Red coat band practice field started during this time and the beaver dam was removed.

In general, samples in the summer months exceeded the criteria more frequently compared to the winter months during storm and baseflow events. The winter of 2017 had 60% and 80% of the samples exceeding the standard at Birdsong and Armadillo respectively while no samples exceeded the standard at the Below Dam site during baseflow. No samples of baseflow or stormflow exceeded the criteria in 2016 at the Below Dam site. Samples at the Below Dam site exceeded the criteria during stormflow events in 2017 both during summer and winter by 50% and 20%, respectively. The reason that samples exceeded the criteria in 2017 more often than 2016 could be due to more frequent rainfall in 2017. Higher stormflow concentrations implied that runoff was the primary source of bacteria.

The average *E. coli* concentrations reported by Brown and Caldwell were lower during 2016 and 2017 than our study sites (Figure 7). This could be because of roughly 30 storm events collected in our study in comparison with 8 wet events collected by Brown and Caldwell.

Source Tracking Analysis

Microbial source tracking (MST) results for our study indicated that humans, ruminants, and dogs were sources of fecal contamination. All sites were sampled for 8 baseflow events; Birdsong and Armadillo were sampled for 13 stormflow events and Below Dam was sampled for 8 stormflow events. The percentage of samples that showed presence of the different markers is shown in Figure 8a.

MST detects the gene copies in both culturable and non-culturable cells, which we will call “genetic *E. coli*”, whereas IDEXX only detects cells that can grow, which we will call “culturable *E. coli*” (Lavender et al., 2009). In this study, the MST gave the *E. coli* gene copies in TSC (Target Sequence Copies) /100 mL or GC/100 mL (gene copies/100 mL) and the IDEXX gave the cell count in MPN (Most Probable Number) /100 mL. The two techniques are different and do not necessarily correlate. The genetic *E. coli* concentration is generally expected to be higher than the culturable *E. coli*, but results are not consistent as some experiments have shown culturable *E. coli* higher than genetic *E. coli* (Noble et al., 2010).

On sunny calm days when there is no outside contamination, genetic counts are expected to be lower (Fujioka et al., 1981) and the concentration could go undetected by the qPCR method. Genetic *E. coli* in this study were present on 6 out of 8 baseflow events (Figure 8a) at Birdsong and Armadillo, whereas culturable *E. coli* concentrations were significant in these events. Genetic *E. coli* were detected at all 8 baseflow events at the Below Dam site. During stormflow, genetic *E. coli* were detected for all samples at all the three sites and stormflow concentrations were significantly higher than baseflow concentrations for both genetic and culturable *E. coli* at all the three sites.

Combining the baseflow and stormflow samples together at the three sites (Figure 8b), the average genetic *E. coli* concentrations were 4 log₁₀ TSC /100 mL at the Birdsong site, 5 log₁₀

TSC/100 mL at the Armadillo site and 3 log₁₀ TSC/100 mL at the Below Dam site. These concentrations were comparable to the culturable *E. coli* with 3 log₁₀ MPN/100 mL at Birdsong, 4 log₁₀ MPN/100 mL at Armadillo and 2 log₁₀ MPN/100 mL at Below Dam. A one-way ANOVA run for genetic *E. coli* showed that the average concentration at the Below Dam site was significantly lower than the other two sites. This was like the trend exhibited by culturable *E. coli*. Both results indicated that *E. coli* concentrations dropped in the lake. Since the 10-fold drop did not occur with TN and TP concentrations (Figure 4) the reduction in *E. coli* concentrations was probably due to die off bacteria and not dilution.

To determine the probable sources of *E. coli*, the correlation between culturable *E. coli* and each marker was tested. Standardized regression coefficients β were calculated for genetic and culturable *E. coli* using the marker data as the independent variable and the culturable *E. coli* as the response variable. Significant positive correlations existed between the genetic and culturable *E. coli* at Birdsong ($R = 0.84$), Armadillo ($R = 0.61$) and Below Dam ($R = 0.78$) (Figure 8c), confirming that the markers and culturable *E. coli* were sampling the same populations.

Dog markers were detected twice at each site under baseflow conditions (Figure 8a). This suggested dog taking a dip in the water at the Below dam and Birdsong sites. Armadillo has residents that live close to the creek, and a dog house was spotted in one of the residences during a stream walk by the monitoring team in 2016. Dog was the most commonly detected marker after *E. coli* and there was a good correlation ($R > 0.6$) with culturable *E. coli* at all the sites (Figure 8c) indicating dogs were a dominant source. The highest concentration for the dog marker was approximately 6 log₁₀ TSC/100 mL at Armadillo. A source tracking study in Michigan found that *E. coli* in a storm sewer exceeded 10000 MPN/100 mL with no illicit connection in system. The sources were identified as pets (cats and dogs) and raccoons (Ram et al., 2007).

During baseflow, the human marker (HF183MGB) was detected only once at Birdsong and undetected at Armadillo and Below Dam (Figure 8a). The absence of human markers during baseflow indicated that there were no illicit discharges within the watershed. During stormflow, the human marker was detected 7 out of 13 times at Birdsong, 9 out of 13 times at Armadillo and 5 out of 8 times at Below Dam (Figure 8a). However, the gene copies were mostly below the detection limit of $0.70 \log_{10}$ TSC/100 mL.

The highest concentration of human marker occurred in a storm event at Armadillo with approximately $6 \log_{10}$ TSC/100 mL (Figure 8b). This indicated that there may be occasional sewer leakage during storms in the Armadillo watershed. The sewer line runs parallel to the lake shore line at the point where Armadillo Creek drains into Lake Herrick. At the Below Dam site, there was a positive correlation ($R = 0.62$) between the human marker and culturable *E. coli* (Figure 8c). One study found that human makers were more persistent than other bacteria once released into the environment (Ishii et al., 2006).

Under baseflow conditions, the ruminant marker (Rum-2-bac) was found once at Armadillo (Figure 8b) and twice at Below Dam, but undetected in the predominantly forested Birdsong tributary. Since there are no cows or goats in the Lake Herrick watershed, deer are the likely source for this marker. Deer have been spotted by the monitoring team at night on Armadillo Creek. Deer may find refuge in forested areas but forage at the forest edge and in suburban areas at night along streams which provide a route into these areas (Guber et al., 2016; Parajuli et al., 2009). Under stormflow conditions, the ruminant marker was detected 3 out of 13 times at Birdsong, 4 out of 13 times at Armadillo and 2 out of 8 times at Below Dam. However, the correlation between the ruminant marker and culturable *E. coli* was not significant (Figure 8c), suggesting deer were not a dominant source of the *E. coli*.

Overall, our *E. coli* results indicated that most of the bacteria entered the lake during stormflow, which could have been due to bacteria washed off in runoff or scoured from bacteria that were surviving in stream sediment. Concentrations in the tributaries exceeded the *E. coli* standard during baseflow and stormflow but concentrations below the dam were below the standard indicating die-off of bacteria in the lake. Armadillo Creek was the dominant source of the bacteria with an approximately 6-fold higher concentration in stormflow compared to Birdsong Creek. Dogs were a more likely source of this bacteria than humans or deer. The fact that human sources were uncommon in the Lake Herrick watershed indicated less risk of human pathogens being present (J. Soller et al., 2015).

Principal Component Analysis (PCA)

In the PCA analysis under baseflow conditions, Factor 1 explained 26% of the variance in the data and Factor 2 explained 15% of the variation (Figure 9a). The Armadillo and Below Dam sites behaved differently since they had non-zero values at opposite ends of the Factor-1 axis, whereas the Birdsong site was not correlated with Factor 1. TN, NO₃⁻, conductivity and *E. Coli* were associated with the Armadillo site. In terms of Factor 2, TP, TPN and TP were at the opposite end from the Birdsong site suggesting that concentrations were lower in this tributary compared to Armadillo, as our data showed. Sample pH, DIP, DON, and temperature plotted near zero for both factors indicating that they did not explain much of the variance.

Under stormflow conditions, Factor 1 explained 24% of the variance in data and Factor 2 explained 18% of the variance. TPN, NO₃⁻ and TN plotted together around AR for Factor 1 (Figure 9b). DON, DIP, and conductivity plotted near zero for both factors and did not explain much of the variance in data. Turbidity was correlated with Birdsong on Factor 2 which may have been due to the renovation work at the Oconee Forest Pond and Red coat band practice field. *E. coli*, TP and

TPP were correlated and they plotted close to Armadillo in terms of Factor 1, indicating perhaps that this tributary was the dominant source for these contaminants. The Armadillo and Below Dam sites plotted at opposite ends of the Factor-1 axis indicating the difference in concentrations between these sites. The general absence of any grouping of turbidity or conductivity with any of the contaminants indicated that these were not very good indicators for these pollutants.

CONCLUSIONS

In our study we found that:

- More than 50% of the combined samples were P-limited and less than 10% of the samples were N-limited at all sites. Baseflow samples were mostly P-limited with excess N and the system shifted to co-limited when P increased during stormflow.
- TP concentrations for storm samples were significantly higher than the baseflow sample concentrations. TN concentrations were remarkably similar between baseflow and stormflow samples and there were no significant differences between samples at all sites.
- More than 95% of the samples exceeded the Eco-region criteria for TN and 54% to 72% of the samples exceeded the reference Eco- region criteria for TP.
- TP and its forms did not differ statistically between sites, but TN concentrations were higher at Armadillo compared to Below Dam, indicating N transformations in the lake. Ammonium was higher at Below Dam compared to Armadillo and Birdsong, also indicating N transformations. TPN concentrations were higher at Armadillo compared to the forested Birdsong site, indicating fertilizer sorbed to eroded or scoured sediment could have been the source.

- The dominant form of P was TPP followed by DOP with less than 10% of the P being inorganic (DIP). Also, DOP increased two-fold during stormflow at all the sites, signifying that organic P dominated the dissolved P pool.
- The dominant form of N was NO_3^- , followed by TPN or DON with NH_4^+ less than 15% of the N at all sites. Nitrate concentrations generally decreased during stormflow with increased particulate and organic forms. Below Dam differed from the other two sites with increased NH_4^+ and decreased NO_3^- , suggesting transformations in the lake.
- Samples exceeded the *E. coli* criteria of 235 MPN/100 mL more during the summer than the winter. Almost all of the storm samples exceeded the criteria at the inlet sites during stormflow, implying a relationship between higher flow and bacteria. The storm flow *E. coli* concentrations at Armadillo and Birdsong were roughly 30-fold higher than during baseflow. Also, the year 2017 had more samples exceeding the criteria compared to 2016 because of the frequent rainfall events during 2017.
- Stormflow *E. coli* concentrations at Armadillo were approximately 6 and 80-fold higher than Birdsong and Below Dam. Stormflow concentrations below the dam were below the standard indicating die off bacteria in the lake. Source tracking results showed that dogs were a more likely source of this bacteria than humans or deer. The fact that human sources were uncommon in the lake indicated reduced risk for human infection (J. Soller et al., 2015) and supports the decision to reopen Lake Herrick to limited recreational use.
- The PCA analysis showed that Armadillo and Birdsong behaved differently under baseflow conditions with TN, NO_3^- , conductivity and *E. Coli* more associated with Armadillo. During stormflow Armadillo and Below Dam plotted on opposite ends of the Factor-1 axis. *E. coli*, TP, TPP were more associated with Armadillo which had higher

concentrations of these pollutants compared to Birdsong. Turbidity plotted closer to Birdsong during stormflow conditions. In general turbidity and conductivity plotted opposite to each other with the turbidity increasing during stormflows and conductivity decreasing during stormflows, but neither were good predictors of N, P, or *E. coli*

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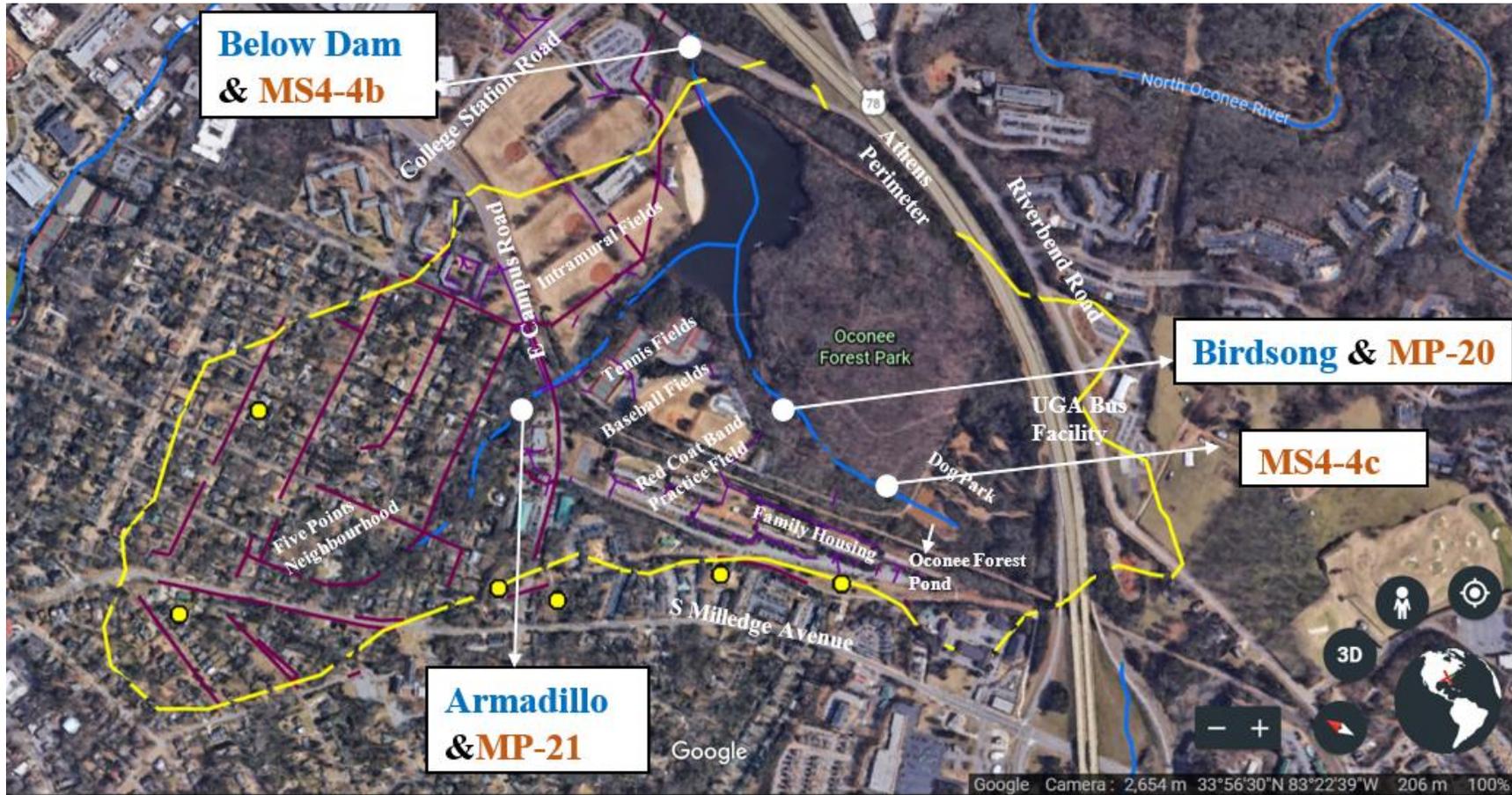
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FIGURE 1. LAKE HERRICK WATERSHED EXTENT



Legend

- Septic Systems
- Storm Lines
- Sewer Lines
- Lake Herrick Watershed
- Streams

Study Sites: Birdsong (BS), Armadillo (AR) and Below Dam (BD)

B+C Sites: MS4-4c, MS4-4b, MP-20 and MP-21

Source: Google Earth

FIGURE 2A. STOICHIOMETRIC RATIO AGAINST TOTAL PHOSPHORUS

Stoichiometric linkages established by (Guildford et al., 2000) used for data from the sites to establish the limiting nutrient.

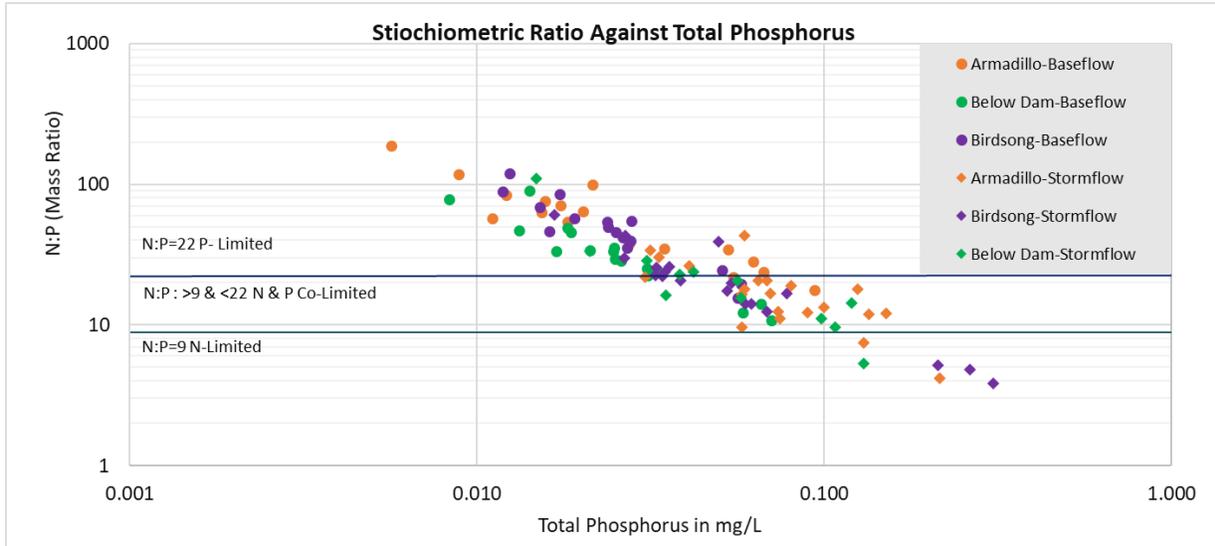


FIGURE 2B. STOICHIOMETRIC RATIO AGAINST TOTAL NITROGEN

Stoichiometric linkages established by (Downing et al., 1992) used for data from the sites to establish the limiting nutrient.

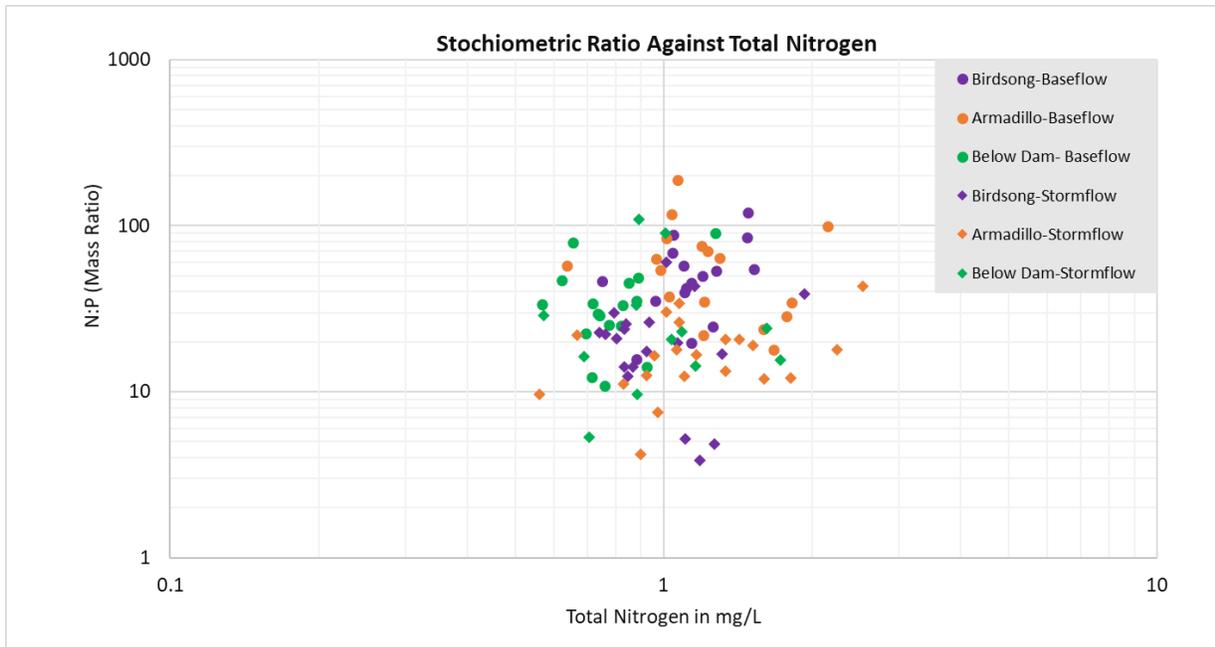


FIGURE 3A. STREAM FLOW AND PRECIPITATION AT THE BIRDSONG SITE

Primary axis shows the flow in m³/s and TP in mg/L. Secondary axis shows the precipitation in inches and TN in mg/L at the Birdsong site.

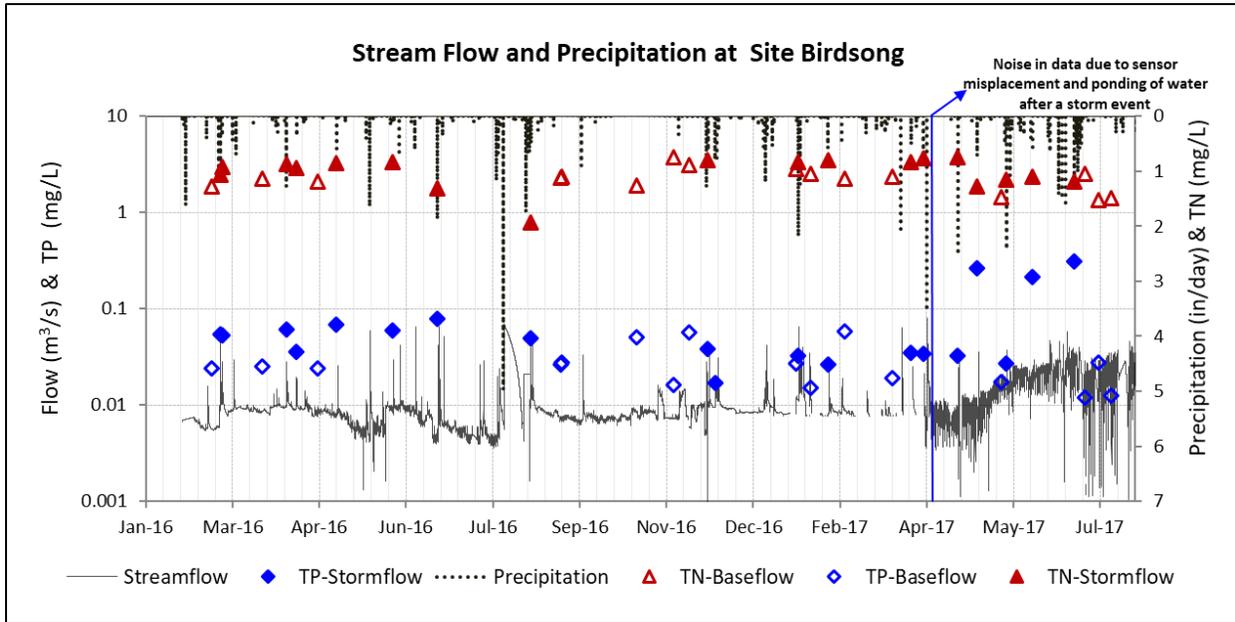


FIGURE 3B. STREAM FLOW AND PRECIPITATION AT THE ARMADILLO SITE

Primary axis shows the flow in m³/s and TP in mg/L. Secondary axis shows the precipitation in inches and TN in mg/L at site the Armadillo site.

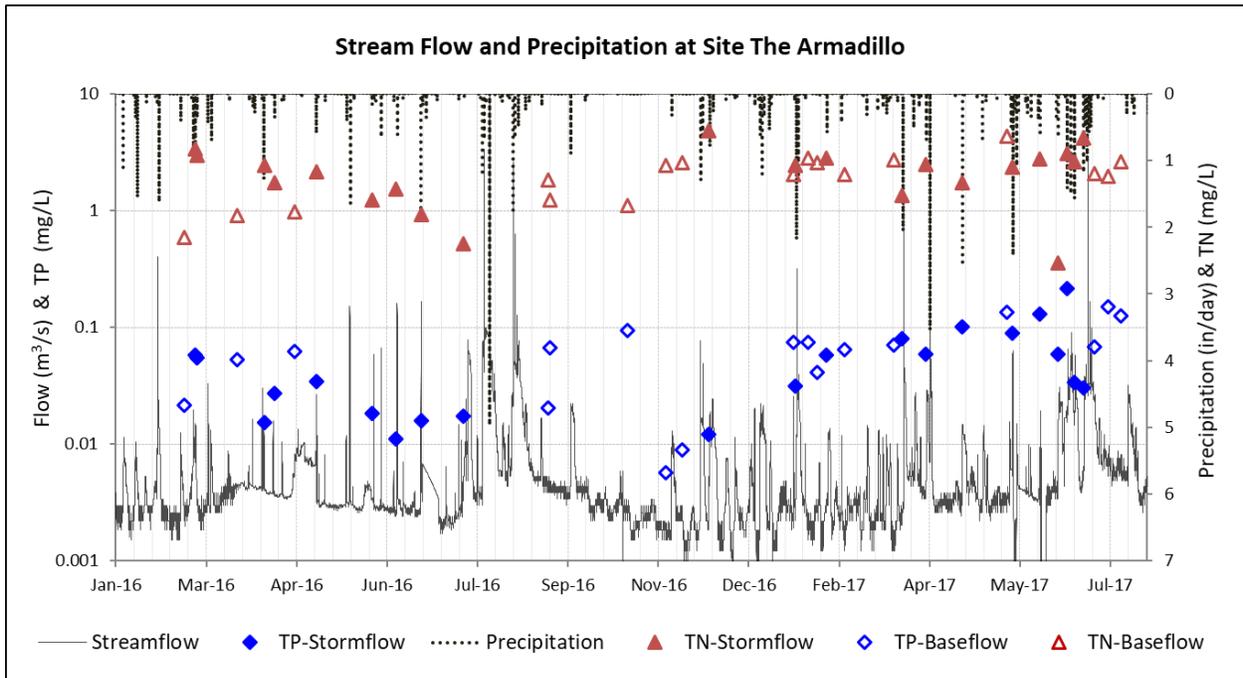


FIGURE 3C. STREAM FLOW AND PRECIPITATION AT THE BELOW DAM SITE
 Primary axis shows the flow in m³/s and TP in mg/L. Secondary axis shows the precipitation in inches and TN in mg/L at the Below Dam site.

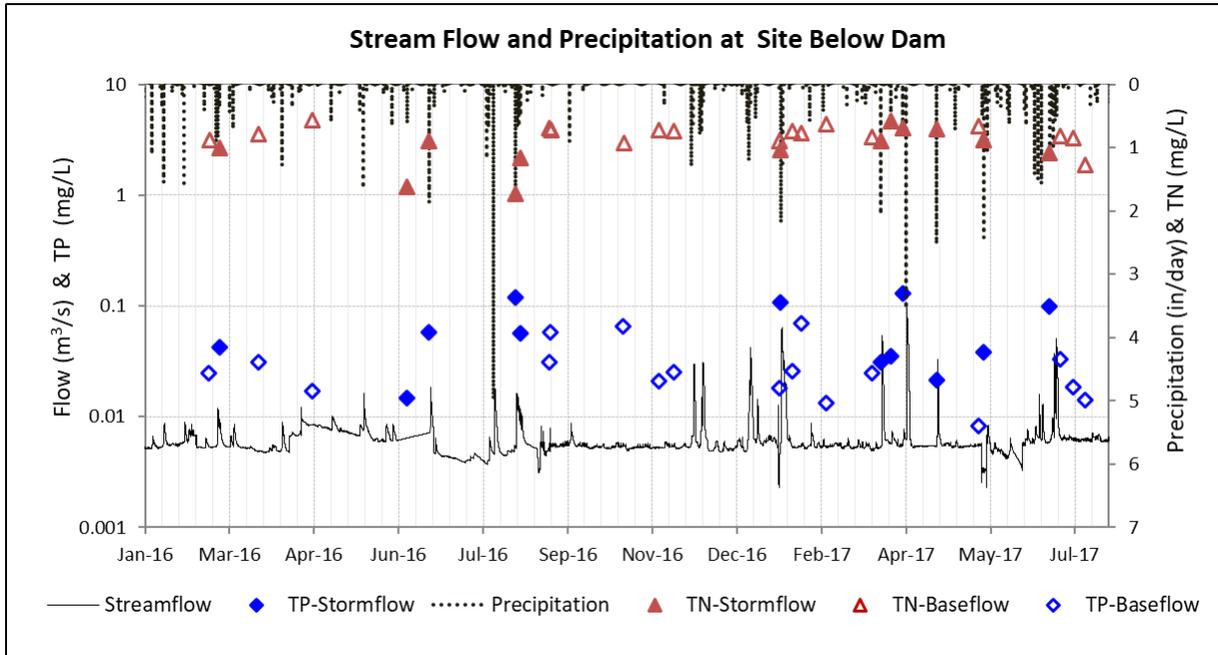


FIGURE 4A. PHOSPHORUS CONCENTRATION AT THE BIRDSONG SITE

Primary axis shows the phosphorus concentration in $\mu\text{g/L}$. X-axis shows the composition of different forms for the baseflow samples represented by BF followed by the date the sample was collected. SF followed by the date indicates stormflow samples.

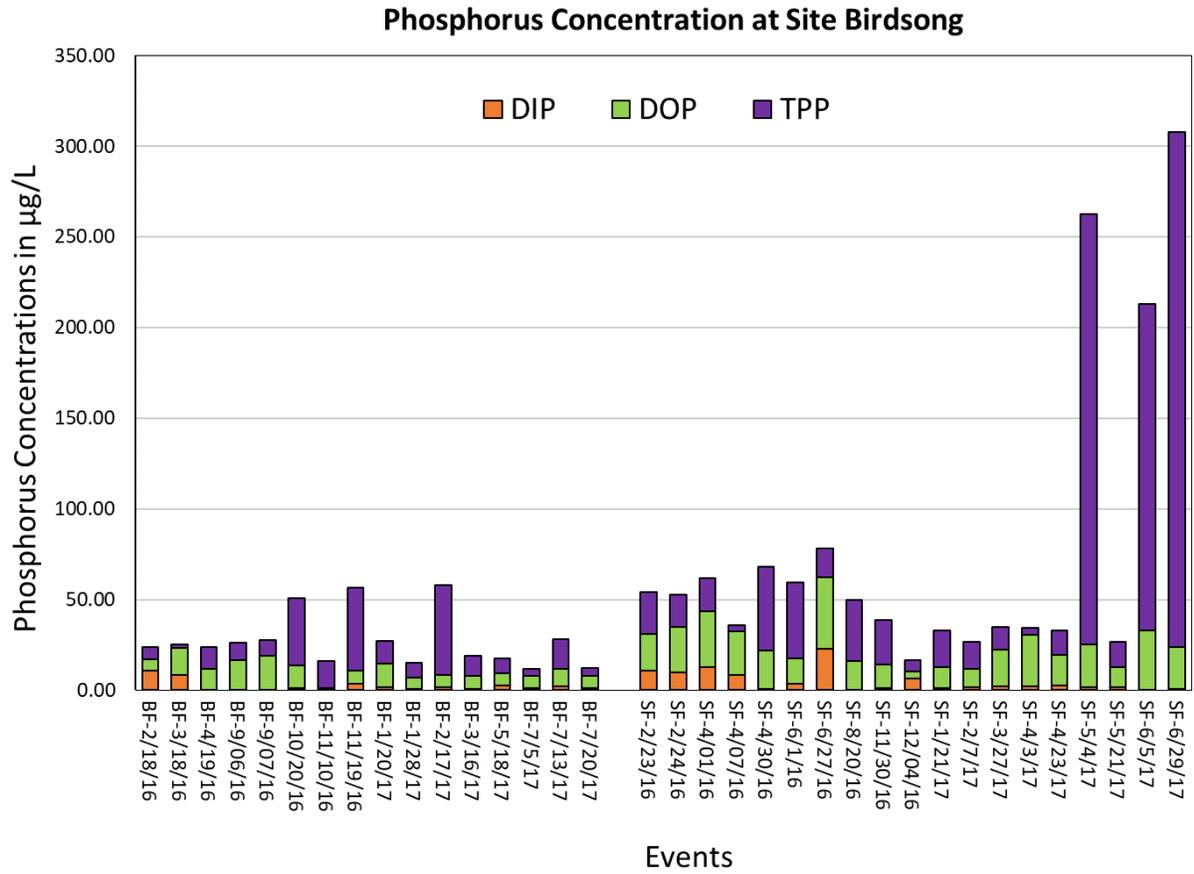


FIGURE 4B. NITROGEN CONCENTRATION AT THE BIRDSONG SITE

Primary axis shows the phosphorus concentration in mg/L. X- axis shows the composition of different forms for the baseflow samples represented by BF followed by the date the sample was collected.SF followed by the date indicates stormflow samples.

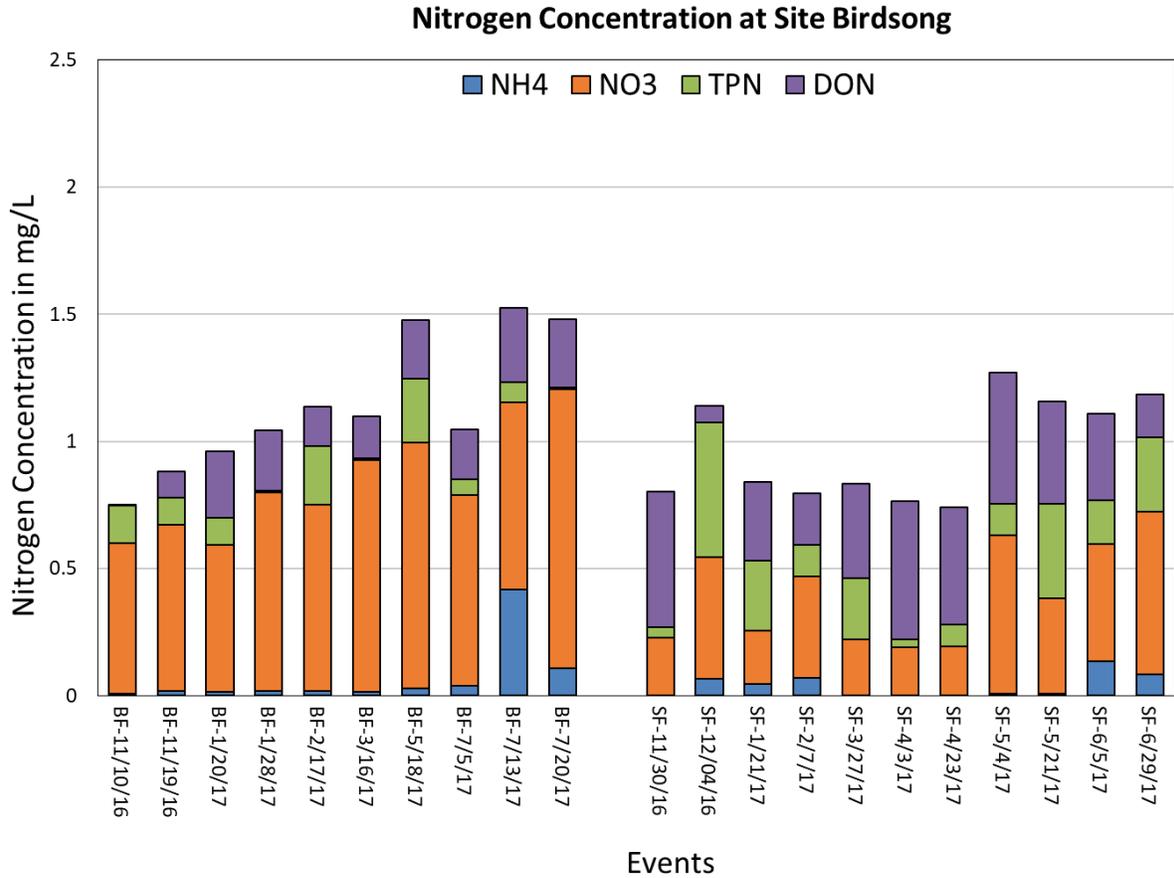


FIGURE 4C. PHOSPHORUS CONCENTRATION AT THE ARMADILLO SITE

Primary axis shows the phosphorus concentration in $\mu\text{g/L}$. X- axis shows the composition of different forms for the baseflow samples represented by BF followed by the date the sample was collected. SF followed by the date indicates stormflow samples.

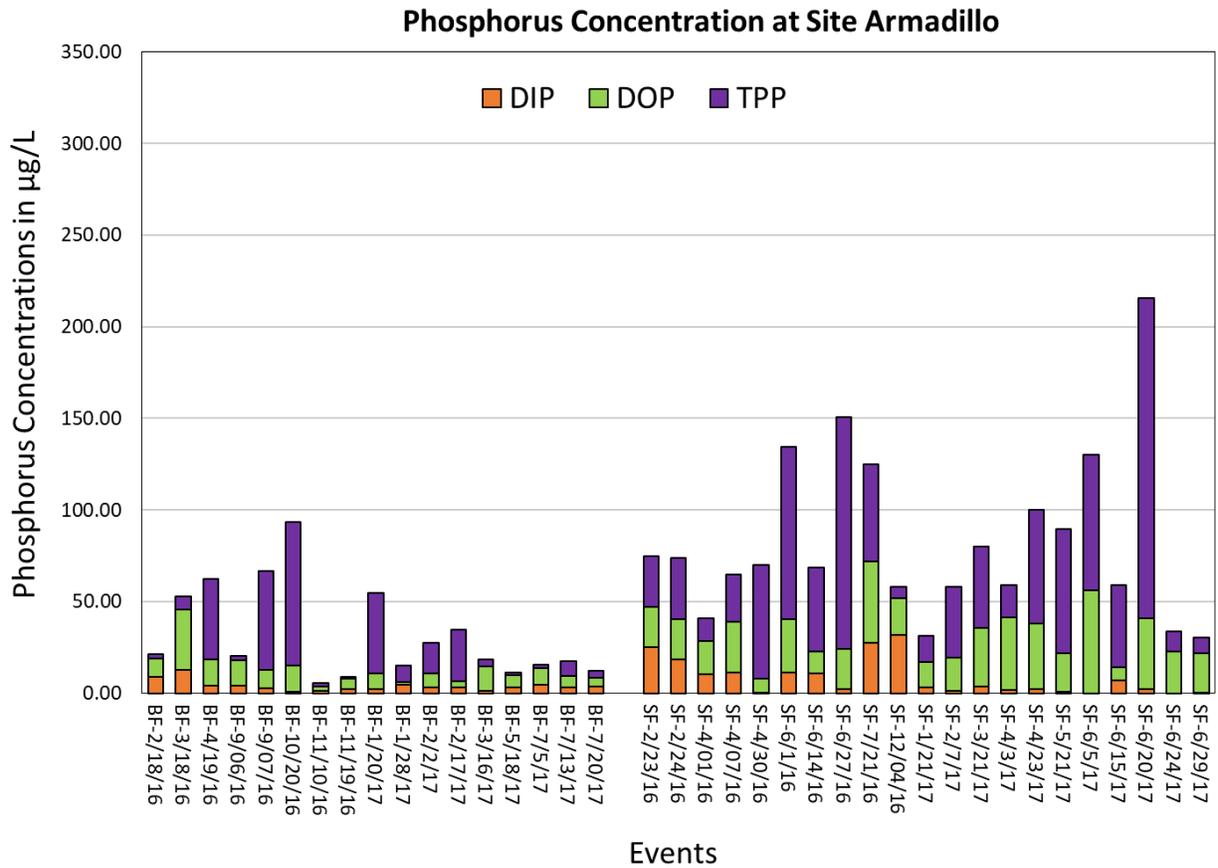


FIGURE 4D. NITROGEN CONCENTRATION AT THE ARMADILLO SITE

Primary axis shows the phosphorus concentration in mg/L. X- axis shows the composition of different forms for the baseflow samples represented by BF followed by the date the sample was collected. SF followed by the date indicates stormflow samples.

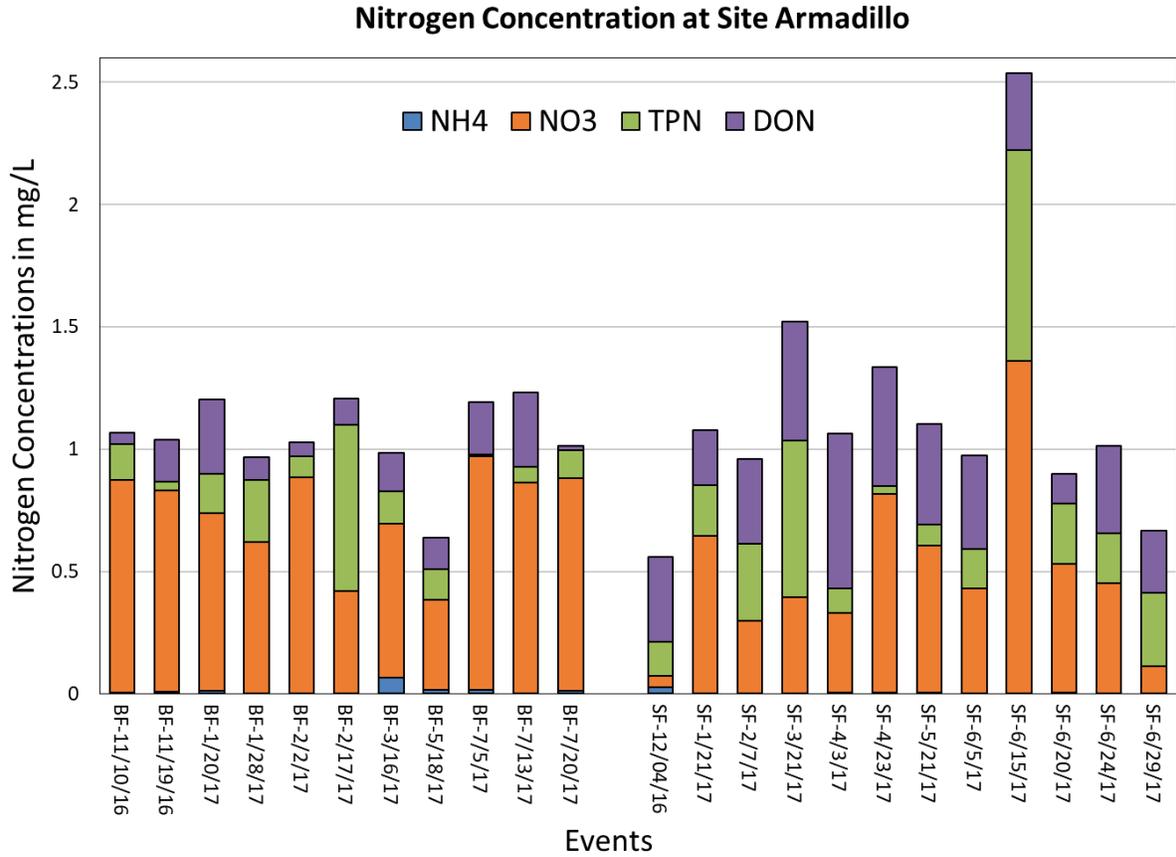


FIGURE 4E. PHOSPHORUS CONCENTRATION AT THE BELOW DAM SITE

Primary axis shows the phosphorus concentration in $\mu\text{g/L}$. X- axis shows the composition of different forms for the baseflow samples represented by BF followed by the date the sample was collected. SF followed by the date indicates stormflow samples

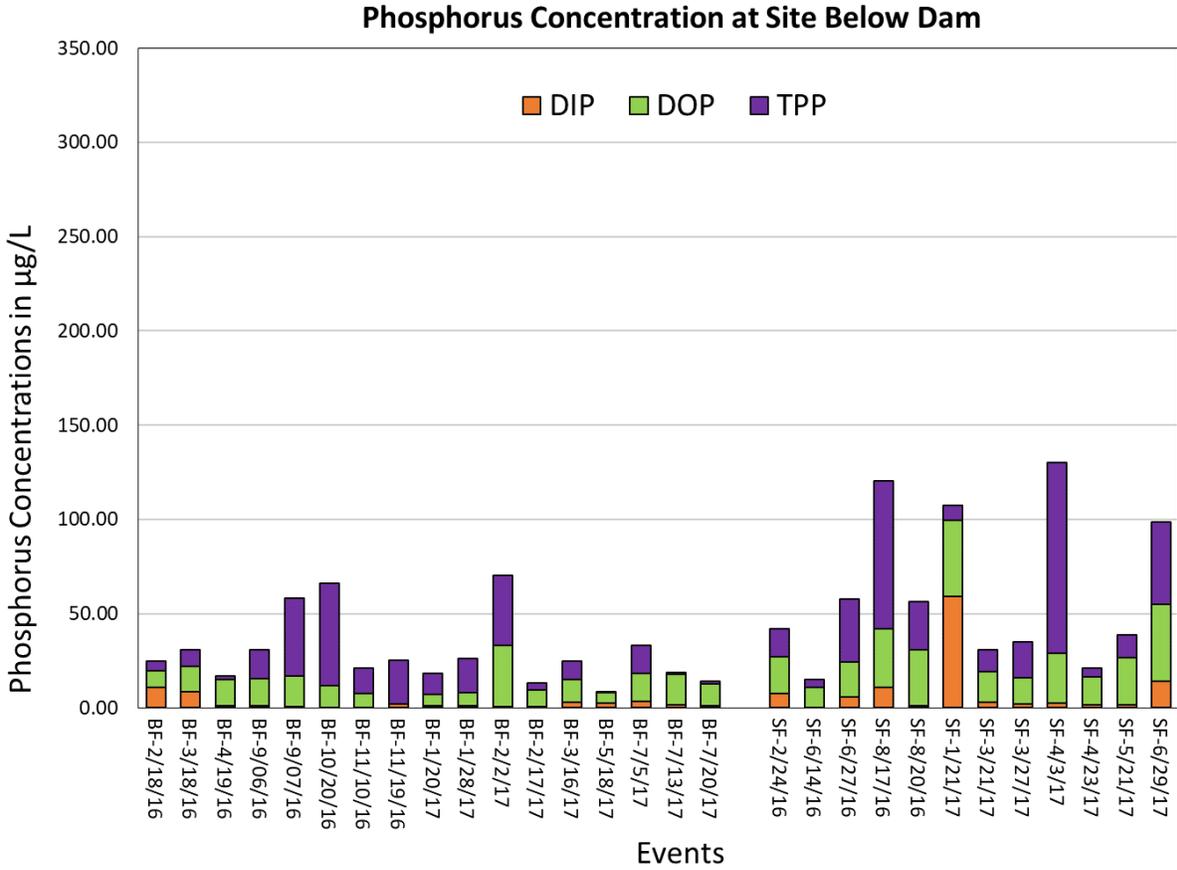


FIGURE 4F. NITROGEN CONCENTRATION AT THE BELOW DAM SITE

Primary axis shows the phosphorus concentration in mg/L. X- axis shows the composition of different forms for the baseflow samples represented by BF followed by the date the sample was collected..

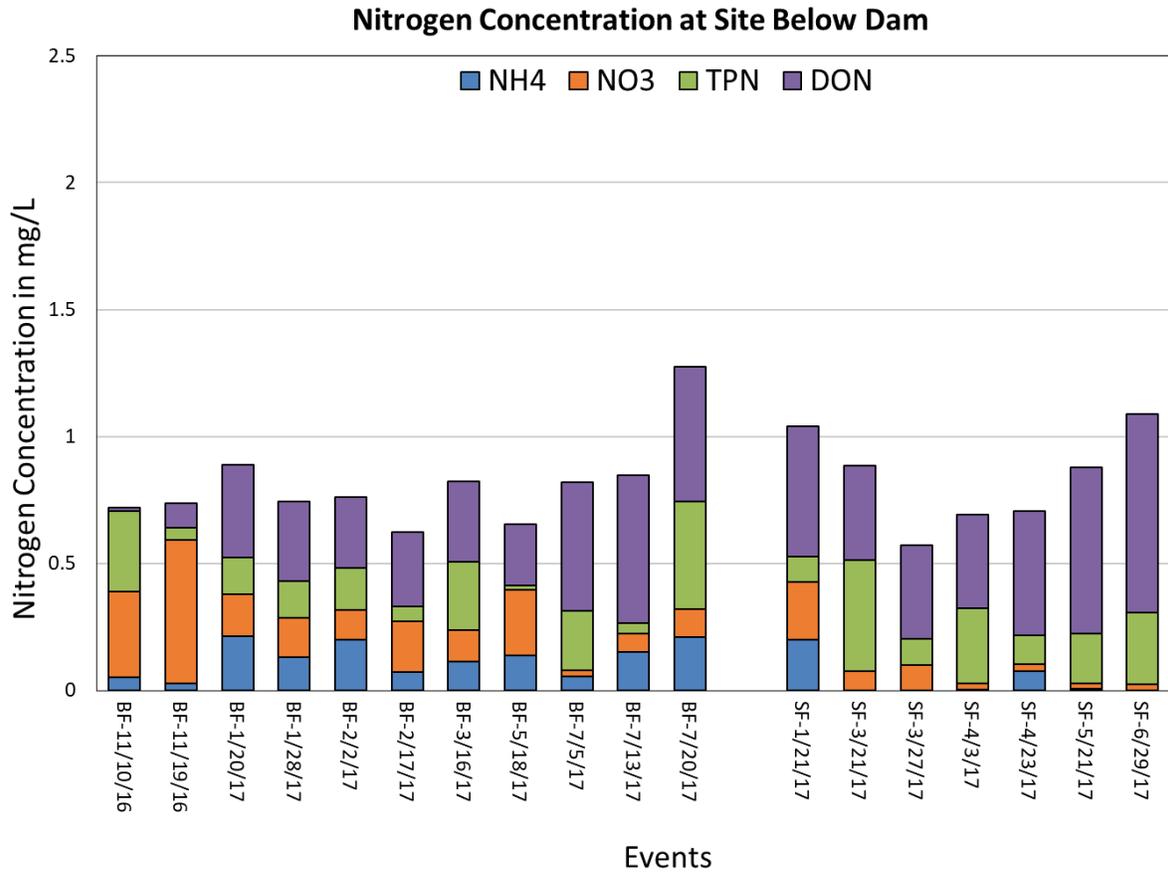


FIGURE 5A. AVERAGE TN REPORTED BY BROWN AND CALDWELL AND THREE STUDY SITES
 Average TN in mg/L (2012-2017) reported by Brown and Caldwell (sites MS4-b on below the dam, MP-21 on Armadillo, MP-20 on Birdsong, and MS-4c near the Oconee Forest Pond and former dog park) along with our study sites (BD, AR, and BS).

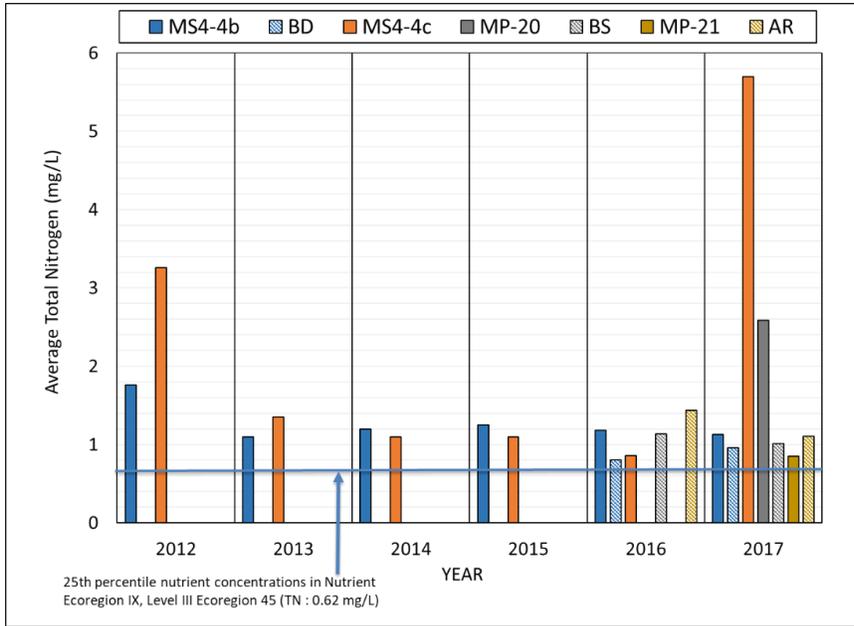


FIGURE 5B. AVERAGE TP REPORTED BY BROWN AND CALDWELL AND THREE STUDY SITES
 Average TP in mg/L (2012-2017) reported by Brown and Caldwell (sites MS4-b below the dam, MP-21 on Armadillo, MP-20 on Birdsong, and MS-4c near the Oconee Forest Pond and former dog park) along with our study sites (BD, AR, and BS)

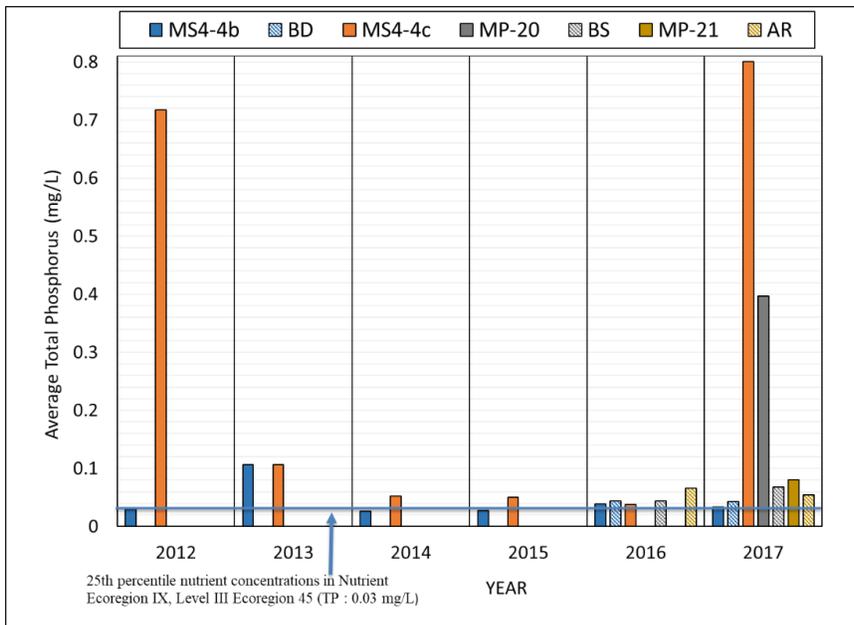


FIGURE 6. *E. COLI* MEASUREMENTS DURING BASEFLOW AND STORMFLOW

E. coli counts measured using IDEXX kits under baseflow and stormflow conditions converted to log₁₀ scale. The number of samples are shown next to the box plot. The sites are abbreviated as: Birdsong BS, Armadillo AR and Below dam BD.

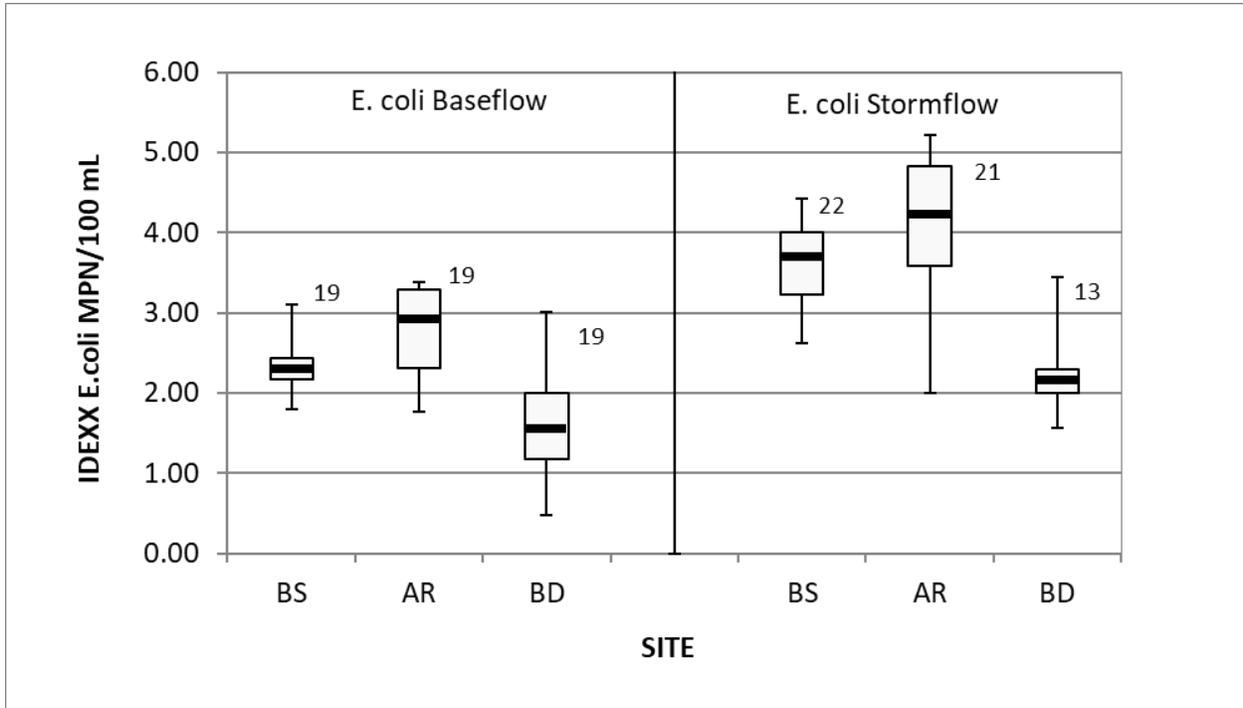


FIGURE 7. AVERAGE *E. COLI* REPORTED BY BROWN AND CALDWELL AND THREE STUDY SITES
 Average *E. coli* (2012-2017) reported by Brown and Caldwell (sites MS4-b below the dam, MP-21 on Armadillo, MP-20 on Birdsong, and MS-4c near the Oconee Forest Pond and former dog park) along with our study sites (BD, AR, and BS).

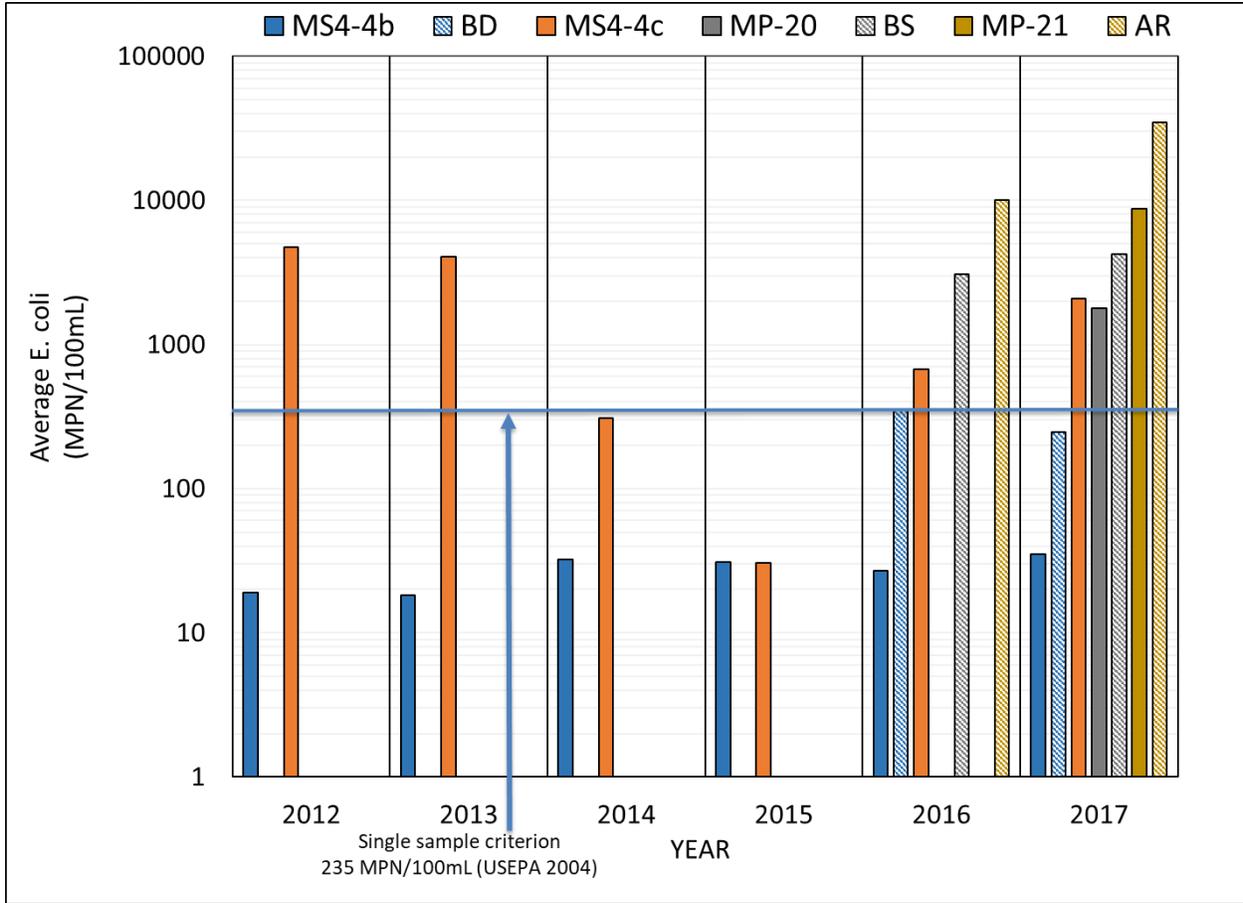


FIGURE 8A. PERCENTAGE OF MARKERS DETECTED AT EACH SITE.

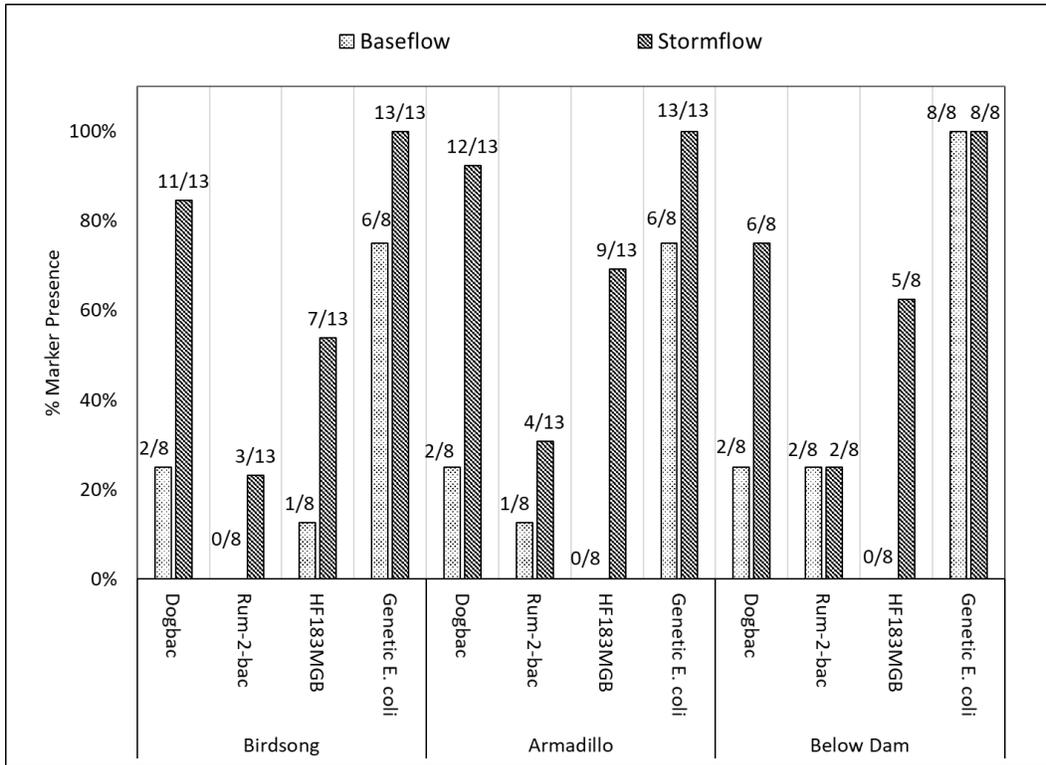


FIGURE 8B. CONCENTRATION OF MARKERS IN LOG₁₀ GENE COPIES (GC)/100 mL AT EACH SITE
 Concentration of markers in log₁₀ Gene Copies (GC)/100 mL on the x-axis against the sites abbreviated as BS (Birdsong), AR (Armadillo) and BD (Below Dam)

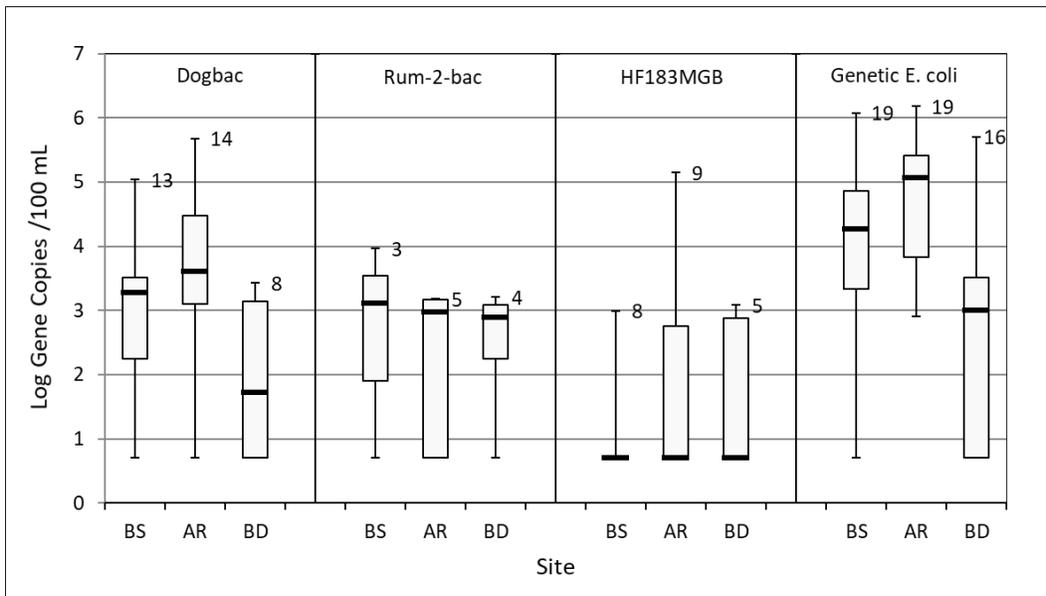


FIGURE 8C. STANDARDIZED CO-EFFICIENT B OF CULTURABLE *E. COLI* AND QPCR MARKERS AT EACH SITE

The standardized co-efficient β is the same as the Pearson's correlation co-efficient. The 95% confidence interval are measured for standardized co-efficient β and shown on the graph. If 95% confidence interval for the true slope (β) does not contain zero, they show that there is a significant positive linear association between the culturable *E. coli* and the marker. The significant linear association are flagged with an asterisk after the marker's name on the y axis.

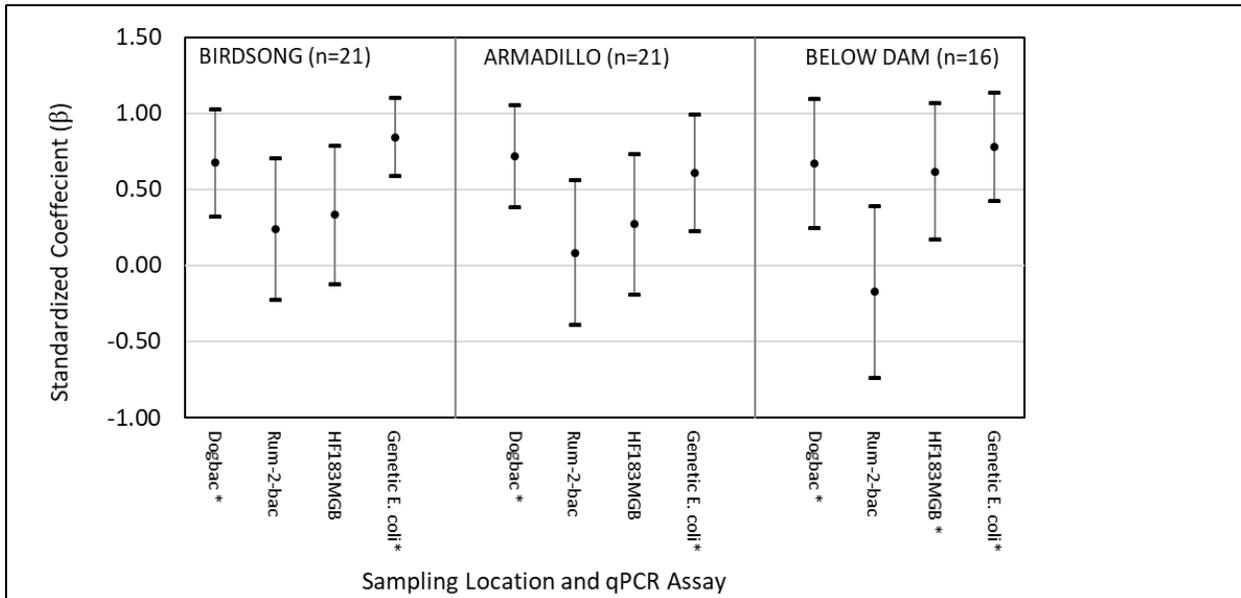


FIGURE 9A. PRINCIPAL COMPONENT ANALYSIS AT THREE SITES DURING BASEFLOW

Principal Component Analysis between TN, TPN, DON, NO₃⁻, TP, TPP, DIP, Conductivity, Turbidity, Temperature, pH, *E. coli* and the sites (BS: Birdsong, AR: Armadillo and BD: Below Dam) during baseflow conditions.

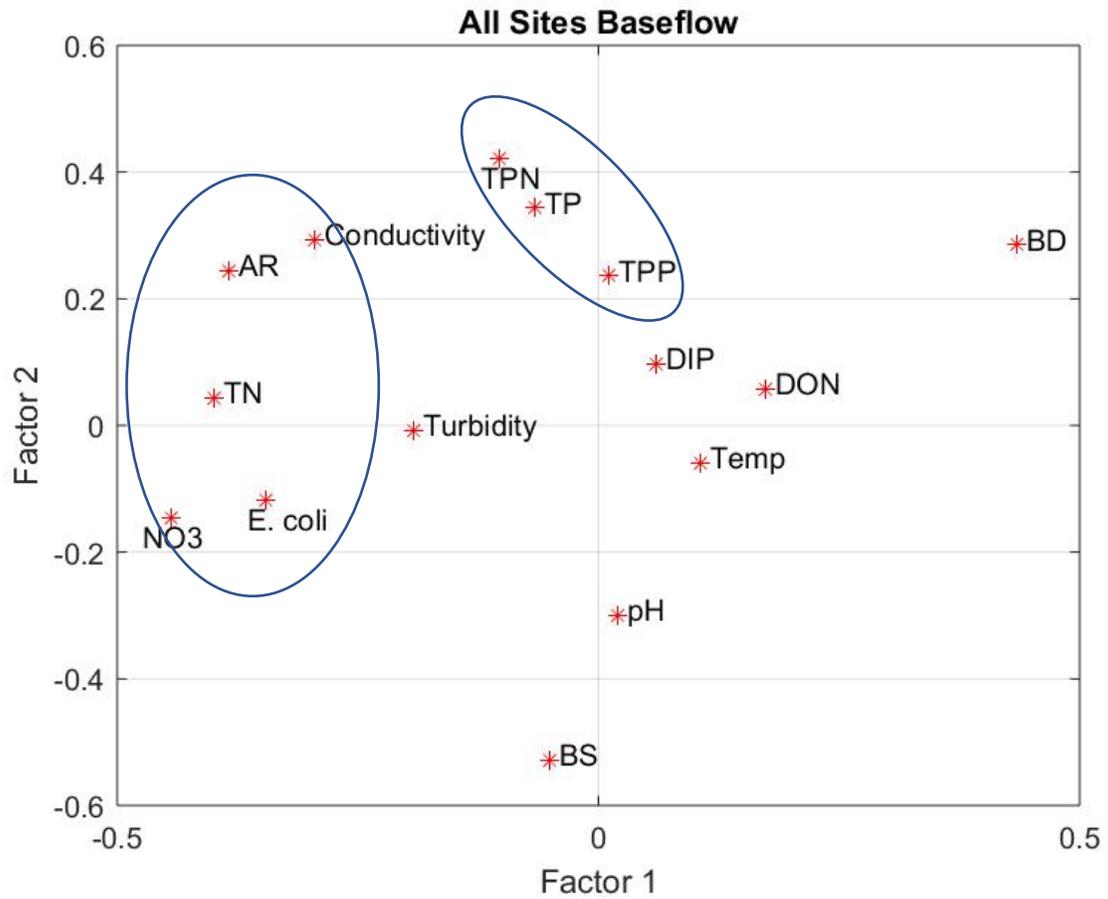


FIGURE 9B. PRINCIPAL COMPONENT ANALYSIS AT THREE SITES DURING STORMFLOW
 Principal Component Analysis between TN, TPN, DON, NO₃⁻, TP, TPP, DIP, Conductivity, Turbidity, *E. coli* and the sites (BS: Birdsong, AR: Armadillo and BD: Below Dam) during Stormflow conditions.

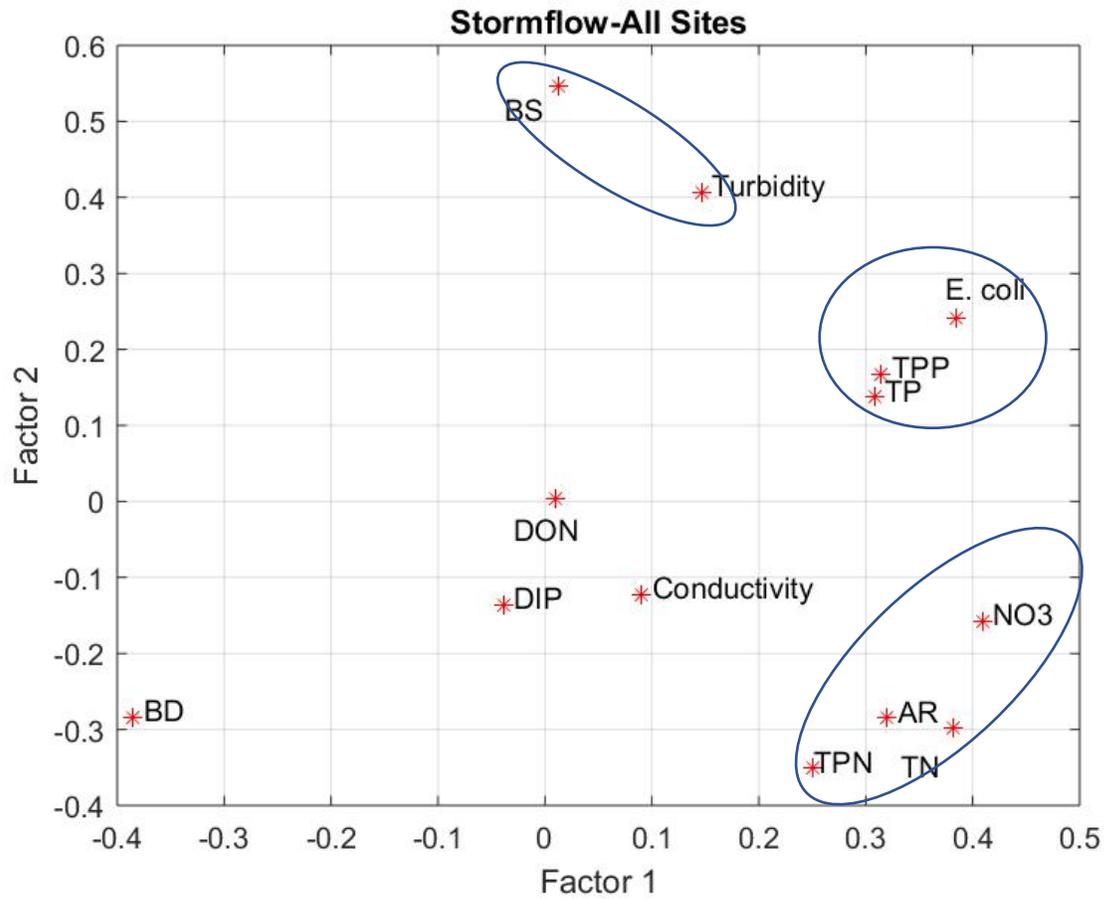


FIGURE 10. LAKE HERRICK WATERSHED MAP SHOWING SEWER LINES RUNNING PARALLEL TO STREAMS
 Source:(Christopher, 2015)



TABLE 1. *E. COLI* CONCENTRATIONS AT THREE SITES DURING THE MONITORING PERIOD

Shows the *E. Coli* concentrations at different sites during the monitoring period (2016-2017). All the *E. Coli* concentrations are in MPN/100 mL. Percentage of Baseflow and Stormflow samples exceeding the recreational water quality, single time sample criteria of 235 MPN/ 100 mL are shown in last two columns.

Season	No of Samples	Range	Mean Baseflow	Mean Stormflow	% Baseflow >235 MPN/100 mL	% Stormflow >235 MPN/100 mL
BIRDSONG						
Summer 2016	6	150-11960	518	7590	67%	100%
Winter 2016	11	100.5-10905	190	4499	20%	100%
Summer 2017	13	63-26340	147	10005	0%	100%
Winter 2017	11	141-7890	458	4643	60%	100%
ARMADILLO						
Summer 2016	6	100-142636	753	47664	67%	67%
Winter 2016	10	100-82390	977	20451	40%	100%
Summer 2017	14	181.5-164300	1247	45557	83%	100%
Winter 2017	10	57.5-92340	1202	32331	80%	100%
BELOW DAM						
Summer 2016	6	100-150	133	117	0%	0%
Winter 2016	6	3-123	43	36	0%	0%
Summer 2017	10	5-2740	202	878	17%	50%
Winter 2017	9	10-1275	37	365	0%	20%

Table 2 Conductivity and Turbidity at the three sites during the monitoring period

Parameters	Count	Minimum	25th tile	Average	75th tile	Maximum
Birdsong						
Baseflow						
Conductivity ($\mu\text{S}/\text{cm}$)	14.0	55.8	63.3	72.1	96.4	80.4
Turbidity (NTU)	17.0	5.4	9.7	31.9	130.0	41.8
Stormflow						
Conductivity ($\mu\text{S}/\text{cm}$)	18.0	42.0	52.2	68.3	74.1	148.6
Turbidity (NTU)	20.0	12.0	39.7	77.2	115.8	238.0
Armadillo						
Baseflow						
Conductivity ($\mu\text{S}/\text{cm}$)	13.0	68.7	83.5	92.5	108.8	104.2
Turbidity (NTU)	17.0	0.0	2.3	25.7	113.0	32.5
Stormflow						
Conductivity ($\mu\text{S}/\text{cm}$)	18.0	32.1	58.6	71.6	88.3	103.2
Turbidity (NTU)	20.0	8.8	17.5	44.1	65.0	130.0
Below Dam						
Baseflow						
Conductivity ($\mu\text{S}/\text{cm}$)	13.0	47.6	59.8	72.0	110.7	83.7
Turbidity (NTU)	16.0	1.8	2.6	8.6	65.0	8.1
Stormflow						
Conductivity ($\mu\text{S}/\text{cm}$)	12.0	23.0	52.9	66.5	79.0	164.5
Turbidity (NTU)	13.0	4.3	8.1	13.0	16.0	29.5

CHAPTER 3

SUMMARY AND RECOMMENDATIONS

Lake Herrick open to public for recreation is a good indication of the sustainable efforts toward preserving and improving the quality of the lake. However, careful use of lawn fertilizers is required to maintain and lower the N level in streams. The increase in P during stormflows are from the sediments washed during rainfall and the most likely source is leaves and perhaps grass clippings in the watershed area. The higher *E. coli* loading from Armadillo could be due to higher impervious sub catchments compared to the Birdsong. Also, the linear association of *E. coli* with nutrients showed urban runoff to be the major contributor of the pollutants. Source tracking results played a significant role as it ruled out both human and ruminant sources at inlet tributaries and identified dog sources as a major contributor. Hence careful attention should be paid to picking up pet wastes and excluding dog pens from riparian areas. Human markers detected at low levels during stormflow could have been due to the exfiltration from sewer lines that run parallel to almost all tributaries (Figure 10). The results could be used to develop a watershed and water quality model with continuous BMP simulation to observe and test the improvements to overall water quality.