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Descriptive and Statistical Assessment of *Escherichia coli* in a Suburban Lake for Management Purposes

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An abstract of
A thesis submitted to the Faculty of the
Rollins School of Public Health of Emory University
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Abstract

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INTRODUCTION Current research efforts focus on contamination in water bodies with obvious risk of exposure and economic impact, such as larger lakes with beaches. Improved surveillance methods increasingly link small scale outbreaks to recreational freshwater. This linkage has implications for lake management. A small neighborhood lake in suburban Georgia is the focus of this investigation into environmental predictors of Escherichia coli (*E. coli*). The study will address four aims: 1) determine if *E. coli* concentrations exceed recommended standards for recreational water, 2) assess correlations between variables to determine likely predictors of *E. coli* levels 3) develop a predictive model that may be used to 4) inform future lake management decisions.

METHODS Data was compiled from publicly available sources including Georgia Adopt-A-Stream, National Oceanographic and Atmospheric Administration Climate Data Online, National Aeronautics and Space Administration Earth Observatory, and community-hired contractor lake service reports. *E. coli* counts were collected from three sites: the inflowing creek, the outflow edge of the lake, and the outflow creek. Associations between environmental factors were assessed by Pearson and Spearman correlations to determine parameters for a predictive model for *E. coli* in Lake Avondale. Environmental variables considered include air temperature, conductivity, dissolved oxygen (DO), fecal coliform, insolation, month, pH, season, sampling site, and water temperature. Multivariate linear regression and logistic regression models were developed to investigate the relationships of environmental factors with the dependent variable, *E. coli* concentration and presence, respectively.

FINDINGS Multivariate linear regression found conductivity, log-transformed precipitation, and sampling site predict log-transformed *E. coli*. Investigation of interaction terms resulted in an over-specified model of *E. coli* concentration by conductivity, sampling site, insolation, pH, log-precipitation, water temperature, dissolved oxygen, season, interaction between dissolved oxygen and season. Logistic regression found conductivity and dissolved oxygen predict *E. coli* presence. Limitations include small sample size and convenience sampling at irregular intervals across space and time. Results suggest key environmental factors such as precipitation, conductivity, and dissolved oxygen be considered or controlled during lake management decisions. For a community actively engaged in the management of a local lake concerns regarding water quality are valid. Extending that concern to questions of health risks while perhaps not a dire concern is a reasonable discussion to promote evidence-based lake management.

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Contents

Chapter I: Introduction

Background

Epidemiology and surveillance of recreational water-associated disease outbreaks began in earnest in 1978 with expansion of the existing United States' waterborne-disease surveillance system (Yoder et al., 2004). A recreational water-associated outbreak is defined by at least two people with similar illnesses, an illness linked to a contaminant in recreational water either by location or time of exposure (Hlavsa et al., 2015). These outbreaks are reported voluntarily by public health departments across the country (Hlavsa et al., 2015; Yoder et al., 2004). Reports are then compiled in the National Outbreak Reporting System (NORS) and Waterborne Disease and Outbreak Surveillance System (WBDOSS) which are maintained collaboratively by the Centers for Disease Control and Prevention (CDC), Environmental Protection Agency (EPA), and Council of State and Territorial Epidemiologists (CSTE) (Hlavsa et al., 2015; Yoder et al., 2004). The number of states reporting outbreaks appears to be increasing since the early 1990s (Hlavsa et al., 2015; Kramer et al., 1996, Yoder et al., 2004). During that time, the number of recreational water-associated outbreaks has also risen (Hlavsa et al., 2015; Kramer et al., 1996; Yoder et al., 2004). It is unclear if this rise is results from more cases or improved surveillance; regardless there is both risk and illness that needs to be addressed.

Much research on recreational water and associated illness has focused on beaches (CDC, 1996, Edge & Hill, 2007; Edge et al., 2010; Francy & Darner, 2006; Francy et al., 2006; Francy et al., 2003; Francy et al., 2013; Gorham & Lee, 2015; Hou et al., 2006; Nevers & Whitman, 2005; Olyphant, 2005; Olyphant & Whitman, 2004; Paunio et al., 1999; Stidson et al., 2012; Whitman et al., 2004; Zimmerman, 2006). A recent assessment of the United States surveillance data for 2011-2012, documented 90 outbreaks associated with recreational exposure to contaminated water (Hlavsa et al., 2015). Twenty of those outbreaks were linked to untreated recreational freshwater (Hlavsa et al., 2015). Twenty-one of those

90 outbreaks included at least 479 cases of illness due to recreational water (Hlavsa et al., 2015). Compare those cases to the at least 431 cases associated with drinking water exposure over the same time period (Beer et al., 2015). Resulting illnesses were primarily acute gastrointestinal associated with one or multiple bacteria, parasites, toxins, or chemicals (Beer et al., 2015; Hlavsa et al., 2015; CDC, 2015; WHO, 2009}. The variety of associated symptoms may not motivate all cases to seek medical attention and be reported.

Outbreak detection, investigation, and incidence reporting are all limited by missing data (Hlavsa et al., 2015). Along with severity of symptoms, several other surveillance factors can affect missing data including: the size of the outbreak, the frequency of recreation (e.g. residential versus public waterbody), the accuracy of contaminant detection, and the compilation of case reports in a common database affect the number of reported cases (Hlavsa et al., 2015). As a result, these surveillance factors affect detection of outbreaks (Andersson & Bohan, 2001; Beer et al., 2015; Hlavsa et al., 2015; Yang et al., 2012).

Despite underreporting, both research and case studies continue to link surface freshwater to gastrointestinal illness (Ackman et al., 1997; Bruce et al., 2003; CDC, 1996Cransberg et al., 1996; Feldman et al., 2002; Hlavsa et al., 2015; Keene et al., 1994; Kramer et al., 1996; McCarthy et al., 2001; Paunio et al., 1999; Rosenberg et al., 1977; Yoder et al., 2004). These case studies include untreated lake water in a variety of environmental contexts (Ackman et al., 1997; CDC, 1996, Feldman et al., 2002). When there is a pathway for human exposure to those contaminants, there is risk of illness (EPA, 2011).

Small scale outbreaks from small bodies of freshwater do occur. Surface waters used for recreation can be the source of exposure to a variety of contaminants with ill health effects (WHO, 2003). Disease outbreaks with an identified surface water source are often associated with busy public swimming beaches, water sources with high levels of human activity (Beer et al., 2015; Hlavsa et al., 2015).

However, other recreational activities that do not involve full body contact with water such as wading and fishing can lead to low level exposure (EPA, 2011). Outbreaks occurring on a smaller, neighborhood scale may be even more likely to go undetected. However, such small isolated bodies of water may be potential sources of localized disease. Small outbreaks, residential settings of exposure, and mild illness are three of many obstacles to detecting outbreaks (Hlavsa et al., 2015). Water quality assessment of small, untreated, recreational, freshwater lakes such as those located in urban or suburban environments is expanding these epidemiologic and environmental health studies beyond the public beach environment {Honkonen & Rantalainen, 2013; Melymuk et al., 2014; Melymuk et al.,, 2011; Nevers & Whitman, 2005; Wada et al., 2006; Young & Thackston, 1999}.

With growing public health awareness and improved surveillance methods, detection of smaller outbreaks is increasingly possible (Hlavsa et al., 2015). Establishing likelihood of illness involves two preceding steps: identifying possible sources of contamination capable of causing illness and determining possible routes of exposure for those contaminants (Heymann, 2015). There is a need to develop simple methods for monitoring small neighborhood lakes in order to manage the lake in a way that minimizes adverse public health effects. This study will focus on assessment of the water quality of untreated, recreational, freshwater lake located in a suburban community as modeled by potential environmental determinants of *E. coli*. Methods for monitoring small, neighborhood lakes are necessary for effective, evidence-based management of the lake ecosystem with public health in mind.

A growing number of case studies as well as research studies suggest an association between gastrointestinal distress and ingestion of contaminated surface water. Multiple studies suggest or even demonstrate transmission of pathogenic *E. coli* O157:H7 from exposure to freshwater (Ackman et al., 1997; Bruce et al., 2003; CDC, 2016; Feldman et al., 2002; Hlavsa et al., 2015; Keene et al., 1994; Kramer et al., 1996; McCarthy et al., 2001; Paunio et al., 1999; Rosenberg et al., 1977; Yoder et al., 2004). A 1995 CDC case study in Illinois linked cases of twelve patients infected with *E. coli* O157:H7 to swimming in a 50-acre untreated lake (CDC, 1996). Another case study investigated by Ackman et al., 1997 associated twelve *E. coli* O157:H7 infections to swimming in a 5-acre, 1.5 meter deep lake at a park in New York (Ackman et al., 1997).

Sources for microbial contamination vary from surface runoff to wastewater to disturbed sediment (Honkonen & Rantalainen, 2013; Laenen & Risley, 1997; Melymuk et al., 2014; Olyphant et al., 2003; Olyphant & Whitman, 2004; Selvakumar & Borst, 2006; Tornevi et al., 2014; Unckless & Makarewicz, 2007; USGS, 2002). Wastewater from sewer outflows or septic systems can also impact lake water quality (Fong et al., 2007; Hill et al., 2006; Sales-Ortells & Medema, 2014; Selvakumar & Borst, 2006; Young & Thackston, 1999). Studies of freshwater lake contamination show animal feces deposited along the shore can contribute enteric pathogens to the water (Edge & Hill, 2007, Colford, 2007; Jellison, 2009). Such pathogens include the enteric pathogens *E. coli*, *Campylobacter*, and *Cryptosporidium*.

Indicator organisms are a proxy for pathogens which are often difficult to measure or detect (Haack et al., 2009). Pathogens include bacteria, protozoa, and viruses (Heymann, 2015). Enteric pathogens affect the digestive system, particularly the intestines ("Enteric,"). Protozoa are shed by carriers in lower volumes than bacteria or viruses (Feachem et al., 1983). However, enteric bacteria generally have a higher infectious dose (Feachem et al., 1983). Particularly with small outbreaks, it is difficult to trace the aetiologic agent to its source. Yet, enteric bacteria such as *E. coli* and *Campylobacter* have caused outbreaks in recreational freshwater (Hlavsa et al., 2015) while runoff and waterfowl are known to contribute these same bacteria to lake water (Fallacara et al., 2001; Moriarty et al., 2011).

Study Site and Population

Located in the Upper Ocmulgee River Watershed, the City of Avondale Estates located in DeKalb County, Georgia is a 1.23 square mile community home to about 3,360 residents (Adopt-A-Stream, 2011-2012; Griffin, 1983). Lake Avondale, located in what is now named Bess Walker Park, has been a gathering place for community members since the city’s foundation in 1924 ("BBQ [image]," n.d.; C. Warren, personal communication, November 19, 2015; "Fast Facts," n.d.; Google, 2016; "History," n.d.). The park is open to a variety of events and recreational activities including fishing and dog-walking (The Lake House at Avondale, 2015; Warren, 2015). Though swimming and boating are currently prohibited, some adults recall swimmers in the lake during their childhood (C. Warren, personal communication, November 19, 2015; The Lake House at Avondale, 2015). The Lake Avondale Advisory Board (LAAB), committee of community members, has managed the lake since 2004 (C. Warren, personal communication, November 19, 2015; The Lake House at Avondale, 2015). The LAAB advises the city manager on matters including waterfowl populations, fishing, park vegetation, and drainage around the park, runoff, and lake dredging (City of Avondale Estates, n.d.; C. Warren, personal communication, November 19, 2015). LAAB also approves local residents interested in conducting school or citizen science projects such as measuring water quality and maintaining native vegetation (C. Warren, personal communication, November 19, 2015). LAAB duties encompass several environmental factors that influence water quality including runoff from roads and residences, accumulation of sediment in the lake, and what water flows downstream (Georgia Adopt-A-Stream, 2011-2012).

Problem Statement

Growing public health awareness and improved surveillance methods make detection of smaller outbreaks more likely (Hlavsa et al., 2015). Development of simple methods for monitoring neighborhood lakes will facilitate and inform lake management decisions that minimize adverse ecological and public health effects. Recreational activities from swimming to wading to fishing all carry some risk of exposure to contaminants in the water (EPA, 2011). In the best interest of maintaining water bodies suitable for community enjoyment, the Lake Avondale Advisory Board (LAAB) that oversees Lake Avondale, a 5-acre suburban lake (Brown & Slovisky, 2015b) in Avondale Estates, Georgia , uses analysis of water quality to inform lake management decisions (C. Warren, personal communication, April 15, 2016). The LAAB is interested in improving on evidence-based decision making of Lake Avondale.

Purpose Statement

To effectively and efficiently manage the lake environment, there is a need to understand factors influencing water quality of Lake Avondale. Past water quality assessments based on lake uses (e.g. fishing, children touching the water) were used to inform frequency of sampling and parameters to measure (C. Warren, personal communication, April 15, 2016). Through that process concerns over goose presence at the lake arose (C. Warren, personal communication, April 15, 2016). It follows, therefore, that the purpose of this project is to provide the Lake Avondale Advisory Board (LAAB) with resources to inform lake management questions as they arise. The goal of evaluating lake water quality though potential environmental predictors of *Escherichia coli (E. coli*) concentrations in Lake Avondale will be achieved through four specific aims.

Specific Aims

1. Determine if *E. coli* levels have exceeded World Health Organization (WHO) recommended standards for recreational waters
2. Assess potential correlations between environmental variables to determine likely predictors of *E. coli* levels in Lake Avondale
3. Attempt to develop a predictive model describing *E. coli* concentrations in Lake Avondale based on observed and literature-supported associations between *E. coli* and environmental variables
4. Provide recommendations to the Lake Avondale Advisory Board (LAAB) for managing *E. coli* levels in the lake and future work in the area of neighborhood lakes

Significance Statement

This project is a response to community-initiated concerns for public health and a desire for greater knowledge. The outputs of this project will be a resource for the Lake Avondale Advisory Board (LAAB) both when managing Lake Avondale and responding to community questions about the lake and public health. Ongoing lake management may include monitoring water quality and assessing exposure. This investigation is site-specific to Lake Avondale and its microclimate. Yet relationships between environmental predictors of water quality found here may suggest relationships to investigate in other lakes. Furthermore, the practice of using these relationships for evidence-based lake management can hopefully be applied to other small urban or suburban lakes.

Research Question

Which environmental variables can be used to predict *E. coli* concentrations in Lake Avondale?

Hypothesis

Canada goose feces are a significant predictor of *E. coli* concentrations in Lake Avondale.

Ethics

Emory University Institutional Review Board (IRB) reviewed study protocols. The IRB exempt this project from full review.

Chapter II: Literature Review

Recreational Water-Associated Outbreaks

Disease outbreaks have been traced to contamination of both drinking and recreational water (Ferguson et al., 2003; Hauri et al., 2005; Hlavsa et al., 2015; Keene et al., 1994; Kramer et al., 1996; Levy et al.,, 1998; McCarthy et al., 2001; Mead et al., 1999; Paunio et al., 1999; Pintar et al., 2009; Rosenberg et al., 1977; Washington Department of Health, 2014; WHO, 2009; Yoder et al., 2004). The majority of outbreaks are recorded across the United States during the months of June, July, and August (Hlavsa et al., 2015). Investigations of recreational waters often focus on swimming beaches, areas of high exposure where accidental ingestion of water is more common (CDC, 1996). However, fewer studies describe pathogen transmission from untreated freshwater (e.g. lakes, rivers) used for recreation (Feldman et al., 2002; Gorham & Lee, 2015; M. C. Hlavsa et al., 2015). However, Potential exposure to waterborne pathogens, toxins, or chemicals when visiting the park is a reasonable concern (Edge & Hill, 2007a; Hlavsa et al., 2015; Yoder et al., 2004).

Influx of human sewage and runoff are often-cited pathways of contamination, however runoff from roads and lawns can also contribute to poor water quality by adding chemicals, nutrients, and microbes (Barbosa & Hvitved-Jacobsen, 1999; Bjorklund et al., 2009; Ferguson et al., 2003; Honkonen & Rantalainen, 2013; Kayhanian et al, 2007; Naffrechoux, et al., 2000; Ribeiro et al., 2012; Roinas et al., 2014; Selvakumar & Borst, 2006; Zhang et al., 2009). Runoff containing animal feces can contribute to contamination of water with fecal pathogens (Sterk et al., 2016; Tyrrel & Quinton, 2003).

Once contaminated there are several routes that can lead to human exposure to the contaminant potentially resulting in illness. While an outbreak summary for the year 2007-2008 found the majority of the 134 recreational water outbreaks were due to ingestion (81/134, 60.4%) other exposures comprised the remaining 40% with contact being the second-most common exposure at 18.7% (25/134)

(Hlavsa et al., 2011). Inhalation accounted for 13.4% (18/134), combined routes of exposure for 4.5% (6/134), and unknown routes for 3.0% (4/134) (Hlavsa et al., 2011).

Literature strongly suggests that Canada geese feces can contribute to outbreaks of enteric diseases from human exposure to recreational waters containing goose feces (Gorham & Lee, 2015). Limited data exists on strain-specific links of intestinal microbes of Canada geese to etiologic agents of reported outbreaks (Lu et al., 2009). However, identification and implication of specific microbial strains during outbreaks is an emerging technology (CDC, 2016). This microbial source tracking is a recent strategy applied to identifying contributions by Canada geese to fecal contamination of surface waters (Gorham & Lee, 2015; Lu et al., 2009).

Pathogens of Interest

This investigation is concerned with waterborne pathogens, including bacteria, parasites, and viruses. The presence and concentration of each of these pathogens in water can vary based on a number of environmental factors (Clasen et al., 2015). Given the wide variety of organisms, this investigation will focus on enteric pathogens. Bacteria and parasites account for the majority of gastroenteritis cases resulting from exposure to untreated freshwater 1990s (Hlavsa et al., 2015; Kramer et al., 1996; Yoder et al., 2004); though a few cases resulting from viruses were reported in the most recent assessment (Hlavsa et al., 2015). Other pathogens typically associated with foodborne outbreaks, such as *Salmonella*, outbreaks can be found in contaminated water that infects humans directly or subsequently contaminates food (Benjamin et al., 2013).

Enteric Bacteria

Enteric bacteria can have human, animal, or environmental hosts (Heymann, 2015; Levy et al., 2012; Luo et al., 2015). Common bacteria in this category include *Escherichia coli* (*E. coli*) (discussed in a later section), *Salmonella, Shigella,* and *Campylobacter*. These bacteria are excreted in large numbers and can also reproduce in the environment away from their host and live for days to months (Feachem et al., 1983). Though the work of Gorham and Moriarty 2012 suggest survival time is influenced by climate, finding that *Campylobacter jejuni* from Canada goose feces survives closer to 1 week in the winter rather than a couple days in the summer (Gorham & Lee, 2015; Moriarty et al., 2011). Regardless of season, while bacteria tend to occur in high quantities, they also have a relatively high infectious dose compared to viruses and parasites (Feachem et al., 1983).

Salmonella, commonly associated with poultry, has been detected in Canada geese but seems to be rare (Gorham & Lee, 2015; CDC, 2015b). *Campylobacter* is a common cause of diarrhea (CDC, 2014). Interestingly, Carter 1987 found that *Campylobacter* spp. did not correlate with fecal coliforms in untreated surface waters (Carter et al., 1987) though it is found in the presence of *E. coli* (Bolton, 1982).

Protozoan Parasites

In addition to bacteria, protozoan parasites are common causes of disease due to contamination from human or animal feces (Heymann, 2015). These enteric protozoa can persist in the environment for days to months, however they do not reproduce outside of the human host but animals can act as a passive reservoir (Dawson, 2005; Feachem et al., 1983; Heymann, 2015; Zhou et al., 2004). *Giardia* and *Cryptosporidium* are two protozoa found in surface waters that can cause gastrointestinal problems in drinking water (Swistock et al., 2006). Health effects tend to be more severe for children and the immunocompromised (Heymann, 2015).

Parasites have a low infectious dose compared to bacteria, on the order of hundreds of organisms (e.g. a median infectious dose of 132 oocysts for *Cryptosporidium parvum*) (DuPont et al., 1995). Furthermore, parasites can survive in the environment for a relatively long time, up to months (Heymann, 2015).

Enteric Viruses

Enteroviruses have a low infectious dose relative to enteric bacteria and enteric protozoa (Heymann, 2015; Levinson, 2014; Ryan & Ray, 2014). The strains infectious to humans are usually unique to humans, though some human viruses are vector-borne (Heymann, 2015). Like enteric protozoa, viruses do not reproduce in the environment but may persist for months (Lawley, 2013; Seitz et al., 2011).

Rotavirus, is a common enteric virus. However, Victoria et al., 2014 found that it did not correlate well with bacterial indicators such as *E. coli* (Liang et al., 2015). Points raised in explanation of this absence of correlation include different structures and effective inactivation methods for each type of organism (Liang et al., 2015).

Other Pathogens

Research and media coverage address additional adverse health effects from exposure to recreational water. Recent such news casts include West Nile Virus and the "brain-eating amoeba" *Naegleria* fowleri (CDC, 2003; Visser, 2015). In August 2015, West Nile virus was detected in mosquitos from Lake Avondale in DeKalb County, Georgia during the expected summer emergence of the virus (Visser, 2015). Vector-borne illnesses such as West Nile virus are transmitted via different exposure pathways (Day, Tabachnick, & Smartt, 2015; Hayes et al., 2005) than the gastrointestinal diseases described above. Similarly, illness such as primary amoebic meningoencephalitis is caused by the free-living amoeba *Naegleria* (Griffin, 1983; Wellings et al., 1977). *Naegleria fowleri,* a pathogenic, free-living amoeba is capable of living in freshwater and tolerant of high temperatures, but not an enteric pathogen (Griffin, 1983).

Pathogens such as West Nile Virus and *Naegleria fowleri* will not be assessed by this investigation. It is important to note that there are multiple species of each of the above-mentioned organisms and that note all species are pathogenic to humans (Gorham & Lee, 2015). Given the different disease-causing organisms and different exposures, the models and management strategies of interest for this project apply only to enteric pathogens for which *E. coli* is a suitable indicator organism.

Indicator Organisms

Indicator organisms are used as a proxy measure for the presence of other organisms (Haack et al., 2009; K. Levy et al., 2012). Proxy measures forego the need to measure pathogens directly in water, an attempt to provide a standard measure in a multitude of settings. However, there are a number of limitations of this method, not limited to the varying degrees of association between a given indicator and the pathogen of interest (Haack et al., 2009; K. Levy et al., 2012). However, *E. coli* currently remains a common indicator of fecal contamination and indicator of human enteric pathogens in freshwater (K. Levy et al., 2012). In some cases, relative relationships between pathogen presence and indicator *E. coli* presence make this microbe a useful indicator (Francy et al., 2013).

Sources of Contamination

Surface water contamination occurs from a variety of sources including wastewater discharge, runoff, and human activity (Bjorklund et al., 2009; Hill et al., 2015; Honkonen & Rantalainen, 2013; Jellison et al., 2009; Scopel et al., 2006; Tyrrel & Quinton, 2003; Wada et al., 2006; Waltham et al., 2014; Wu et al., 2014). Contamination from these sources includes both chemical and biological agents (Bjorklund et al., 2009; Hill et al., 2015; Honkonen & Rantalainen, 2013; Jellison et al., 2009; Scopel et al., 2006; Tyrrel & Quinton, 2003; Wada et al., 2006; Waltham et al., 2014; Wu et al., 2014). Runoff contamination includes chemicals washed from roadways and animal feces washed off surrounding land (Bjorklund et al., 2009; Edge & Hill, 2007; Naffrechoux et al., 2000; Wada et al., 2006; Waltham et al., 2014; Zhang et al., 2009). Agricultural and farm runoff in particular can contribute both chemicals and biological contaminants to nearby waterways (Hill et al., 2006). Biological contributions from runoff are primarily animal feces while wastewater discharge such as from sewer outflows is primarily human fecal matter (Edge & Hill, 2007;

Hill et al., 2006; Melymuk et al., 2014; Young & Thackston, 1999). Storm water outflows may combine with wastewater outflows adding a mixture of contaminates (Edge & Hill, 2007; Hill et al., 2006; Melymuk et al., 2014). Even in urban areas, runoff usually contributes more to fecal coliform concentrations than sanitary sewer overflows, at least for streams (Young & Thackston, 1999).

Measures of Microbial Water Quality

Coliform Bacteria

Coliform bacteria is a broad category of bacteria some of which cause waterborne illnesses (Swistock et al., 2006). Fecal coliforms from both human and animal waste is one sub-group of coliform bacteria (Swistock et al., 2006). Total coliform bacteria and *E. coli* are common water quality measurements of greatest concern where water is used for human drinking, animal drinking, or swimming (Swistock et al., 2006). The World Health Organization (WHO), EPA, and state of Georgia have regulations for total or fecal coliforms and *E. coli* allowable in drinking and recreational waters (Georgia Adopt-A-Stream, n.d.-a; EPA, 2003; WHO, 2003). See Table 3 for water quality standards. *E. coli* removal can occur by attachment of the bacteria to suspended algae or sediment and subsequent sinking of the newly formed aggregate (Ansa et al., 2011).

Once deposited in the environment, survival of these bacteria depend on a variety of factors. Such factors include temperature, solar radiation, turbidity, salinity, tidal versus non-tidal water bodies, deposition with suspended sediment, and competition with other microorganisms (Whitman et al., 2004).

E. coli

E. coli is a nearly ubiquitous microbe and type of fecal coliform bacteria (Swistock et al., 2006). Certain strains of *E. coli* can be pathogenic, however not all *E. coli* are pathogenic (Katouli, 2010; Leimbach et al., 2013). Non-pathogenic *E. coli* are naturally found in the gastrointestinal tracts of both humans and many animals (Heymann, 2015). However, certain strains of *E. coli* are pathogenic and can cause severe, self-limiting gastrointestinal distress (Heymann, 2015). These pathogenic *E. coli* are typically acquired via fecal-oral routes from environmental sources (Gorham & Lee, 2015). *E. coli* O157:H7 and O111 are common recreational water pathogens, causing 33% (7/20) of outbreaks between 2011 and 2012 (Hlavsa et al., 2015). As numerous other microbes are enteric pathogens that also cause gastrointestinal distress of varying degrees of severity and often have fecal sources, the near ubiquity of *E. coli* in feces suggests that it is a reasonable indicator organism (Moe, Sobsey, Samsa, & Mesolo, 1991). Simple methods for detection of an indicator organism such as *E. coli* or fecal coliforms in general suggests that it will be a reliable indicator of other waterborne enteric pathogens (Bain et al., 2014; K. Levy et al., 2012; Moriarty et al., 2011).

In a study of five lakes, (Francy et al., 2006) (Erie) found variation in the strength of correlations between *E. coli* and a number of environmental or water quality variables including rain with a 1-day lag, log-turbidity, turbidity, water temperature, water depth (Francy et al., 2006).

Conductivity

Conductivity is a measure of aqueous ions able to conduct electrical current, essentially a measure of salinity (Lundstrom & Fundingsland). This measure is related to other measures of ions such as total dissolved solids, hardness, alkalinity, and pH (Lundstrom & Fundingsland). Water of “good” quality has a low conductivity, on the order of 200 micro-Siemens per centimeter (µS/cm) or less (*Interpretation of Water Results: Standard Water Analysis (SWA),*, n.d.) whereas conductivity for poor quality water, highly saline water, is on the order of more than 50,000 µS/cm (Queensland Health Forensic and Science Services, n.d.). In the study by Psilovikos 2006 of a deltaic lake, low conductivity values of conductivity, on the order of 100-300 µS/cm, was associated with high water levels even when dissolved material that contributes to conductivity is expected to be high (Psilovikos et al., 2006).

Dissolved Oxygen

Dissolved oxygen is one indicator used by the EPA in the most recent 2012 National Lakes Assessment (NLA) survey that monitors environmental stressors and biologic condition of lakes across the United States (EPA, 2015). Oxygen dissolves into lake water from the air or is released into the water during photosynthesis by algae, for example (Lock, 1914). Dissolved oxygen is necessary for aerobic life, including algae and fish (Davidson, 2014). Organic material such as decaying plant matter or fecal wastes reduce levels of dissolved oxygen (Davidson, 2014). Van Buuren and Hobma 1991 found that dissolved oxygen concentrations greater than 0.5 milligrams per liter (mg/L) can help remove fecal bacteria (as cited in Ansa et al., 2011). Poor aeration in deep areas of the lake results in less dissolved oxygen (Swistock et al., 2006).

Insolation

Solar radiation can be measured as total insolation or reduced to just ultraviolet (UV) radiation. Whitman 2004 found that while associated UV-radiation was a poor proxy for total insolation, likely because much UV-radiation is scattered before it reaches bacteria in the water. Insolation influences inactivation of bacteria such as *E. coli* in freshwater bodies (Whitman et al., 2004). In addition to DNA damage, solar inactivation mechanisms are thought to include photo-oxidation of cells, a process enhanced by the presence of oxygen and organic matter in the water (Whitman et al., 2004). The effect of insolation is itself influenced by local weather factors including cloudiness, which reduces inhibits solar inactivation (Whitman et al., 2004). To provide perspective on insolation measurements, a National Renewable Energy Laboratory (NREL) National Solar Radiation Database (NSRD) summarized insolation in Atlanta, Georgia between 1961 and 1990 (Marion & Wilcox, 1994). The ranges NREL found were roughly: 3-5 kWh/m^2/day from November through January, 3.5 – 6 kWh/m^2/day in March, 4-6.6 kWh/m^2/day in April, 5 – 7 kilowatt-hours per square meter per day (kWh/m^2/day) in May through August, and 4-6.5 kWh/m^2/day in October and November (Marion & Wilcox, 1994).

Sunlight can inactivate fecal coliforms, however water depth is a confounding factor due to sunlight penetration (Sinton, Hall, Lynch, & Davies-Colley, 2002). Possible confounders of the relationship between insolation and *E. coli* concentration include turbidity, lake level, and wave height, three factors associated with wind (Whitman et al., 2004).

pH

Photosynthesis and respiration cause lake pH to cycle on a daily basis (Ansa et al., 2011; Lock, 1914). At night the lake pH decreases as respiration releases carbon dioxide into the water creating carbonic acid (Ansa et al., 2011; Lock, 1914). pH rises again during the day as algae consume carbon dioxide reducing the carbonic acid and acidity of the lake (Lock, 1914). pH typically fluctuates between 6.5 and 9 (Lock, 1914). pH values outside that range stress fish populations and can be fatal (Lock, 1914). Awuah, 2006 found that variation in pH, such as the daily fluctuation, inhibits survival of *E. coli* (as cited in Ansa et al., 2011), similarly Maynard et al., 1999 found high pH levels can remove fecal bacteria (as cited in Ansa et al., 2011). Limestone can be used to raise pH of ponds (Swistock et al., 2006).

Temperature

Temperature is an important parameter in bacterial growth and survival. Inactivation of *E. coli* and other enteric bacteria is dependent on temperature (Pachepsky et al., 2014). Air and water temperatures are two environmental factors often considered for predictive models of *E. coli* concentration (Olyphant & Whitman, 2004). As temperature increases, inactivation generally increases (Pachepsky et al., 2014). Insolation is a confounding factor that influences both temperature and bacteria survival (Pachepsky et al., 2014). Water temperature is expected to vary with season (Sinton et al., 2002). Olyphant et al. 2003 found that water temperature decreased in a stream during a storm and increased as stream discharge decreased to normal (Olyphant et al., 2003). McQuestin et al. 2009 found temperature to be a key variable in *E. coli* inactivation (McQuestin et al., 2009).

Transparency and Turbidity

Transparency and turbidity are contrasting measures of suspended particulates in the water. Transparency indicates the visual clarity of the water (Swistock et al., 2006). A Secchi disk is a measurement tool immersed in the water until its contrasting lines are no longer visible; it is a measure of clarity rather than turbidity (Swistock et al., 2006). The Secchi disk is black and white circle is lowered into the water until the contrasting sections are barely visible and the depth recorded; the greater the visible depth, the less turbid the water (Swistock et al., 2006). Turbidity is measured in nephelometric turbidity units (NTU) using a turbidimeter (Francy et al., 2013). Turbidity is mostly an aesthetic problem (Swistock et al., 2006). Runoff, bottom-dwelling wildlife, and large zooplankton populations can all make water turbid (Swistock et al., 2006). To distinguish between sediment and zooplankton, hold a glass of water up to the light if the particles move erratically, they are zooplankton (Swistock et al., 2006). Heavy loads of suspended sediment can be harmful to fish by irritating gills and reducing oxygen production (Lock, 1914). An experiment by Unckless and Makarewicz, 2007 showed mixing of bottom sediment into the water column incorporated settled goose feces and nutrients into the water. Turbidity and log-turbidity were positively and significantly related to *E. coli* concentration (Francy et al., 2006).

Exposure

Fecal-oral transmission of is the primary exposure route for waterborne pathogens (Gorham & Lee, 2015). Because swimmers are assumed to have the highest exposure due to full body contact with the water and potential for ingestion, the recreational water standards for swimmers are used as a worst-case scenario for risk assessment EPA, 2011. Recreational waters, by definition, are separated into treated sources, such as swimming pools, spas, and waterparks, and untreated surface waters further divided into freshwater and marine sources (Yoder et al., 2004). This study will focus on untreated recreational freshwater sources. Distribution of infectious microbes can have both public health and economic effects if human exposure, such as through drinking water and/or recreation is leads to an infectious dose (Swallow et al., , 2010).

Water Quality and Dogs

The risk associated with pet dogs drinking from park ponds and lakes is a frequently-asked question that few studies address directly (Unidentified Recreators at Lake Avondale, personal communication, February 28, 2016). Literature suggests dogs may become ill from ingesting some of the same water contaminants as cause human illness. Risk of such illnesses is heightened for puppies. For example, dogs can be affected by *Campylobacter* and cyanotoxins ("Overview of Enteric Campylobacteriosis," 2015; Washington Department of Health, 2014).

Lake Management

Factors of Importance

A basic understanding of the watershed can provide context for understanding the flow of nutrients, the key concept in managing a lake ecosystem (Connolly, 1990). Nutrients and contaminants vary with land use across the watershed, for example livestock, fertilizer application, and septic systems all introduce nutrients to the watershed (Connolly, 1990). The first wave of runoff during a rainstorm is a source of nutrients, especially phosphorus (Connolly, 1990). The term integrated pond management (IPM) refers to the method of managing a pond based on the ecosystem rather than only individual factors (Connolly, 1990).

Natural characteristics of the water body and its watershed that can affect water quality include source of water, types of rock, types of soil, and the rate water moves in and out (Connolly, 1990; Swistock et al., 2006). From a management standpoint, humans have little influence on these factors, however human activity near the pond often correlates with water quality problems (Swistock et al., 2006).

The primary uses of the lake will determine which water quality parameters to focus on (Connolly, 1990; Swistock et al., 2006). Important parameters to consider are fecal coliform bacteria, *E. coli*, parasites, blue-green algae, turbidity, water temperature, pH, dissolved oxygen, metals (copper, iron, manganese), nutrients (phosphorus, nitrate-nitrogen, ammonia-nitrogen), and chemicals (pesticides, aquatic herbicides) (Swistock et al., 2006). Key parameters for lakes used primarily for fishing, irrigation, or aesthetic beauty as primary lake uses (see Appendix F).

Nutrients

Nitrogen, phosphorus, and potassium are nutrients essential to growth of plants and algae (Connolly, 1990; Swistock et al., 2006). Nitrogen and phosphorus are the two primary nutrients of concern with phosphorus often the limiting nutrient (Connolly, 1990). Sources of nitrogen and phosphorus include direct application, runoff including fertilizers, runoff including animal waste, and septic systems (Swistock et al., 2006). Increasing levels of nutrients can lead to excess nutrients, or a eutrophic lake, stimulating aquatic plant growth and reducing dissolved oxygen levels out of balance with biological oxygen demand (see Appendix B) (Connolly, 1990; "Eutrophication,"). It should be noted that abundant aquatic plants alone do not signify eutrophication (Connolly, 1990).

Nitrogen in ponds is usually found as ammonia or nitrate (Swistock et al., 2006). Human and animal waste is the predominant source of ammonia, which is toxic to aquatic life, particularly at ammonia-nitrogen concentrations greater than 0.1 mg/L (Swistock et al., 2006). Nitrate-nitrogen concentrations are considered excessive when the concentration exceeds 3 mg/L (Swistock et al., 2006).

Phosphorus in ponds is usually found as phosphate (Swistock et al., 2006). Only a small amount of phosphate, approximately 0.01 mg/L (10 parts per billion, ppb), is needed to stimulate plant and algae growth (Connolly, 1990; Swistock et al., 2006). Should a lake become eutrophic, aluminum sulphate (alum) can be applied to bind with phosphorus rendering it chemically unavailable and removing it from the water column as a precipitate (Connolly, 1990). Phosphorus is chemically available from some sediments, however less phosphorus is released into oxygenated waters, emphasizing the role of aeration in control of phosphorus concentrations (Connolly, 1990).

Pesticides

Pesticides, particularly insecticides, applied around the pond can affect water quality especially when windy or when followed by heavy rain however the concentrations dissipate fairly rapidly (Swistock et al., 2006). For this reason it is recommended that application of pesticide within 50 feet of a water body be done with a drop spreader (Connolly, 1990). Copper-based algaecides can build high concentrations of copper in ponds after repeated use (see section on Metals) (Swistock et al., 2006).

Metals

High concentrations of iron can result in orange precipitate (Swistock et al., 2006). A metallic taste can result from iron concentrations greater than 0.3 mg/L, manganese concentrations greater than 0.05 mg/L, or copper concentrations greater than 1.0 mg/L (Swistock et al., 2006). When water is used for irrigation, these same iron and manganese concentrations may harm plants (Kronlein et al., 2014; Swistock et al., 2006).

Buffer Region

Buffer strips around ponds are vegetated areas that help filter out pollutants (Connolly, 1990). A buffer strip of vegetation around the pond, such as four to ten feet of unmowed grass is recommended on a gentle slope (Swistock et al., 2006). While trees can minimize heat reaching the pond, they can also block wind and inhibit aeration of the pond (Connolly, 1990). Inflow streams, outflow streams, and drainage ditches should not be straight channels but rather meander to reduce velocity and thus the amount of sediment deposited in the pond (Connolly, 1990). Additional methods for reducing velocity include dams, weirs, sediment ponds (Connolly, 1990).

Color

Related to turbidity, transparency, and microorganisms is pond color. Plankton populations along with suspended sediments and organic matter determine the pond color (Lock, 1914). Abundant planktonic algae population produces a green tint to the pond (Lock, 1914). Rain can wash sediments into the pond turning it a brown color that disappears over a few days as the sediment settles to the bottom (Lock, 1914).

Algae and Fish

When lake chlorophyll concentrations greater than 13 g/L the lake is considered eutrophic (Ansa et al., 2011). Algal blooms generally appear after prolonged periods of hot weather (Swistock et al., 2006). Some blue-green algae are harmful if used to water livestock, however the long strands or mats formed by filamentous algae are not harmful to animals (Swistock et al., 2006). Reducing phosphorus levels in the water is a recommended strategy to reduce occurrence of algal blooms (Connolly, 1990).

In freshwater, cyanobacteria is a greater concern than algae as cyanobacteria are more likely to form blooms, dense surface mats, with concentrations of toxins great enough to be of concern (WHO, 2003). Cyanotoxins are produced by dense populations of cyanobacteria which can be ingested and in some cases inhaled (WHO, 2003). Reporting of algal bloom-associated intoxication, such as from cyanobacterial toxins, is expected to improve as CDC is expanding NORS to study it (Hlavsa et al., 2015).

Some algae is good for fish production and the life cycle of the pond (Lock, 1914). However, high concentrations of pesticides, including aquatic herbicides, in water can be detrimental to fish populations (Swistock et al., 2006). Rapid die-off of microalgae is most common after use of aquatic herbicides to prevent excessive growth or during hot, dry weather (Swistock et al., 2006). Such rapid die-off of the microalgae can consume dissolved oxygen as the algae decomposes thus causing stress, disease, or suffocation of fish (Lock, 1914; Swistock et al., 2006). Mechanical aeration can also reintroduce needed oxygen to the pond (Lock, 1914; Swistock et al., 2006). In general, fish ponds have a pH near 7 and negative effects are apparent below pH of 6.0 (Swistock et al., 2006).

Canada Geese and Other Waterfowl

A variety of waterfowl including Canada geese (*Branta canadensis*), ducks, black swans, cranes, and gulls are known carriers of both indicator organisms and pathogens which are present in their feces (Moriarty et al., 2011; Pacha et al., 1988). Geese present concerns to freshwater including increased abundance of pathogens and excess nutrients (as cited in Unckless & Makarewicz, 2007). According to Pettigrew 1998, excess nutrients due to low nitrogen-phosphorus ratios in goose feces affect the lake ecosystem, in particular by encouraging blooms of cyanobacteria rather than a diverse phytoplankton population (as cited in Unckless & Makarewicz, 2007). Human pathogens carried by waterfowl include the bacteria *Campylobacter jejuni* (Rutledge et al., 2013), the protozoan *Giardia lamblia* (Graczyk et al., 1998), and viruses such as avian influenza (CDC, 2015a).

Multiple studies conclude that distribution of human pathogens by Canada geese is a legitimate public health concern (Feare et al., 1999; Graczyk et al., 1998; Zhou et al., 2004). The fecal contributions of Canada geese (*Branta canadensis*) to water quality have been the focus of numerous studies (Bedard & Gauthier, 1986; Connolly, 1990; Swallow et al., 2010; Unckless & Makarewicz, 2007). Research suggests that feces deposited directly into the water settles quickly and is detected in water quality measurements only if the sediment is mixed into the water column either through wind and wave action or benthic activity.

While environments and size of Canada goose flocks vary, each goose produces a large quantity of feces which can influence water quality. Defecation rates of geese can vary significantly with food source as determined by the habitat (Bedard & Gauthier, 1986). Data collection methods for goose defecation rates are often biased or imprecise and recommended data collection methods vary for large, compact flocks of feeding geese compared to small flocks or family groups (Bedard & Gauthier, 1986). it should be noted that the geese were observed in three habitats with different food sources, the number of observations varied for each data collection event, and several methods of data collection were tested at each habitat (Bedard & Gauthier, 1986). With the limitations in mind, estimated mean hourly defecation rates in a flock of greater snow geese (*Chen caerulescens atlantica* {Linnaeus}) in the Quebec, Canada were 12.23 +/- 0.15 defecation events per hour (Bedard & Gauthier, 1986). This rate is at the upper end of the range estimated for Canada geese. Canada geese can defecate between 28 to 92 times per day, for a healthy adult goose that totals 0.5 to 3 pounds of feces (wet weight) per day (French & Parkhurst, 2009; Hussong et al., 1979; Swallow et al., 2010) a high fecal output regardless of which estimate is used. Given the large quantity of feces per goose and that most Canada goose droppings are most likely to be found within 10 meters (33 feet) of the water study of environmental transmission of pathogens from geese to humans is warranted (Feare et al., 1999). Gorham and Lee, 2015 speculate that goose feces create a potential risk for children playing or families picnicking in the beach sand through either direct or indirect contact with fecal matter as pathogens elute from feces (Gorham & Lee, 2015). This potential exposure is supported by the findings of Whitman and Nevers, 2003 which determined beach sand adjacent to a lake to be a reservoir for *E. coli* (Whitman & Nevers, 2003).

Canada geese also contribute nutrients to the lake in the forms of nitrogen and phosphorus. On a yearly basis, an individual Canada goose can excrete 521 g to 1,410 g (1.15 - 3.11 lbs) of Kjeldahl nitrogen and 163 g to 638 g (0.36 - 1.41 lbs) of phosphorus (Swallow et al., 2010). On a monthly basis, an individual goose can excrete 50 g of phosphorus (Connolly, 1990).

In addition to the nutrient load, studies found Canada geese (*Branta Canadensis*) can carry several human pathogens including the bacteria *E. coli* (Fallacara et al., 2001; Swallow et al., 2010)*, Salmonella spp., Streptococcus spp.* (Swallow et al., 2010), *Listeria* spp. (Converse , 1999), and frequently

Campylobacter (Fallacara et al., 2001; Kassa, Harrington, & Bisesi, 2004; Moriarty et al., 2011; Pacha et al., 1988; Rutledge et al., 2013). Protozoa have been isolated from Canada goose feces as well, including *Giardia* sp. (Graczyk , 1998; Converse , 1999; Kassa , 2001) and multiple genotypes of the protozoan *Cryptosporidium* (Jellison et al., 2009; Graczyk , 1998; Kassa , 2001; Zhou , 2004; Moriarty et al. , 2011). Evidence suggests *Campylobacter* spp. is more commonly found in goose feces than *Cryptosporidium* (Fallacara et al., 2001; Heitman et al., 2002; Moriarty et al., 2011; Ogden et al., 2009; Sterk et al., 2016; Zhou et al., 2004). However, several studies suggest that when present viable *Cryptosporidium parvum*, can pass through goose feces to contaminate water then infect humans who have contact with that water (Dietz & Roberts, 2000; Graczyk et al., 1997; Graczyk et al., 1998).

Precipitation

Precipitation is a key factor in assessing water quality, particularly given the transience of contaminants (Francy et al., 2013; Nevers & Whitman, 2005). Precipitation may effectively dilute contaminant concentrations (Wu et al., 2014). Modeling can be used to evaluate how precipitation relates to run-off into the watershed (Laenen & Risley, 1997).

Upper Ocmulgee River Watershed

Water quality is a concern throughout watershed as upstream water has the potential to affect water quality downstream. Lakes and ponds are an integral part of the watershed as a whole. Water upstream can affect water quality downstream, (Ferguson et al., 2003; Olyphant et al., 2003; USGS, 2002), which can include drinking water intakes (Tornevi et al., 2014) or recreational areas. Lake Avondale drains into Cobbs Creek which runs into South River which then winds south to Jackson Lake (Georgia Adopt-A-Stream, 2014).

Bess Walker Park, Avondale Estates

Bess Walker Park is a park in the City of Avondale Estates in DeKalb County, Georgia. It is a city of 1.23 square miles home to about 3,360 residents (Adopt-A-Stream, 2011-2012; Griffin, 1983). The park is open to a variety of events and recreational activities including dog-walking and fishing in Lake Avondale (The Lake House at Avondale, 2015; Warren, 2015).

Lake Management in Bess Walker Park

Since 2004, the Lake Avondale Advisory Board (LAAB), committee of community members, has managed the lake (C. Warren, personal communication, November 19, 2015; The Lake House at Avondale, 2015). The LAAB has addressed a wide range of issues including waterfowl populations, fishing, park vegetation, and drainage around the park, runoff, and lake dredging (City of Avondale Estates, n.d.; C. Warren, personal communication, November 19, 2015). LAAB also approves local residents interested in conducting school or citizen science projects such as measuring water quality and maintaining native vegetation (C. Warren, personal communication, November 19, 2015). .

Several years ago the lake water level was lowered in order to install gabions as bank support to prohibit erosion (O. Griffin, personal communication, January 22, 2016). At least one worker commented on the bad smell emanating from the lowered lake (O. Griffin, personal communication, January 22, 2016).

Park representatives estimate that dredging the lake occurs every 20 years or so and the lake is due, if not overdue for dredging (O. Griffin, personal communication January 22, 2016). Some residents who recall draining the lake also recall an unpleasant smell (O. Griffin, personal communication, January 22, 2016).

A couple years ago, higher curbs were put in to help prevent road runoff from reaching the lake and direct more water flow to the storm drains at either end of the lake. The storm drains were recently widened to accommodate a greater volume of water. However, some water from the roads still makes it to the lake.

In landscaped areas such as Bess Walker Park, human manipulation of the environment to control road runoff, bank erosion, or sediment load, to name a few examples, can also influence the lake ecosystem and therefore water quality (Lock, 1914).

Though swimming and boating are currently prohibited, some adults recall swimmers in the lake during their childhood (C. Warren, personal communication, November 19, 2015; The Lake House at Avondale, 2015).

Fauna of Bess Walker Park

Fish in the lake include catfish, carp, and bass. People are permitted to keep any fish caught from the lake. Turtles live in the lake. Ducks and Canada geese visit the lake. Occasionally a heron, hawk, deer, or weasel is seen. (O. Griffin, personal communication, January 22, 2016)

Geese of Bess Walker Park

Geese have been visiting Bess Walker Park for many years. Geese are typically removed once per year in March or April when they fly in from the east for the day to eat the young grass. However, in 2015 the geese appeared sporadically throughout the spring preventing the coordination of their removal and relocation. The geese are tagged to track their potential return, but to the best of the City’s knowledge none of the relocated geese have returned to Lake Avondale. Ten to thirty geese are relocated during the annual removal, though smaller numbers of geese, six or seven, may be seen at the lake during other seasons. Signs around the lake prohibit feeding geese, however residents often still feed the ducks which in effect, also ends up feeding the geese. (O. Griffin, personal communication, January 22, 2016). Ducks and geese are regularly found at the pond, however Canada geese are of particular concern due to their feces.

Canada geese are typically removed from the park on an annual basis. When the geese become a nuisance, the city Park Manager calls the United States Department of Agriculture Animal and Plant Health Inspection Service Wildlife Services (USDA APHIS WS) to have the geese removed from the park and relocated. Over the past ten years, since 2006, anywhere from 0 to 19 geese have been relocated from the park (E. Miller, personal communication, March 11, 2016).

Pesticides and Bess Walker Park

Pesticides are applied to the lake lawn by an external company as needed. A sulfur-free, water-friendly pesticide is used on the lawn of Bess Walker Park. No pesticide is applied within ten feet of the lake. Specific pesticides used on the lawns of Bess Walker Park include Ortho Weed-B-Gon Pro, Halts Pro 65 WDG (Prodiamine), and 17-0-5 Professional Fertilizer. (O. Griffin, personal communication, January 22, 2016). Ortho Weed-B-Gon Pro is an alkaline pesticide, soluble in water ("Materials Safety Data Sheet: Ortho Weed-B-Gon Pro,"). The three primary components, 2,4-D, Mecoprop-p (MCPP-p), dicamba, and dimethylamine salt dissociate rapidly in the environment, on the order of days to weeks ("Materials Safety Data Sheet: Ortho Weed-B-Gon Pro,"). Testing on fish and mallard ducks found these three components toxic to both species on the order of 100 mg/L and 4,000-10,000 mg/L, respectively ("Materials Safety Data Sheet: Ortho Weed-B-Gon Pro,"). Effects of prodiamine were also assessed in fish, Mallard ducks, and invertebrates ("Materials Safety Data Sheet: Prodiamine,"). Doses of at least 0.55 mg/L over four days were toxic to fish and invertebrates, while doses of 10,000 mg/L were toxic to ducks over 8 days ("Materials Safety Data Sheet: Prodiamine,"). While also an alkaline pesticide, unlike Ortho Weed-B-Gon Pro, prodiamine is insoluble and stable in water ("Materials Safety Data Sheet: Prodiamine,"). However, prodiamine will sink over 24-hours where it persists in the soil but remains immobile and does not bioaccumulate ("Materials Safety Data Sheet: Prodiamine,").

Maintenance included treatment of algae and exotic, invasive weeds, which can deplete dissolved oxygen required by fish and present aesthetic issues, are controlled using EPA-approved algaecides and herbicides. (J. Brown, personal communication, February 3 2016; J. Brown, personal communication, February 8, 2016).

Results of Studies Investigating the Problem – Interrelationships Between Variables

Numerous environmental factors have been shown to influence pathogen survival. Once evaluated, these factors can be used in statistical models used to associate environmental and water quality variables in order to predict the presence of disease-causing organisms (Francy, Stelzer, Duris, et al., 2013; Nevers & Whitman, 2005). In a study of *E. coli* in public beaches and recreational lakes in Ohio, Francy et al., 2013 determined the best predictors of *E. coli* concentrations were rainfall, turbidity, water temperature, wind direction, and wind speed (Francy, Stelzer, Duris, et al., 2013). Nevers and Whitman, 2005 also developed a regression model for *E. coli* in recreational beaches including the variables precipitation, turbidity, wind, wave height, wave period, and chlorophyll (Nevers & Whitman, 2005). While not always significantly correlated with *E. coli,* turbidity and water temperature did have a positive relationship with *E. coli* concentration and were included in most models used by Francy et al., 2013. Key factors found by Ferguson et al., 2003 were temperature, sunlight, pH, nutrients, and water potential (related to moisture content in soils) (Ferguson et al., 2003). Air temperature was also a factor in a study of pathogen persistence in feces deposited on pasture land (Moriarty, Weaver, Sinton, & Gilpin, 2012).

Rainfall is a commonly studied factor relating to freshwater quality and also outbreaks of waterborne disease (Sterk et al., 2016; Tornevi et al., 2014). Heavy rainfall was a stronger predictor of fecal contamination than turbidity as the heavy rain attenuated the effect of turbidity on bacteria concentrations (Tornevi et al., 2014). Tornevi , 2014 found rainfall is exponentially associated with bacteria concentrations (Tornevi et al., 2014). Generally, days of wet weather were associated with poorer water quality regardless of season (Tornevi et al., 2014).

A time component appears repeatedly when discussing measurements of water quality. Olyphant and Whitman 2004 found a slight relationship (r=0.41) between morning and afternoon measures of *E. coli* at a Chicago beach. The findings of Olyphant and Whitman 2004 suggest temporal variability in *E. coli* concentrations. Jensen 2003 also included season measured by calendar month as a confounder for water temperature (Jensen et al., 2003).

Risk Assessment

Completion of an exposure pathway transmitting a hazardous water contaminant to a person at a high enough dose to cause illness is necessary to determine immediate risk (Gorham & Lee, 2015). Ishii , 2014 used quantitative microbial risk assessment (QMRA) to evaluate multiple pathogens in a small irrigation lake frequented by geese as an alternative to fecal indicators like the *E. coli* used in this study (Ishii et al., 2014; Sales-Ortells & Medema, 2014). Several factors influenced pathogen concentrations. While no significant correlation was found between number of geese and pathogen concentration, pathogen concentrations were generally higher when geese were present (Ishii et al., 2014). QMRA results suggest that when used to irrigate fresh vegetables, the vegetables would have potential to transmit illness to consumers particularly during spring and fall when pathogen concentrations were highest (Ishii et al., 2014). Risk was affected by pathogen persistence in the environment which was dependent on environmental factors such as temperature, and sunlight (Ishii et al., 2014). The authors suggest multipathogen quantification is a more reliable method of risk assessment than using fecal-indicator bacteria such as *E. coli* (Ishii et al., 2014). Sales-Ortells and Medema 2014 conducted risk assessment of recreational activities including accidental ingestion of water droplets and aerosols, as well as hand-to-mouth contact while fishing (Sales-Ortells & Medema, 2014). These authors note the importance of site-specific pathogen data and frequency of exposure (Sales-Ortells & Medema, 2014).

Current problem and relevance to target population

Many of these parameters have been measured in Lake Avondale. However, measurements were collected on an inconsistent basis by multiple groups. While evaluated for short term lake management decisions or school projects, the datasets have yet to be compiled and analyzed for trends and the possibility of predicting water quality. Given the community of Avondale Estates has expressed concerns regarding lake water quality and public health, improving the surveillance and predictive capabilities of LAAB while providing a stronger knowledge basis for the broader community is an opportunity to assuage those concerns.

Chapter III: Manuscript

Abstract

INTRODUCTION Current research efforts focus on contamination in water bodies with obvious risk of exposure and economic impact, such as larger lakes with beaches. Improved surveillance methods increasingly link small scale outbreaks to recreational freshwater. This linkage has implications for lake management. A small neighborhood lake in suburban Georgia is the focus of this investigation into environmental predictors of Escherichia coli (*E. coli*). The study will address four aims: 1) determine if *E. coli* concentrations exceed recommended standards for recreational water, 2) assess correlations between variables to determine likely predictors of *E. coli* levels 3) develop a predictive model that may be used to 4) inform future lake management decisions.

METHODS Data was compiled from publicly available sources including Georgia Adopt-A-Stream, National Oceanographic and Atmospheric Administration Climate Data Online, National Aeronautics and Space Administration Earth Observatory, and community-hired contractor lake service reports. *E. coli* counts were collected from three sites: the inflowing creek, the outflow edge of the lake, and the outflow creek. Associations between environmental factors were assessed by Pearson and Spearman correlations to determine parameters for a predictive model for *E. coli* in Lake Avondale. Environmental variables considered include air temperature, conductivity, dissolved oxygen (DO), fecal coliform, insolation, month, pH, season, sampling site, and water temperature. Multivariate linear regression and logistic regression models were developed to investigate the relationships of environmental factors with the dependent variable, *E. coli* concentration and presence, respectively.

FINDINGS Multivariate linear regression found conductivity, log-transformed precipitation, and sampling site predict log-transformed *E. coli*. Investigation of interaction terms resulted in an over-specified model of *E. coli* concentration by conductivity, sampling site, insolation, pH, log-precipitation, water temperature, dissolved oxygen, season, interaction between dissolved oxygen and season. Logistic regression found conductivity and dissolved oxygen predict *E. coli* presence. Limitations include small sample size and convenience sampling at irregular intervals across space and time. Results suggest key environmental factors such as precipitation, conductivity, and dissolved oxygen be considered or controlled during lake management decisions. For a community actively engaged in the management of a local lake concerns regarding water quality are valid. Extending that concern to questions of health risks while perhaps not a dire concern is a reasonable discussion to promote evidence-based lake management.

Introduction

Epidemiology and surveillance of recreational water-associated disease outbreaks began in earnest in 1978 with expansion of the existing United States' waterborne-disease surveillance system (Yoder et al., 2004). Recreational water-associated outbreaks consist of at least two people with an illness linked to a contaminant in recreational water either by location or time of exposure (Hlavsa et al., 2015). Since the early 1990s the number of reported outbreaks from has increased (Hlavsa et al., 2015; Kramer et al., 1996; Calderon, & Juranek, 1996; Yoder et al., 2004). A recent assessment for 2011-2012, found 20 of 90 outbreaks were linked to untreated recreational freshwater (Hlavsa et al., 2015) and at least 479 cases of illness due to all recreational water (Hlavsa et al., 2015). Reported numbers are likely under-estimates as outbreak detection, investigation, and incidence reporting are all limited by missing data (Hlavsa et al., 2015). Surveillance factors affecting recorded case counts include: untreated and therefore undocumented symptoms, popularity of a recreation area (e.g. residential lake versus public beach), accuracy of contaminant detection, and compilation of case reports in a common database (Hlavsa et al., 2015).

With growing public health awareness and improved surveillance methods, detection of smaller outbreaks is increasingly possible (Hlavsa et al., 2015). Small outbreaks, residential settings of exposure, and mild illness are three of many obstacles to detecting outbreaks (Hlavsa et al., 2015). Water quality assessment of small, untreated, recreational, freshwater lakes such as those located in urban or suburban environments is expanding these epidemiologic and environmental health studies beyond the public beach environment (Honkonen & Rantalainen, 2013; Melymuk et al., 2014; Melymuk et al., 2011; Nevers & Whitman, 2005; Wada et al., 2006; Young & Thackston, 1999). Recreational activities that do not involve full body contact with water such as wading and fishing can lead to low level exposure to microbial contaminants EPA, 2011. Studies of freshwater lake contamination show animal feces deposited along the shore can contribute human enteric pathogens to the water (Edge & Hill, 2007

Colford, 2007; Jellison et al., 2009). Such pathogens include *E. coli*, *Campylobacter*, and *Cryptosporidium* which have caused outbreaks in recreational freshwater (Hlavsa et al., 2015). Surface runoff, disturbed lake bottom sediments, and waterfowl are known to contribute these same bacteria to lake water (Fallacara et al., 2001; Honkonen & Rantalainen, 2013; Laenen & Risley, 1997; Melymuk et al., 2014; Moriarty et al., 2011; Olyphant et al., 2003 & Harper, 2003; Olyphant & Whitman, 2004; Selvakumar & Borst, 2006; Tornevi et al., 2014 2014; Unckless & Makarewicz, 2007; USGS, 2002). As an indicator organism and proxy for pathogens which are often difficult to measure or detect (Haack et al., 2009) *E. coli* is often chosen as the model for water quality and enteric pathogens. There is a need to develop simple methods for monitoring small neighborhood lakes in order to manage the lake in a way that minimizes adverse public health effects.

This study will focus on assessment of the water quality of untreated, recreational, freshwater lake located in a suburban community as modeled by potential environmental determinants of *E. coli*. This project is a response to community-initiated interest in understanding water quality to inform lake management decisions. Lake Avondale in Avondale Estates, Georgia is a 5.12 acre lake with an average depth of 7.60 feet (Brown & Slovisky, 2015a). Lake regulations include prohibition of swimming, boating, and feeding geese (C. Warren, personal communication, November 19. 2016). Ongoing management of Lake Avondale may include monitoring and surveillance for health outcomes associated with recreating at the lake and minimizing potential human exposure to pathogens. Methods for monitoring small, neighborhood lakes are necessary for effective, evidence-based management of the lake ecosystem with public health in mind. Outputs of this project as it will be a resource for the Lake Avondale Advisory Board (LAAB) in both managing Lake Avondale as well as responding to community questions about the lake and public health.

To effectively manage lake conditions and advise residents, there is a need to understand the factors that influence the microbial water quality of Lake Avondale. This investigation will use *E. coli* data as an indicator for enteric bacteria. The goal of this project, therefore, is to perform a pilot-scale analysis to evaluate whether various lake water quality and environmental factors are potential predictors of *E. coli* concentrations in Lake Avondale. The project will address this goal through four specific aims: 1) determine if *E. coli* levels have exceeded recommended standards for recreational waters, 2) assess possible correlations between environmental variables to determine likely predictors of *E. coli* levels in Lake Avondale, 3) attempt to develop a predictive model describing *E. coli* concentrations in Lake Avondale based on observed and literature-supported associations between *E. coli* and environmental variables, 4) provide recommendations for future studies and considerations when monitoring or managing neighborhood lakes.

Methods

Data Sources

Data on environmental variables was extracted from lake service reports provided by LAAB and supplemented with publicly-available data. Variables across all data sets included air temperature, altitude, conductivity, dissolved oxygen, debris in lake, *E. coli*, fecal coliforms, insolation, lake depth, pH, precipitation, presence of aquatic vegetation, sampling date, sampling group, sampling site, sediment thickness, transparency, water depth, water temperature, water quality index, weather, and wildlife (Table 1, Appendix G) . However, not all variables were available for each date or each sampling site.

Publicly available Georgia Adopt-A-Stream data was the source for the majority of variables. While accumulated data spanned from January 2010 to November 2015, observations for many variables were recorded both infrequently and at irregular intervals. Data for the primary outcome variable, *E. coli*, was recorded during five selected months (May, June, August, September, and November) in 2011 and one month (February) in 2012. While two groups in Avondale Estates are registered with Adopt-A-Stream, only one group, the LAAB group, measured *E. coli*. Consequently only AAS data from the LAAB group was included in analyses. Fecal coliforms were measured by 3M Petrifilm, but the method used specifically for *E. coli* is not mentioned (Georgia Adopt-A-Stream, 2014). Single measurements were taken for air and water temperature using a thermometer (Georgia Adopt-A-Stream, n.d.-b). Conductivity data was also collected as single measurements, rather than in duplicate, and was measured using a calibrated meter (Georgia Adopt-A-Stream, n.d.-b). Dissolved oxygen was measured by two duplicate samples for precision and analyzed via fixation with manganous sulfate, alkaline potassium iodide azide, and sulfuric acid then titrated with sodium thiosulfate (Georgia Adopt-A-Stream, n.d.-b). Using an octa-slide 2 viewer pH test, pH was measured by two samples with the second measurement within 0.25 standard pH units used to validate the previous measurement (Georgia Adopt-A-Stream, n.d.-b)}. Additional details regarding AAS sampling techniques at Lake Avondale were not available.

Since the site-specific AAS data did not include precipitation or insolation, daily data for these two variables was obtained separately from the nearest location possible covering the range of time over which AAS *E. coli* data was collected (Francy, Stelzer, Duris, et al., 2013). Daily precipitation data was acquired from National Oceanic and Atmospheric Administration Climate Data Online (NOAA CDO) data for a site in Decatur, Georgia, less than five miles away from Lake Avondale. Insolation data was acquired from the National Aeronautics and Space Administration (NASA) Earth Observations (NEO).

Data reported in Lake Avondale service reports was collected by one environmental consulting firm contracted by Avondale Estates. Data was collected by various pairs of service technicians. During a full water quality analysis, lake water samples were analyzed using a variety of testing equipment. Turbidity is measured in nephelometric turbidity units (NTU). Nutrients such as ammonia-nitrogen and nitrite-nitrogen as well as alkalinity and hardness using a Hach , LaMotte, or American Marine manufactured testing equipment. Hanna equipment is used to measure temperature, pH, and conductivity while dissolved oxygen is measured with equipment from American Marine. Clarity was measured by Secchi disk on each visit to a lake. Fecal coliform samples were stored on ice and taken to an accredited laboratory for analysis by membrane filtration of a 100mL sample. All water quality measurements included in service reports were analyzed in triplicate and averaged to produce the final result. (J. Brown, personal communication, February 8, 216; J. Brown, personal communication, April 19, 2016).

Data Analysis

First, univariate exploratory data analysis was conducted on all variables. Descriptive statistics were examined to assess the distribution of each variable (Table 1). Variables were evaluated for normality of the distribution by comparing visual inspection of the probability plot and histogram with skewness and kurtosis statistics. Precipitation and insolation, larger datasets from outside sources, were evaluated for both all dates obtained and for the reduced dataset corresponding to dates with *E. coli* data. All later analyses were restricted to dates with *E. coli* measurements (n=29). *E. coli* values were collapsed to make a binary variable for presence-absence analysis of *E. coli* presence where 0 CFU/100mL indicated absence and concentrations greater than 0 CFU/100mL indicated presence of *E. coli*, the dependent variable, and fecal coliform variables were log-transformed for analysis to meet parametric assumptions of a normal distribution and equality of variances (Francy et al., 2013; Zimmerman, 2006). To permit the natural log-transformation *E. coli* observations of 0 colony-forming units per 100 milliliters (CFU/100mL) were assigned a value of 0.05 CFU/mL prior to the calculation. Post-transformation, these assumptions were confirmed using the skewness and kurtosis statistics. These two statistics were evaluated alongside histograms to assess normality for the remaining environmental variables.

Second, likely associations were assessed between all variables. Bivariate correlations between all variables were conducted using parametric Pearson's r correlation and the non-parametric Spearman rank correlation analyses. Variables with normal distributions were evaluated using Pearson correlation.

Categorical variables and variables with skewed distributions were evaluated using Spearman correlation (Weiss, 2007). Methods for Pearson correlation were based on the methods of Francy et al. 2006, Francy et al. 2013, and McEgan et al. 2013 while Spearman correlations were based on Psilovikos et al. 2006. Separate bivariate correlations were performed for potential environmental predictors with log-transformed *E. coli*, the presence/absence of *E. coli* data, and the environmental variables with each other.

Simple linear regression was also performed to determine if any individual variables had a linear relationship with the primary outcome, log-transformed *E. coli* concentration. The linearity of each variable with respect to the log-transformed *E. coli* outcome variable was assessed based on review of p-values and visual observation of scatterplots. Other variables significantly correlated with each other were investigated for confounding and interference with *E. coli* Environmental variables significantly associated with *E. coli* concentration or presence were included as parameters in multivariable regression models.

Multivariate linear regression analysis was performed to create predictive models of *E. coli* based on the methods of Francy et al., 2006, Francy et al., 2013, and McEgan et al., 2013. The final model was used to determine environmental variables most likely to significantly influence concentrations of *E. coli* in Lake Avondale. All models were evaluated at 95% confidence ($\alpha = 0.05$) unless otherwise stated. Variables associated with *E. coli* were included for review of all possible linear regression models.

Using the variables determined to be associated with *E. coli* all possible linear regression models were reviewed. These models were evaluated to determine which to consider for the final model. Evaluation was based on the highest coefficient of determination, r^2 value most similar to the model including all relevant variables, and to the best Mallow's C_p value. Akaike information criterion (AIC) and Bayesian information criterion (BIC) were also assessed to make sure they were comparatively low relative to other models. Backward selection with a cut-point of □=0.1 was used to reduce the full 8-variable model to statistically significant variables (Hering et al., 2014; Nicolas-Chanoine et al., 2013). Selected models were then re-evaluated using both stepwise and backward selection of variables. A p-value of 0.1 was used as the condition for any parameter to stay in the backwards analysis. The same significance level of 0.1 was used for both entry and remaining in the stepwise analysis. Potential final models were selected based on r^2 value and p-value for the model. These models were evaluated for multicollinearity by assessing variance inflation (VIF). Models displaying multicollinearity were adjusted by selecting between multicollinear variables until the model no longer contained multicollinear variables. This selection was based on the quality of the data for the variables, the frequency that variables appeared in other models, and the predictive variables in existing literature. Models passing the test for multicollinearity were verified by testing for influential points or outliers using jackknife residuals, Cook’s D, and leverage values for model residuals. The normality of residuals was evaluated for the final model. Multivariate linear regression models for *E. coli* concentration were compared to logistic models of log-transformed *E. coli* presence-absence models.

A multivariable logisitic regression model was created to assess the association of environmental factors with the presence of *E. coli* based on procedures modified from McEgan et al., 2013. The outcome variable, *E. coli*, was transformed into a bivariate categorical variable indicating either *E. coli* present (greater than 0 CFU/100mL) or not present (equal to 0 CFU/100mL, or not detected). Independent environmental predictor variables were first screened in bivariate logistic regression with *E. coli* presence using exact procedures to determine which variables to include in the model. Variables with a p-value of 0.05 were considered for inclusion. Both backward and stepwise selection were used in the multivariate logistic regression model with a cut-point of $\alpha = 0.10$. Assessment of collinearity amongst variables by VIF followed by tests for interaction before selecting a final model. The resulting log odds were back-transformed to the most probable number of *E. coli*.

Analyses were performed using Microsoft Excel (version 15.0), SAS 9.4 (SAS Institute Inc., Cary, NC), SAS Power and Sample Size 13.1 (SAS Institute Inc., Cary, NC), and G*Power 3.0.10 (. Following the conclusion of the study, results were shared with the Lake Avondale Advisory Board (LAAB).

Results

Descriptive Results

Describing and calculating variables. Conductivity and pH were very slightly right-skewed but were considered normally-distributed during analysis (Table 1). *E. coli*, fecal coliforms, and precipitation were right skewed with probability plots that appear logarithmic. These three variables were log-transformed to normalize the distribution.

Observations were grouped in time and space. The majority of data comes from the year 2011. *E. coli* data were collected in 2011 and 2012 with the majority of measurements (23/29) collected in 2011 (Frequency results not shown). The majority of observations were also collected at the site located on the south bank of Lake Avondale above the dam.

For several variables there was insufficient variation between observations to create a density plot for that variable. One potential predictor, transparency, was removed from analysis due to an insufficient number of observations and limited variability. All five transparency observations corresponded to *E. coli* values of 0 CFU/100mL at the same site, the south bank of the lake above the dam.

Analysis of variance (ANOVA) found that while there are differences in *E. coli* concentrations between sites, *E. coli* did not vary significantly between the three sampling sites (Figure 1). However, conductivity, dissolved oxygen, and pH did vary significantly by site.

Environmental variables differed slightly between sites (Figure 2). Conductivity was highest at the downstream site and lowest at the lake site. Conductivity also consistently increased from February to

November. Dissolved oxygen increased from December to April to varying degrees between the sites. Water and air temperature showed similar trends, increasing from April to June 2011 then plateauing through the summer and decreasing throughout 2012. This trend looks slightly different downstream of the lake, however there was still an increase from spring into summer 2011 followed by a general decrease in temperature.

Compliance with recreational water regulations. The majority of samples were collected in the lake rather than the inflowing and outflowing streams (frequency analysis not shown here). The creeks had median *E. coli* counts above the EPA limit for recreational water, and the downstream sampling site was entirely outside that limit (Figure 1). *E. coli* concentrations were compared to concentrations stated in EPA and Georgia regulations on recreational waters (see Appendix C). While individual samples taken at a single point in time could be compared to regulations they do not address the per month component of some regulations. We were unable to calculate a geometric mean for each month to compare to regulations. Incompliant samples were found each year that AAS sampling took place (Table 2). Incompliant samples were found year round, however the majority of incompliant samples were in the spring and summer months (Table 3).

Across years with available data, in 2011, 50% (12/24) of samples were incompliant with the U.S. Environmental Protection Agency regulation for recreational water of 126 CFU/100mL in any of five samples collected during a 30-day period (Table 2). In 2015, 0 of 14 samples were out of compliance with this regulation on the dates and times samples were collected. Overall, there were a greater number of incompliant samples during spring and summer months compared to fall and winter (Table 3).

Correlation Results

The second aim of the study was to assess potential correlations between environmental variables to determine likely predictors of *E. coli* levels in Lake Avondale. We conducted bivariate correlations and compared Pearson's r results to Spearman's rank correlation results. Variables with significant correlation at 99% and 95% confidence were included, as were variables with meaningful correlations suggested by biological relationships or by existing literature. Pearson correlations evaluated the association between normally-distributed variables and both *E. coli* concentration and *E. coli* presence.

Separate bivariate comparisons of *E. coli* concentration and *E. coli* presence with each of the predictor variables showed positive and significant correlations with precipitation at the 95% confidence level and for some precipitation variables at the 99% confidence level (Table 4). A significant, positive, bivariate relationship between log-*E. coli* and conductivity, dissolved oxygen, pH, and precipitation. The only significant bivariate correlations with *E. coli* presence were conductivity, and dissolved oxygen (Table 4).

Correlations also exist between the environmental variables (Table 4). Dissolved oxygen is strongly associated with sampling site and time components (year, month, season) as well as air and water temperatures. Conductivity is also significantly associated with sampling site and time, as well as pH. In addition to dissolved oxygen, water temperature is significantly associated with pH, air temperature, insolation, and year.

Linear Regression Results

The third aim of the study was used to assess environmental predictors of *E. coli* concentrations in Lake Avondale. To address this aim, we applied both multivariate linear regression and logistic regression to the data. The variables found to have a linear relationship with *E. coli* concentration were fecal coliform and dissolved oxygen (Table 6). Assessment among environmental parameters found air temperature was associated with dissolved oxygen and water temperature (figure not included). Water temperature was also associated with insolation, pH, and year. Dissolved oxygen was also associated with month, season, and year. Insolation was associated with year and week day. Conductivity was associated with year and sampling site. Season was associated with precipitation.

The full model included eight variables selected based on significant correlations and absence of multicollinearity: conductivity, dissolved oxygen, pH, log-precipitation, air temperature, insolation, site, and season (Table6, Table 7). Due to one missing value for pH, the model used 28 of 29 observations. Using backward selection from the full model with a cut-off of 0.05 for staying in the model, the reduced version of the model included three variables –Conductivity, log-precipitation, and site (Table 6, Table 8). Step-wise selection was also attempted with a significance level of 0.05 for entry and staying in the model. This model produced only conductivity as a predictor of *E. coli* concentration.

Water temperature and month were not included in the models due to high variance inflation (VIF > 10) when included with air temperature and season, respectively indicating these variables predict the same outcome.

When validating the model, there were two dates identified as outliers at this significance level (α= 0.05) based on jackknife residuals greater than 1. When these two points were removed, bringing the sample size down to 26, the reduced model subsequently included conductivity, dissolved oxygen and log-precipitation (Table 6, Table 9).

Together conductivity, precipitation, and site explain 33% of the observed variation in *E. coli* concentrations in Lake Avondale with 95% confidence. Ideally, the study would have 95% confidence (α = 0.05) and 80% power (beta = 0.20). As power is especially influenced by sample size, the actual power achieved by this study is much lower than 80%. At 95% significance the power for the reduced model is 33% and 37% at 90% significance.

Investigation of interaction terms between environmental variables led to selection of one model (Table 6, Table 10). Parameters included in the model for backward selection were water temperature, insolation, precipitation, pH, conductivity, dissolved oxygen, season, and interaction between dissolved oxygen and season. The resulting reduced model was obtained through backward selection ($\alpha = 0.10$) and included the variables conductivity, precipitation, season, dissolved oxygen, and the interaction term. When run using backward selection with $\alpha = 0.05$, conductivity was the only predictor.

Logistic Regression Results

Of the 29 samples with *E. coli* measurements, ten observations had no detectable *E. coli*, 18 had *E. coli* present, and one observation was missing. Given the presence of *E. coli* at the sampling site, the results of logistic regression return conductivity (odds ratio = 1.12 (95% confidence interval (CI): 0.99-1.26, p-value = 0.085) and dissolved oxygen (odds ratio = 0.49 (95%CI: 0.21-1.15, p-value=0.100) as predictors of *E. coli* presence. While the logistic model is significant (p-value = 0.004), neither individual predictor is significant at the 95% significance level.

Discussion

There were several notable findings from this investigation that can direct future management of Lake Avondale. First, a greater percentage of samples were incompliant with EPA and Georgia standards for recreational water quality during spring and summer months. The variables included in at least one of the four models were conductivity, precipitation, dissolved oxygen, season, and sampling site. Conductivity appears in all four models, while precipitation and dissolved oxygen appear in three. Site and season each appear in one model.

Results suggest the lake may be within EPA guidelines for recreational water during most months, but these results must be interpreted in the context of the dataset. While a monthly or seasonal average may show that *E. coli* levels are below the regulations, the number of observations per month is not representative enough to capture day-to-day variations. Individual days may be out of compliance, but again due to the number of samples we were unable to determine the frequency of such occurrences. More samples per month facilitates calculation of a geometric mean for which there is the EPA *E. coli* guideline of 126 CFU/100mL averaged over five samples collected within 30 days (see Appendix C).

This study used regression analysis to define further the amount of variance in *E. coli* attributable to each variable. The adjusted R-square value, accounting for the number of variables in the model, fell within the range of 0.3 to 0.5 suggesting a moderate relationship between the environmental predictors and *E. coli*. However, the model with interaction terms and six predicting variables may over-specify the model given the small size of the dataset.

While statistical outliers were removed from the linear regression model in the interest of exploratory analysis, we believe they should remain in the model. While the two values may be anomalous for this data set, the dataset is small and the values are biologically plausible. In light of these uncertainties we are unable to decide on the best model for Lake Avondale. However, the models summarize currently available data and common predictors across all models supports the ability of conductivity, dissolved oxygen, and precipitation to inform LAAB on *E. coli* concentrations in Lake Avondale.

The predictive capabilities of any model depends on the quality of the data used to develop it (Francy & Darner, 2006). In this case, given the variety of sampling groups, it is possible that an inconsistent set of procedures was used to collect the data. A major limitation resulting from this small sample size is the absence of a validation data set. It was not possible to initially subset 10% of the data to validate the final or acquire comparable data from another lake in the Atlanta area to act as a validation set. Accuracy and precision of the model also limit the ability to draw causal conclusions based on results. Accuracy of the model may be limited by use of non-site-specific data for precipitation and insolation. This data was from routine monitoring at the city and national level, and was not site specific. Weather patterns can vary within the Atlanta metropolitan area where the study site is located so site-specific data is really more robust.

A low p-value implies but does not guarantee significance as sample size increases. It would be interesting to see if or how associations change in a more time-representative set an observations throughout the year as well as a 24-hour period. It could be that associations reflect when it was convenient for sample collectors to collect samples based on their daily schedules or the weather, for example.

Nevertheless the models suggest similar variables to the other models, returning conductivity, precipitation, and dissolved oxygen while suggesting there is a relationship between season and dissolved oxygen also found in the literature. Routine sampling of dissolved oxygen throughout the year could elucidate this relationship for Lake Avondale. Additional relationships between variables that may describe findings include a relationship between dissolved oxygen and organic material, specifically algae and aquatic vegetation (J. Brown, personal communication, February 8, 2016). Measurement points for data not collected at the sampling site, precipitation and insolation, were determined primarily by proximity (Francy, Stelzer, Duris, et al., 2013). The insolation data point was chosen by which coordinates were closest to the majority of sampling sites. Precipitation data was selected based on proximity to the sampling sites and correlation with rainfall documented at the sites. Several variables rejected from the analysis due to missing values were reported to be significant predictors in the literature; turbidity in particular is a commonly cited predictor of *E. coli* concentration (Francy, Stelzer, Duris, et al., 2013; Nevers & Whitman, 2005; Nevers, Whitman, Frick, & Ge, 2007; Olyphant & Whitman, 2004; Whitman et al., 2004). However, the corresponding AAS transparency variable in this study had too few observations (n=5) almost exclusively collected in the same month and year to perform most of the calculations.

From a lake management perspective, control measures that influence these two parameters could become a priority. The meter-measurement methods of conductivity and dissolved oxygen are much simpler than the culture method used to measure *E. coli.* Thus, conductivity and dissolved oxygen are potential proxies for *E. coli* concentrations that can be monitored by citizen scientists with relative ease,

Data collected is a convenience sample and not spatially or temporally representative of any variable. This convenience can refer, for example, to the ease of access to the sampling site, the sites chosen for sampling as well as the time of day, day of the week, or month that samples were collected. Differences were noted between the sampling sites suggesting that location-specific factors influence results and samples collected near one another are more similar than samples collected from farther away.

Time of sample collection is a factor weakly addressed by this study. This dataset is a convenience sample and not spatially or temporally representative of any variable. The absence of a meaningful time frame is a confounder. There may be yearly variation within a given variable but the time resolution of this dataset is not such that two years can be compared without potential effects of factors such as season or chance. Given the likely influence of season on *E. coli* concentrations, future models should consider further the interaction between dissolved oxygen and season. For the variable for year, the majority of observations were in one of the two years and heavily concentrated in one or two seasons. Such observations are not representative of a full year limiting the variation and therefore the descriptive capabilities of this variable. Aggregating months into seasons, achieved a more representative variable with a time component that could capture some of the many environmental changes that can occur from month to month or year to year. While these time variables may not be causal, they likely correlate with environmental factors such as insolation (Whitman et al., 2004). With this correlation in mind, variables such as time of day could provide insight into relationships between environmental predictors of *E. coli.*

The routine updates to the national level precipitation and insolation data repositories allowed inclusion of those variables without decreasing the sample size of our study due to missing values. However, the precipitation and insolation measurements were not taken at Lake Avondale and therefore have greater uncertainty as they are not site-specific. In contrast, the site-specific AAS data collection was interspersed throughout six non-consecutive months and includes more missing values. The size of the dataset was limited by the number of outcome *E. coli* measurements to a sample size of 29. However several environmental variables had even fewer observations, for example there are only five measurements of transparency, a key predictive variable in the literature (Francy, Stelzer, Duris, et al., 2013; McEgan, Mootian, Goodridge, Schaffner, & Danyluk, 2013; Nevers & Whitman, 2005; Olyphant & Whitman, 2004; Whitman et al., 2004). A limitation of combining datasets is use of inconsistent methods of analysis. While only AAS *E. coli* measurements were available for this study, service reports rely on membrane filtration when evaluating *E. coli*.

The small size of the data set limits the statistical analyses that can be performed resulting in limited power to detect significant differences in associations between variables over time. The uncertainty in the models lends itself to over-protective false positive results as well as false negative predictions declaring water out of regulatory compliance to be within acceptable limits (Brady, 2007). . The limited number of observations and irregular data collection also increase the uncertainty, widening the confidence intervals, around resulting estimates. Outcomes classified from a small number of observations are subject to more misclassification (World Health Organization (WHO), 2009). Despite these limitations the adjusted-r-square value suggests a significant moderate association that can be used to predict *E. coli* concentrations in Lake Avondale.

This study is a site-specific examination and what is found here may not hold true for another lake, due to unmeasured environmental factors or variations in key predictor variables such as precipitation. The predictive capabilities of any model is dependent on the quality of the data used to develop it (Francy et al., 2006). Interpretation of results, therefore, should focus on the significant associations which may be suggestive of environmental predictors of *E. coli* at another site and allow for relative predictions of changes in *E. coli* concentrations in Lake Avondale. Ultimately, even given the limitations of the dataset, the results of the study are informative. The results can help direct future work both in terms of lake management and research on small, neighborhood, suburban lakes.

Monitoring is critical to informed management of both local waters and the watershed (United States Geological Survey (USGS), 2002). To identify lake trends and compare conditions throughout the watershed, it is necessary to monitor long-term following consistent measurement techniques (United States Geological Survey (USGS), 2002).

This investigation promotes evidence-based lake management. As a collaboration between students, researchers, professionals in environmental fields, and community members the project explores a possible means bridging gaps between research and community understanding of the research. This project has potential to motivate small-scale surveillance of environmental exposures at the neighborhood level. When data is collected by community members at the local level, the resulting evidence may suggest key parameters to address regarding lake management.

Conclusion

Despite the small sample size, the results are consistent with the literature. While results must be interpreted cautiously in light of the data quality, a few broad conclusions may be drawn. Greater numbers of fecal indicator bacteria were detected in spring and summer months, the warmest months in Georgia. Several environmental factors were identified for focus in future investigations: conductivity, precipitation, dissolved oxygen, sampling location, and season. Based on correlation analyses conductivity, dissolved oxygen, precipitation, and pH have the strongest associations with *E. coli* concentration or presence in Lake Avondale. Predictive models consistently selected conductivity as a predictor with dissolved oxygen also common. These two factors, conductivity and dissolved oxygen, should be used by the LAAB as a proxy for *E. coli* concentrations in the lake in order to inform management decisions. The high individual measurements of *E. coli* concentration out of compliance with EPA recommendations suggests that the LAAB should not reconsider permitting swimming or full-body contact with the lake water.

This investigation was a collaboration between a neighborhood lake advisory board, city management, local citizens, student researchers, and faculty researchers. The Avondale Estates community has a history of involvement with Lake Avondale: they voluntarily serve on the lake advisory board, ask questions of the board, local scientists and students have tested the lake through Georgia's Adopt-A-Stream program as well as school projects. With such an actively engaged community this project begins to bridge the gap between research in environmental science and local communities. The results of this exploratory project may facilitate increased awareness of interrelationships among environmental factors relevant to environmental health and the potential to motivate small-scale surveillance of environmental exposures at the neighborhood level.

Chapter IV: Conclusion and Recommendations

Conclusions

Despite the inability to rule out chance causes, results are suggestive of trends and are consistent with the literature. While results must be interpreted cautiously in light of the data quality, a few broad conclusions may be drawn. Greater numbers of fecal indicator bacteria were detected in spring and summer months, the warmest months in Georgia. The selected models identified several environmental factors that together predict *E. coli* concentrations in Lake Avondale. Those factors were conductivity, precipitation, dissolved oxygen, location, and season. The Lake Avondale Advisory Board can use these findings to inform or support future decision-making and to help address community questions regarding the lake.

This project was a collaborative exploration to address a community concern. By bringing together community questions, city management, and researchers in environmental health the results of this project intend to provide a knowledge base for continued community engagement with the lake. Such engagement includes evidence-based lake management and potential motivation for small-scale surveillance of environmental exposures at the neighborhood level.

Recommendations for Lake Avondale

The LAAB can use the information reported here to inform future decisions whether those decisions involve water quality management, school projects sampling the lake, or responses to community concerns. The selected regression models identified several environmental factors that influence *E. coli* concentrations in Lake Avondale for focus in future investigations and management decisions: conductivity, precipitation, dissolved oxygen, location, and season. Assessing each environmental variable individually, high dissolved oxygen corresponds to lower *E. coli.* For this analysis, dissolved oxygen on the order of 8 mg/L corresponded to near absence of *E. coli*. However, dissolved oxygen is not the only parameter than can influence *E. coli* concentrations, so *E. coli* may still be present at this level of dissolved oxygen. Conductivity appears to vary by sampling site between 60 and 80 μS/cm all corresponding to low levels of *E. coli*. *E. coli* levels also increase as precipitation accumulates.

In light of these associations, recreators may want to exercise greater caution after precipitation events and when dissolved oxygen levels are high. These times may be the best ties to avoid contact with the lake water.

More samples that can be compared for each environmental variable of interest enhances the precision of statistical analyses by reducing uncertainty in results. Monitoring lake quality for trends and future comparisons will help guide evidence-based decision-making and produce more robust results.

Recommendations for Future Studies

To enhance understanding of environmental influences on water quality in Lake Avondale and add versatility to management options, continued collected of quality data is recommended. Future studies should strive for more robust statistical analyses might include a better informed, validated regression model that can be used to predict the quality of lake water. Recognizing that a variety of feasibility issues exist when collecting data, particularly if collected by volunteers, what follows are several ideal suggestions for future data collection.

- Collect samples at the same sites as previous data collection
- Record time of day when collecting samples
- Note recent environmental changes such as rainstorms, presence of Canada geese, or algae during the several days prior to collecting measurements
- Set-up a rain gauge at common sampling sites.
- Collect samples at regular intervals whether every hour, every week, or the same week each year
- Collect samples before, during (optional), and after a rainstorm
- Continue annual Canada goose relocation
- Count the number of Canada geese present at the lake a few days before sampling

- Compile data in a single location
- Maintain compilation of information provided in lake service reports
- For a more comprehensive analysis of lake water quality, chemical in addition to the bacterial and physicochemical analyses

Extended Discussion of Canada Geese

Currently available data on Canada geese at Lake Avondale was not sufficient to generate site-specific recommendations; however recommendations are possible based on review of the available data in the context of existing literature. While counts of geese present at Lake Avondale at any given time were limited or unavailable, previous research suggests that geese do influence water quality (Fallacara et al., 2001; Feare et al., 1999; Gorham & Lee, 2015; Graczyk et al., 1997; Hussong et al., 1979; Lu et al., 2009; Manny et al., 1994; Moriarty et al., 2011). Goose removal typically occurs once per year (O. Griffin, personal communication, January 22, 2016; E. Miller, personal communication, March 12, 2016). However Canada geese are often present at the lake intermittently throughout the year, sometimes in flocks as large as 30 geese though available documentation notes flocks of 5 to 19 geese (Figure 3) (O. Griffin, personal communication, January 22, 2016). Since a single goose can produce 0.5 to 3 pounds of feces per day (French & Parkhurst, 2009; Hussong et al., 1979; Swallow et al., 2010); therefore, a flock of 10-20 geese can produce large quantities of waste, on the order of 5-30lbs. While the feces will be distributed around the park and not all the feces will end up in the lake or have viable pathogens. While there are records of number of geese removed, differing numbers of geese may be present during other months and on other dates. During relocation, relocaters remove all Canada geese present at the lake at the time (O. Griffin, personal communication, April 19, 2016). Ideally, yearly counts of Canada geese the same week that *E. coli* measurements are collected will be recorded going forward. Relocation of geese may be particularly effective when heavy rain is expected to wash goose feces into the lake.

Volunteer Data Collection

The general public increasingly contributes to scientific data collection as citizen science expands. Lottig et al., 2014 conducted a large-scale water clarity study using data collected entirely by citizens (Lottig et al., 2014). Questions and potential limitations of data collected by citizens rather than trained scientists include level of knowledge and experience, lack of standardized protocols, accuracy and precision of data collection, number of samples collected, and data that may not be representative of the study area spatially or temporally (Lewandowski & Specht, 2015; Lottig et al., 2014). However, with limited resources available for widespread environmental monitoring, citizen-collected data can be a critical contribution to datasets (Lottig et al., 2014). Analysis by Lewandowski et al., 2015 of biological studies comparing professional to citizen-volunteer-collected data found that while professional data was more accurate in 4 of 7 cases as well as less variable than volunteer data the professionally-collected and volunteer-collected data were not significantly different (Lewandowski & Specht, 2015). This evidence encourages continued collection and compilation of data by the Avondale Estates community.

Public Health Implications

From a public health standpoint, exploring water quality through the collaborative effort of students, researchers, and communities promotes spread of knowledge while investigating relationships between environmental factors and directing future work. With ample room for variation in environmental samples due to site-specific conditions, expanding understanding of relationships between environmental factors enhances our ability to describe and eventually predict local water quality hazards. This background understanding of the parameters needed to mitigate unwanted exposure to that hazard, allowing for more timely reduction in negative health outcomes should they arise.

Chapter V. Report to the Community

Which environmental variables cause variation in *E. coli* concentrations in Lake Avondale?

INTRODUCTION

The initial goal behind this investigation was to evaluate the ability of Canada goose feces to predict concentrations of the bacterium *Escherichia coli* (*E. coli*) in Lake Avondale. This microorganism is one measure used to evaluate water quality and can indicate the presence of pathogenic bacteria that can cause illness.

The purpose of this analysis was to provide guidance for effective water quality monitoring and lake management. There is a need to understand the factors that influence the water quality of Lake Avondale. Statistical models were used to evaluate whether lake water quality and environmental factors can predict *Escherichia coli (E. coli*) concentrations in Lake Avondale.

Ongoing management of Lake Avondale may include decision-making based on the water quality findings from this investigation. This study may also direct any future monitoring of water quality.

Conductivity, precipitation, and sampling site were found to influence *E. coli* concentrations in Lake Avondale. However, the available data was limited. I was not able to establish whether any of the variables caused a defined increase or decrease in *E. coli* concentrations, only that they were correlated with changes in *E. coli*. Higher conductivity, lower dissolved oxygen, and more precipitation all tend to raise *E. coli* concentrations in the lake.

E. coli and the related fecal coliforms, are practical standards used to assess water quality with the implications for risk of exposure to unfriendly microbes. Recreational water has different standards than drinking water because the estimated amount of water ingested is different.

The U.S. Environmental Protection Agency (EPA) limit for *E. coli* in recreational freshwater is an average of 126 CFU/100mL from five samples collected over 30 days.

RECOMMENDATIONS FOR LAKE AVONDALE

The resulting predictors from the statistical models such as conductivity, dissolved oxygen, and precipitation can be used as proxies for *E. coli* levels in Lake Avondale and to monitor water quality.

While not included in the analysis, a review of the literature notes Canada geese can contribute to poor water quality, suggesting that Avondale Estates' annual relocation of Canada geese is beneficial to the lake.

RECOMMENDATIONS FOR FUTURE WORK

Recognizing that there are feasibility limitations with implementing a robust long-term monitoring program run by a local community on a volunteer basis there are still actions to consider going forward.

- Set-up a rain gauge at common sampling sites.
- Collect samples before, during (optional), and after a rainstorm
- Collect samples at the same sites at others before you
- Collect samples at regular intervals whether every hour, every week, or the same week each year
- Compile data in a single location
- Continue annual Canada goose relocation
- Count the number of Canada geese present at the lake a few days before sampling

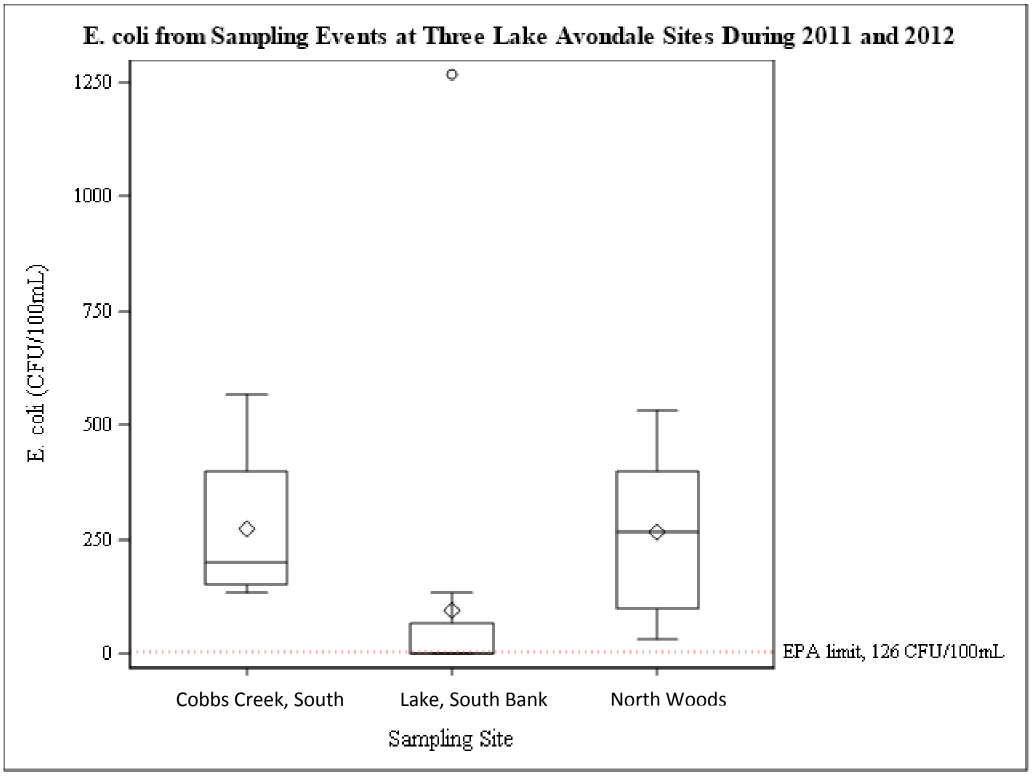
Tables and Figures

Table 1: Descriptive statistics of continuous environmental factors

Descriptive Statistics for Regression Model Parameters

Variable	Units	N	Missing values	Mean	Standard Deviation	Standard Error	Median	Minimum	Maximum	Mode	Skewness	Kurtosis	Variance	Coefficient of Variation
E. coli	CFU/100mL	29	0	155	265	49.3	33.0	0	1267	0	3.01	10.8	70463	171
log-*E. coli*	log-CFU/100mL	29	0	2.17	3.91	0.727	3.50	-3.00	7.14	-3.00	-0.518	-1.61	15.3	181
Air temperature	°C	29	0	20.1	6.58	1.22	19.7	9.75	30.6	13.75	0.082	-1.33	43.3	32.7
Altitude	feet	29	0	977	15.2	2.83	978	945	997	978	-0.979	0.977	233	1.56
Conductivity	micro Siemens/cm	29	0	74.8	11.2	2.08	70	60	100	70	1.02	0.739	126	15.0
Insolation	Watts/square meter	29	0	215	88.1	16.4	236	47.6	316	119	-0.364	-1.26	7757	41.0
Precipitation	inches to hundredths	29	0	50.1	142	26.5	0	0	726	0	4.15	19.2	20296	284
log-precipitation	log-inches	29	0	-0.773	3.47	0.645	-3.00	-3.00	6.59	-3.00	1.06	-0.677	12.1	-449
Water temperature	°C	29	0	20.2	6.69	1.24	19.1	10.1	30.5	13.3	0.231	-1.45	44.7	33.0
Dissolved oxygen	mg/L (ppm)	28	1	7.52	1.44	0.271	7.38	5	9.8	7.3	-0.169	-1.16	2.06	19.1
pH	pH units	28	1	6.91	0.531	0.100	6.75	6.25	8	6.75	1.04	0.066	0.282	7.69
Fecal coliform	CFU/100mL	14	15	734	1462	391	85	10	4700	10	2.32	4.41	2137132	199
log-fecal coliform	log-CFU/100mL	14	15	4.78	2.06	0.551	4.40	2.30	8.46	2.30	0.472	-0.758	4.25	43.2
Rainfall	inches/hour	7	22	0.033	0.029	0.011	0.042	0	0.083	0.042	0.566	0.377	0.001	88.9
log-rainfall	log-inches/hour	7	22	-3.13	0.410	0.155	-3.18	-3.87	-2.48	-3.18	-0.496	2.50	0.168	-13.1
Transparency	cm	5	24	88.5	11.9	5.34	90	75	100	100	-0.183	-2.90	142.5	13.5
Year	-	-	0	2011	0.412	0.077	2011	2011	2012	2011	1.53	0.352	0.170	0.020
Month	-	-	0	6.45	3.17	0.588	6	2	11	5	0.063	-1.14	10.0	49.1

Figure 1: Comparison of *E. coli* concentration by sampling site



Cobbs Creek, South is the creek downstream of the lake. Lake, South Bank is at the southern bank of the lake, and North Woods is the creek upstream of the lake.
The dotted line at126 CFU/100mL indicates the EPA monthly regulation for recreational water based on a geometric mean of 126 CFU/100m (five samples taken over 30 days).

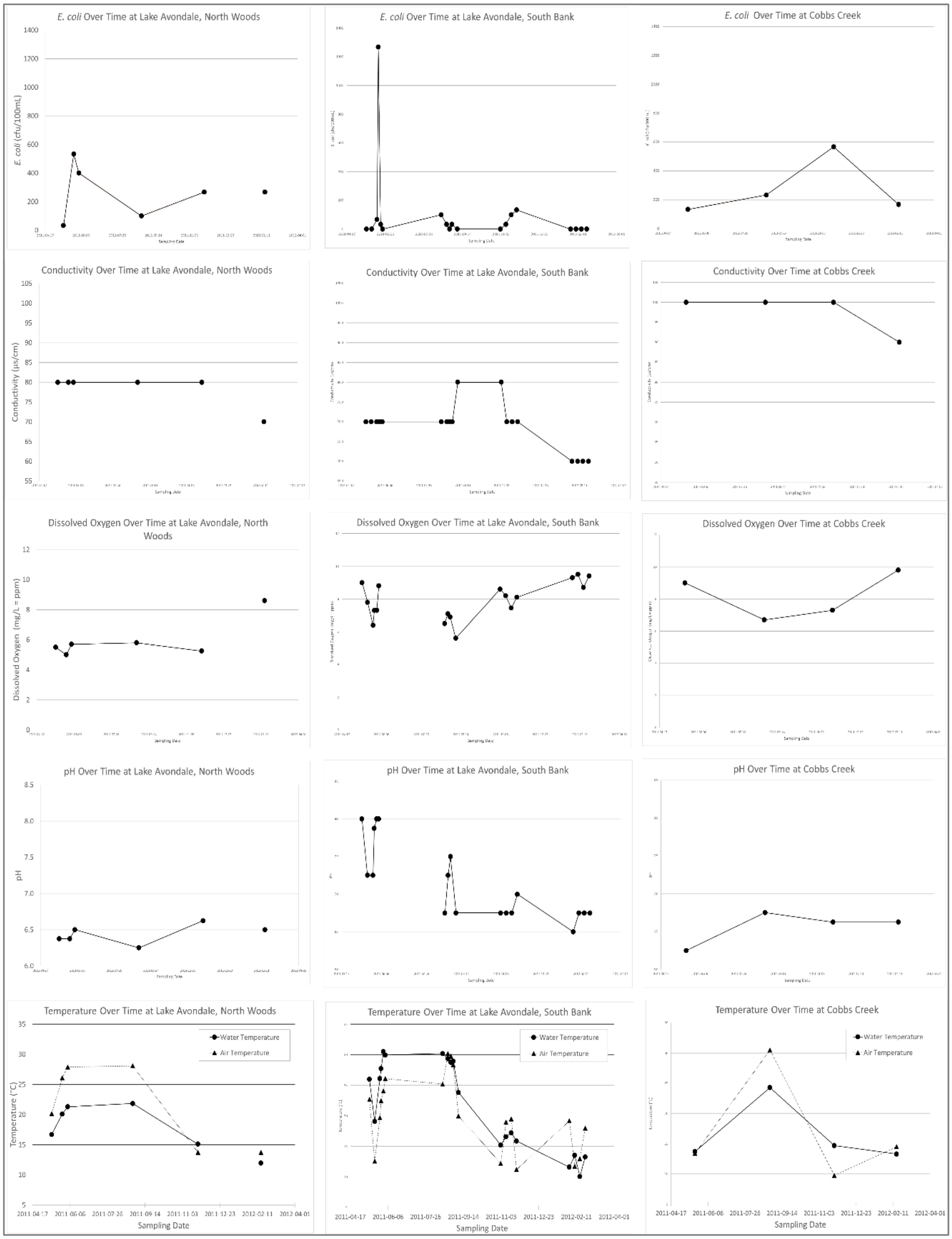
Table 2: Yearly compliance with EPA monthly *E. coli* regulation

Year	Total Samples	Incompliant Samples	Percent Incompliant by Year
2011	24	12	50%
2012	6	2	33%
2013	0	-	-
2014	0	-	-
2015	14	0	0%
Total:	44	14	

Table 3: Monthly compliance with EPA monthly *E. coli* regulation for recreational

Month	Total Samples	Incompliant Samples	Incompliant per Month (%)
January	0	0	0%
February	6	2	33%
March	0	-	-
April	0	-	-
May	8	4	50%
June	2	1	50%
July	0	-	-
August	6	3	50%
September	7	1	14%
October	4	0	0%
November	11	3	27%
December	0	-	-
Total:	44	14	

Figure 2: Comparison of environmental variables across the three sampling sites



The North Woods site is upstream of Lake Avondale and Cobbs Creek is downstream of the lake. Dates correspond to only those when *E. coli* samples were collected.

Table 4: Results of simple linear regression analysis of environmental variables with log-transformed E. coli to assess bivariate association.

Results of Simple Linear Regression with log-*E. coli*

Independent (Predictor) Variable	F Value	p-value	R-square	Adjusted R-square
log- fecal coliform	18.92	**0.002**	**0.70**	0.67
Dissolved oxygen	18.05	**0.003**	**0.69**	0.65
Air temperature	2.88	0.128	0.26	0.17
Conductivity	0.19	0.676	0.02	-0.10
Insolation	0.12	0.742	0.01	-0.11
pH	0.14	0.719	0.02	-0.11
Water temperature	1.87	0.208	0.19	0.09
Week day	0.39	0.548	0.05	-0.07
Month	1.87	0.209	0.19	0.09
Season	1.32	0.283	0.14	0.03
Year	1.59	0.242	0.17	0.06
log-Precipitation	4.3	0.062	0.28	0.22
Precipitation	1.86	0.200	0.14	0.07

Bold variables indicate a statistically significant relationship at the 95% confidence level

Table 5: Comparison of multivariate linear models for predicting *E. coli* concentrations in Lake Avondale based on environmental factors.

Comparison of Multivariate Linear Regression Models

Model	N	DF	RMSE	F-value	p-value	R-Square	Adjusted R-Square
Full Model	28	8	10.2	2.82	0.030	0.54	0.35
Reduced Model	29	3	9.86	6.15	0.003	0.42	0.36
Reduced Model with potential outliers removed	26	3	5.69	15.8	<0.0001	0.68	0.64
Reduced Model, with interaction terms	28	5	5.66	0.002	8.41	0.56	0.46

DF = degrees of freedeom

RMSE = root mean square error

Table 6: Correlation matrix comparing environmental parameters using correlation coefficients and the associated p-values.

Spearman Correlation Coefficients,
Probability > |r| under H0: Rho=0

	Air Temperature	**Altitude**	**Conductivity**	**DO**	**Water Temperature**	**pH**	**Month**	**Year**	**Precipitation**	**log-Precipitation**	**Insolation**
Air Temperature		0.20 0.296 n = 29	0.09 0.654 n = 29	**-0.49 0.008 n = 28**	**0.75 <.0001 n = 29**	0.16 0.415 n = 28	0.09 0.635 n = 29	-0.39 0.036 n = 29	-0.44 0.016 n = 29	-0.44 0.016 n = 29	0.44 0.016 n = 29
Altitude	0.21 0.285 n = 29		-0.11 0.557 n = 29	**-0.50 0.007 n = 28**	0.04 0.855 n = 29	-0.28 0.144 n = 28	-0.03 0.877 n = 29	-0.06 0.755 n = 29	-0.07 0.706 n = 29	-0.07 0.706 n = 29	0.06 0.748 n = 29
Conductivity	-0.02 0.899 n = 29	**-0.54 0.002 n = 29**		-0.43 0.022 n = 28	0.04 0.850 n = 29	-0.47 0.012 n = 28	0.39 0.037 n = 29	-0.46 0.011 n = 29	0.12 0.519 n = 29	0.12 0.519 n = 29	0.24 0.212 n = 29
DO	-0.47 0.012 n = 28	-0.44 0.019 n = 28	-0.28 0.156 n = 28		**-0.49 0.009 n = 28**	0.17 0.394 n = 28	-0.46 0.013 n = 28	**0.64 0.000 n = 28**	-0.04 0.827 n = 28	-0.04 0.827 n = 28	-0.15 0.436 n = 28
Water Temperature	**0.78 <.0001 n = 29**	0.08 0.692 n = 29	-0.06 0.747 n = 29	-0.40 0.036 n = 28		**0.59 0.001 n = 28**	0.33 0.084 n = 29	**-0.70 <.0001 n = 29**	-0.31 0.104 n = 29	-0.31 0.104 n = 29	**0.50 0.006 n = 29**
pH	0.25 0.198 n = 28	-0.04 0.856 n = 28	-0.35 0.066 n = 28	0.17 0.379 n = 28	**0.69 <.0001 28**		0.09 0.653 n = 28	-0.23 0.238 n = 28	-0.02 0.908 n = 28	-0.02 0.908 n = 28	0.14 0.466 n = 28
Month	0.08 0.669 n = 29	-0.03 0.882 n = 29	0.33 0.082 n = 29	-0.43 0.021 n = 28	0.23 0.240 n = 29	-0.02 0.900 n = 28		**-0.72 <.0001 n = 29**	0.22 0.242 n = 29	0.22 0.242 n = 29	-0.21 0.285 n = 29
Year	-0.40 0.031 n = 29	-0.06 0.764 n = 29	-0.38 0.043 n = 29	**0.63 0.0003 n = 28**	**-0.62 0.000 n = 29**	-0.27 0.173 n = 28	**-0.73 <.0001 n = 29**		0.09 0.631 n = 29	0.09 0.631 n = 29	-0.40 0.033 n = 29
Precipitation	-0.11 0.560 n = 29	-0.03 0.888 n = 29	0.00 0.994 n = 29	-0.08 0.673 n = 28	0.08 0.681 n = 29	0.28 0.151 n = 28	0.15 0.442 n = 29	-0.16 0.410 n = 29		**1.00 <.0001 n = 29**	-0.52 0.004 n = 29
log-Precipitaion	-0.46 0.011 n = 29	-0.10 0.598 n = 29	0.12 0.548 n = 29	-0.03 0.882 n = 28	-0.30 0.114 n = 29	-0.03 0.868 n = 28	0.24 0.208 n = 29	0.08 0.667 n = 29	**0.69 <.0001 n = 29**		**-0.52 0.004 n = 29**
Insolation	**0.50 0.006 n = 29**	0.04 0.831 n = 29	0.21 0.279 n = 29	-0.22 0.254 n = 28	**0.57 0.001 n = 29**	0.32 0.098 n = 28	-0.21 0.282 n = 29	-0.40 0.032 n = 29	-0.11 0.555 n = 29	**-0.50 0.006 n = 29**	

Pearson r Correlation Coefficients
Probability > |r| under H0: Rho=0

Pearson correlations are given on the bottom left of the table and Spearman correlations are on the top right section. Two variables, transparency and log-Rain had no significant correlations and were removed from the table for ease of viewing. Fecal coliform and log-fecal coliform are not included as we are more interested in the subset, *E. coli*. Altitude was also removed as it is indicative of sampling site.

Table 7: Parameter estimates for the full eight--variable predictive linear regression model of log-transformed *E. coli* in Lake Avondale

Multivariate Linear Regression - Full Model						
Variable	Parameter Estimate	Standard Error	t-value	p-value	95% Confidence Limits	
Intercept	-10.8	17.9	-0.6	0.555	-48.3	26.7
Conductivity	0.222	0.110	2.03	0.057	-0.007	0.452
Dissolved oxygen	-0.350	0.810	-0.43	0.671	-2.04	1.35
pH	0.123	1.94	0.06	0.950	-3.94	4.19
log-precipitation	0.491	0.249	1.97	0.064	-0.031	1.01
Air temperature	0.171	0.140	1.22	0.236	-0.122	0.464
Insolation	-0.005	0.013	-0.38	0.708	-0.032	0.022
Site	-0.549	0.606	-0.91	0.376	-1.82	0.719
Season	-0.789	0.792	-1	0.331	-2.45	0.868

Table 8: The parameter estimates for the reduced model based on backwards selection from the eight-variable predictive model of log-transformed *E. coli*

Multivariate Linear Regression - Reduced Model						
Predictor Variable	Parameter Estimate	Standard Error	t-value	p-value	95% Confidence Limits	
Intercept	-10.3	4.05	-2.54	0.018	-18.6	-1.94
Conductivity	0.206	0.057	3.60	0.001	0.088	0.324
Precipitation	0.332	0.172	1.93	0.066	-0.023	0.687
Site	-0.719	0.360	-2.00	0.057	-1.459	0.022

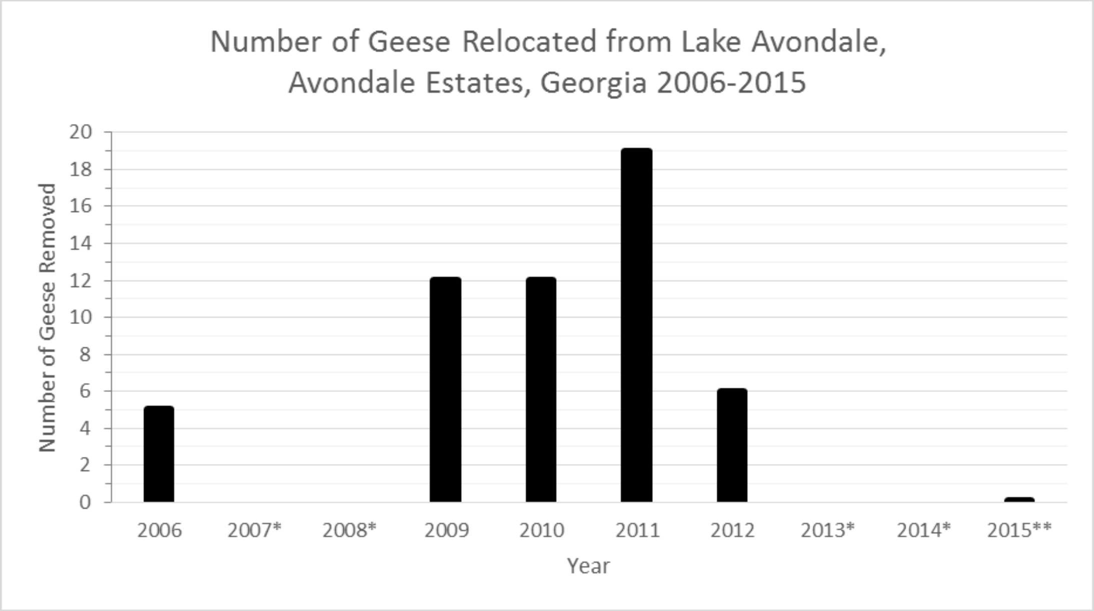
Table 9: The parameter estimates for the reduced model with potential outliers identified by jackknife residuals, removed

Reduced Model, Potential Outliers Removed						
Predictor Variable	Parameter Estimate	Standard Error	t-value	p-value	95% Confidence	
Intercept	-11.2	3.39	-3.30	0.003	-18.2	-4.19
Conductivity	0.226	0.048	4.70	<0.0001	0.127	0.326
Precipitation	0.369	0.147	2.50	0.020	0.063	0.674
Site	0.739	0.300	-2.46	0.022	-1.36	-0.118

Table 10: Parameter estimates for the reduced nine-variable predictive linear regression model including investigation of interaction terms.

Multivariate Linear Regression - Reduced Model, Interaction Terms						
Variable	Parameter Estimate	Standard Error	t-value	p-value	95% Confidence Limits	
Intercept	17.9	10.7	1.67	0.109	-4.34	40.1
Dissolved oxygen	0.809	0.440	1.84	0.080	-0.104	1.72
Season	-7.31	3.62	-2.02	0.056	-14.8	0.194
Season*Dissolved oxygen	0.156	0.053	2.95	0.007	0.046	0.265
Conductivity	-3.11	1.16	-2.69	0.014	-5.52	-0.710
log-Precipitation	0.508	0.176	2.89	0.009	0.143	0.873

Figure 3: Numbers of geese relocated annually from Lake Avondale, Avondale Estates, Georgia



* We found no documentation of goose removal and relocation during these years but this does not reflect presence or absence of geese at the lake during these years.

** Geese relocation was attempted however not performed due to inconsistent presence of geese

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Appendix

Appendix A: Acronyms

ANOVA	Analysis of variance
CDC	Centers for Disease Control and Prevention
CFU	Colony-forming units
CSTE	Council of State and Territorial Epidemiologists
EC	*Escherichia coli (E. coli)*
EPA	United States Environmental Protection Agency
FC	Fecal coliforms
IPM	Integrated Pond Management
LAAB	Lake Avondale Advisory Board
MPN	Most probable number
NEO	National Aeronautics and Space Administration (NASA) Earth Observatory
NOAA	National Oceanic and Atmospheric Administration
NORS	National Outbreak Reporting System
VIF	Variance inflation
WBDOSS	Waterborne Disease and Outbreak Surveillance System
WHO	World Health Organization

Appendix B: Definitions of Terms

Benthic – The bottom of a body of water (e.g. the sediment surface)

Chlorophyll - A pigment that converts light energy into chemical energy during photosynthesis and gives organisms such as algae their green color ("chlorophyll,")

E. coli *Escherichia coli*, a type of coliform bacteria

Enteric – of, relating to, or affecting the intestines ("Enteric,")

Eutrophic lake – lake characterized by low levels of dissolved oxygen and high levels of dissolved nutrients that promote growth of aquatic plant life ("Eutrophication,")

Eutrophic - well-nourished (Connolly, 1990)

Indicator organism - an organism used as a proxy measure for the presence of other organisms; in this case *E. coli* is a proxy for fecal contamination and therefore for enteric pathogens (K. Levy et al., 2012)

Insolation – solar radiation, or more specifically the rate of direct solar radiation received per unit of horizontal surface ("Insolation,")

Kjeldahl nitrogen - Total Kjeldahl Nitrogen, TKN, is the sum of ammonia-nitrogen and organically-bound nitrogen; this sum does not include nitrate-nitrogen or nitrite-nitrogen, distinguishing TKN from total nitrogen (ASA Analytics).

Lake – the definition can vary, but it is generally considered larger and deeper than a pond, sometimes designated as larger than 10 acres (McComas, 2003)

Oligotrophic – poorly nourished (Connolly, 1990)

Pond – the definition can vary but it is generally considered smaller and shallower than a lake, sometimes said to be 10 acres or less (McComas, 2003)

Appendix C: Regulations on Fecal Coliforms and *E. coli*

Table 1: Fecal coliform water quality standards for freshwater

Fecal Coliforms in Georgia			
Standard	**Maximum Concentration**	**Number of Incompliant Samples**	**Data Source**
Freshwater, Recreational	200 CFU / 100 mL	5/14 (36%)	(Georgia Adopt-A-Stream, n.d.)
Fishing, May through October	200 CFU / 100 mL	5/14 (36%)	(Georgia Adopt-A-Stream, n.d.)
Fishing, November through April	1000 CFU / 100 mL	2/14 (14%)	(Georgia Adopt-A-Stream, n.d.)

Table 2: *E. coli* water quality standards for freshwater

E. coli			
Standard	**Maximum Concentration**	**Number of incompliant samples**	**Data Source**
Designated swimming	235 CFU / 100 mL	6/29 (21%)	(Georgia Adopt-A-Stream, n.d.)
Recreational freshwater Geometric mean (5 samples taken over 30 days)	126 CFU / 100 mL	10/29 (34%)	(EPA, 2003; EPA, 1986; Yoder et al., 2004)

Incompliance with EPA Single Sample *E. coli* Limit for Recreational Water (235 CFU/100mL)

Month	**n**	**Number Incompliant**	**Percent Incompliant**
January	0	0	0%
February	6	1	17%
March	0	-	-
April	0	-	-
May	8	2	25%
June	2	1	50%
July	0	-	0%
August	6	0	0%
September	7	0	0%
October	4	0	0%
November	11	2	18%
December	0	-	-
Total:	44	6	14%

EPA = U.S. Environmental Protection Agency
n = number of samples collected
Sampling dates: May 2011 - February 2012

Georgia Adopt-A-Stream. (n.d.). Bacteria and Water Quality. Chapter 1. pp. 17-22. Bacterial Monitoring. Available at http://www.georgiaadoptastream.com/Manuals_etc/Bacterial/B_Ch_1.pdf?5

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Appendix D: Estimated Water Ingestion During Water Recreation Activities

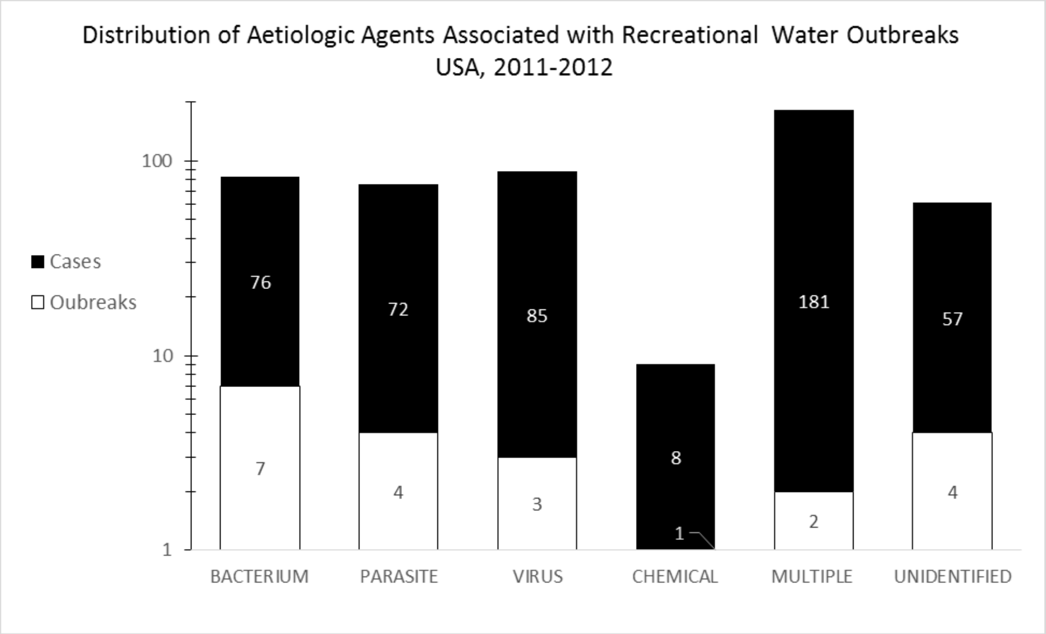
Estimated Water Ingestion During Water Recreation Activities (mL/hr)								
Activity	**Surface Water Studies**				**Swimming Pool Study**			
	N	**Median**	**Mean**	**UCL**	**N**	**Median**	**Mean**	**UCL**
Fishing	600	2	3.6	10.8	121	2.0	3.5	10.6
Wading/Splashing	-	-	-	-	112	2.2	3.7	1.0
Walking	-	-	-	-	23	2.0	3.5	1.0
Canoeing*	766	2.3	3.9	10.8	76	2.6	4.4	14.1

* includes with and without capsizing

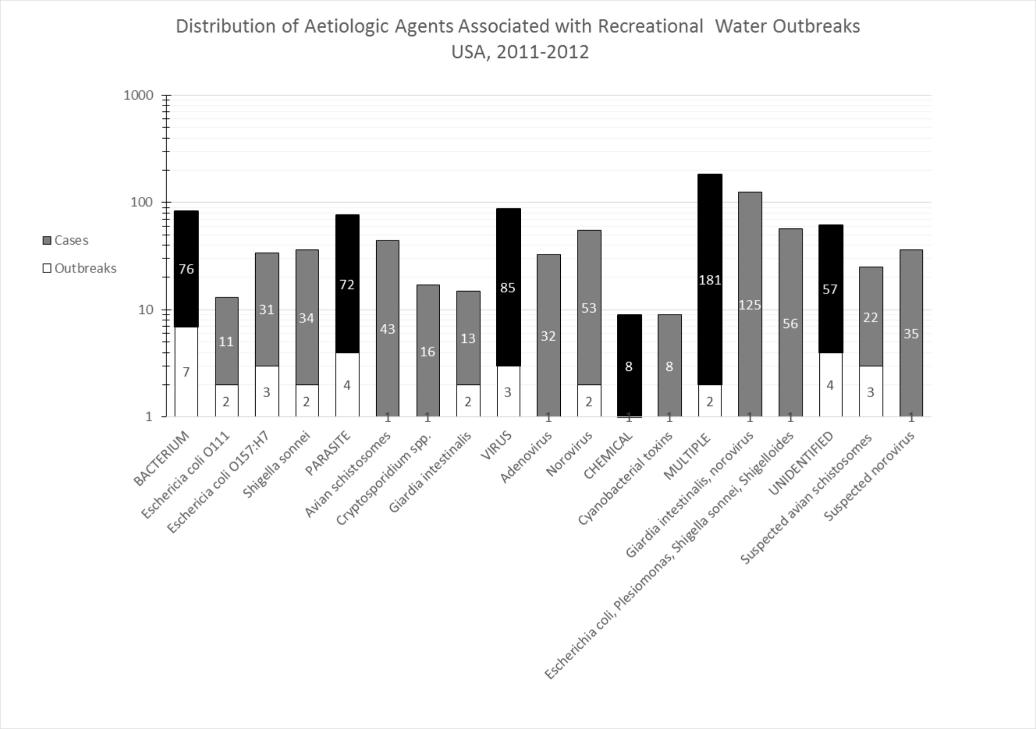
N = Number of participants
UCL = Upper confidence limit (i.e. mean +1.96 x standard deviation)
\- = No data

Source: modified from EPA (2011) and Dorevitch et al. (2011)

Appendix E: Proportions of Microbial Etiologic Agents Associated with Recreational Water Outbreaks



(Hlavsa 2015)



(Hlavsa 2015)

Hlavsa, M. C., Roberts, V. A., Kahler, A. M., Hilborn, E. D., Mecher, T. R., Beach, M. J., . . . Yoder, J. S. (2015). Outbreaks of Illness Associated with Recreational Water--United States, 2011-2012. *MMWR: Morbidity and Mortality Weekly Report, 64*(24), 668-672.

Appendix F: Key Water Quality Concerns for Selected Lake Uses

There are multiple options in terms of what to consider when managing lake water quality. Focus areas can be determined based on the primary function of the lake as well as actionable pathways, which elements of water quality it is most feasible to control.

Recommended Water Quality Parameters to Consider for Management Purposes Based on Primary Function of the Lake

Water Quality Parameter	Primary Function of the Lake		
	Fishing	Irrigation	Aesthetic Beauty
Aquatic herbicides	✓	✓	✓
Pesticides	✓	✓	
pH	✓	✓	
Copper	✓		✓
Dissolved oxygen	✓		✓
Turbidity / Clarity	✓		✓
Ammonia-nitrogen	✓		
Summer maximum water temperature	✓		
Iron		✓	
Manganese		✓	
Blue-green algae			✓
Parasites			✓
Nitrate-nitrogen			✓
Phosphorus			✓

(Swistock et al., 2006)

Swistock, B. R., Sharpe, W. E., & McCarty, T. (2006). Pond facts #5: water quality concerns for ponds. In P. S. C. Extension (Ed.).

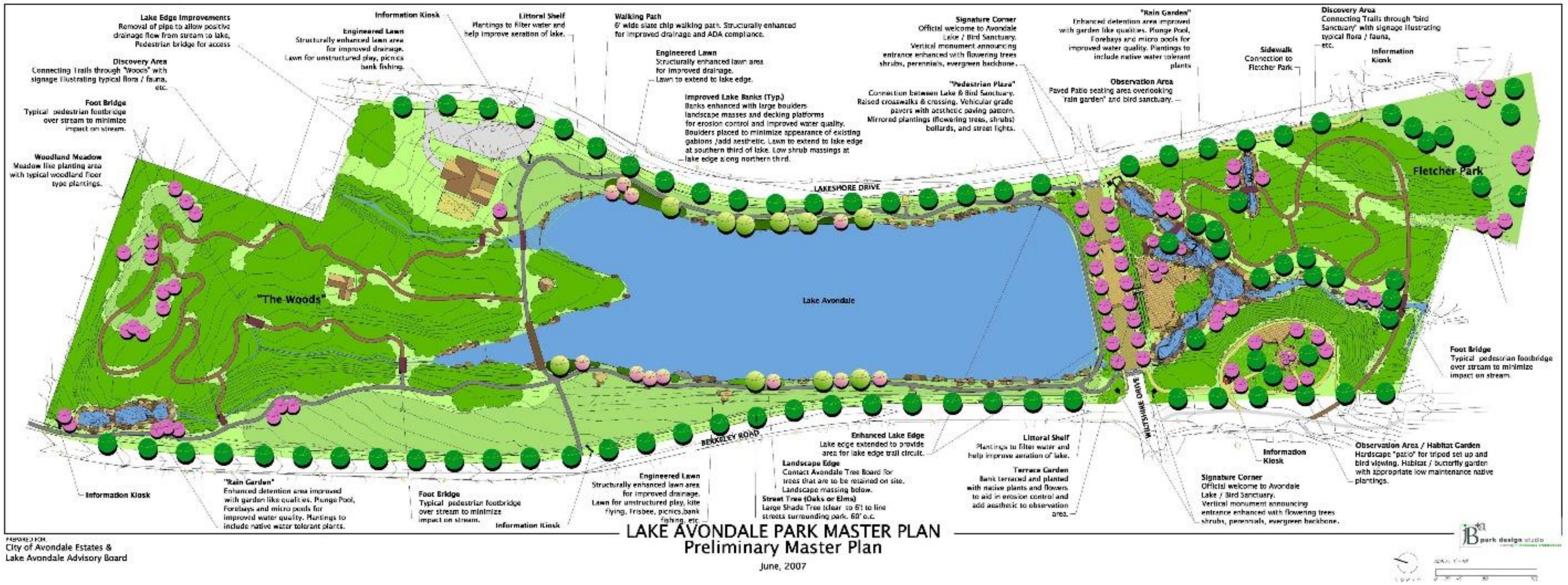
Appendix G: Data Sources

Data Sources		
Variables	**Units**	**Data Source(s)**
Air Temperature	°C	Georgia AAS
Altitude	Feet	Georgia AAS
Conductivity	µS/cm	Georgia AAS
Date of sampling	yyyymmdd	Georgia AAS, Lake Service Report
Dissolved oxygen (DO)	mg/L (ppm)	Georgia AAS
E. coli	CFU/100mL	Georgia AAS
Fecal coliforms	CFU/100mL	Georgia AAS
Canada Geese	count	USDA APHIS WS, Lake Service Report
Insolation (solar radiation)	W/m^2	NEO FLASHFlux TISA data
pH	pH units	Georgia AAS
Precipitation	Inches	NOAA CDO
Rainfall	Inches/hour	Georgia AAS
Transparency	cm	Georgia AAS
Water Temperature	°C	Georgia AAS

AAS: Adopt-A-Stream
APHIS: Animal and Plant Health Inspection Service
CDO: Climate Data Online
CFU: Colony-forming units
FLASHFlux: Fast Long wave And Shortwave Radiative Fluxes
NEO: National Aeronautics and Space Administration (NASA) Earth Observations
NOAA: National Oceanic and Atmospheric Administration

Links to data online:
Georgia AAS: http://www.georgiaadoptastream.org/db/index.html
NEO FLASHFlux TISA: http://neo.sci.gsfc.nasa.gov/view.php?datasetId=CERES_NETFLUX_M
NOAA CDO: https://www.ncdc.noaa.gov/cdo-web/

Appendix H: Map of Plan for Bess Walker Park



(jB+a park design studio, 2007)
http://avondaleestates.org/DocumentCenter/Home/View/100

Map of Lake Avondale and a development plan for Bess Walker Park. The “North Woods” are at the northern end of the lake on the right side of the figure. Outflow from the lake into Cobbs Creek is seen on the right. While not pictured here, private residences face the park from the far-side of the two roads, Lakeshore Drive and Berkeley Road, located east and west the park.

Appendix I: Locations of Sampling Sites

Image credit: Google Earth 7.1.5.1557 (Google Inc.)

Site	**Latitude**	**Longitude**
North Woods (upstream)	33.7689	-84.2655
Cobbs Creek, North	33.7681	-84.265
Lake, North	33.768	-84.2648
Lake Below Dam 2	33.7657	-84.2636
Lake Below Dam 1	33.7656	-84.2638
Cobbs Creek, South	33.7644	-84.2629
Cobbs Creek, Memorial Drive	33.7617	-84.2614

Table 1: Coordinates of the sampling sites by community members registered with Georgia Adopt-A-Stream to collect samples at Lake Avondale

Georgia Adopt-A-Stream:
http://www.georgiaadoptastream.com/db/index.html

Lake Avondale service report sampling sites relative to Google Earth image of the lake.

Image credit: Google Earth 7.1.5.1557 (Google Inc.), United States Geological Survey

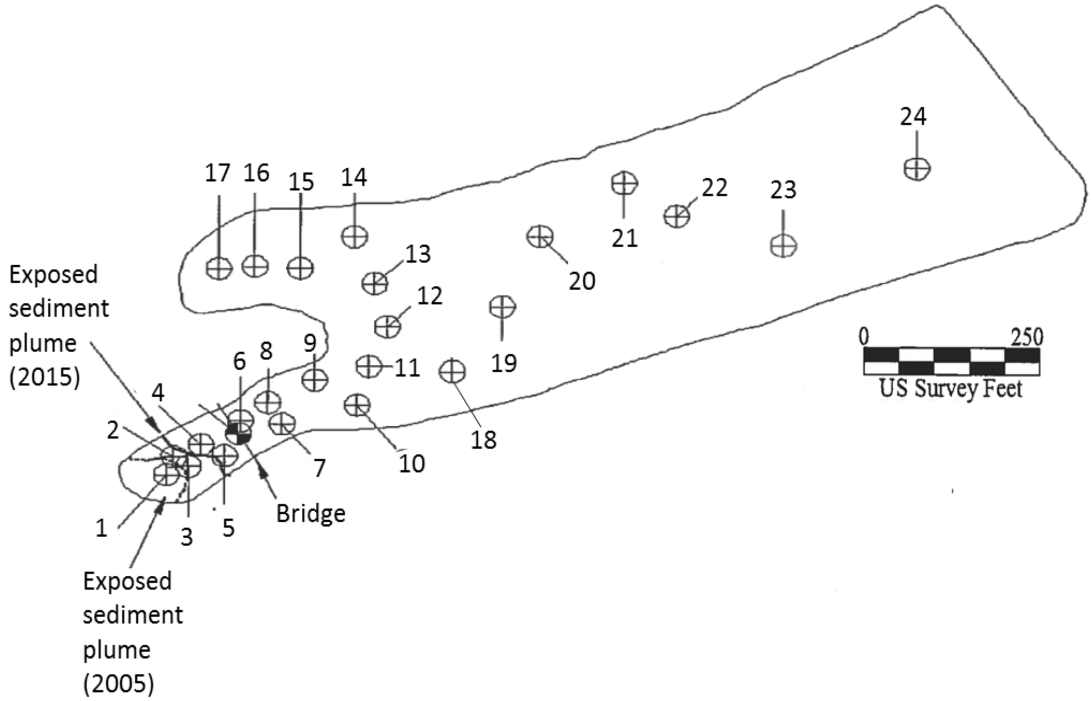


Figure credit: (Brown & Slovisky, 2015a)

Appendix J: Extended Site Description

Frequency of Service Visits by Month (N=11)		
Month	**Frequency***	**Percent**
January	0	0%
February	1	9%
March	1	9%
April	1	9%
May	2	18%
June	1	9%
July	1	9%
August	1	9%
September	1	9%
October	1	9%
November	1	9%
December	0	0%

*N = Total number of visits

Water Clarity (inches) (N=11)		
Clarity	**Frequency**	**Percent**
18	2	18
24	1	9
30	1	9
36	7	64

*N = Total number of service visits

Fish & Wildlife Observations (N=11)		
Wildlife	**Frequency**	**Percent**
Geese - current activity	5	45
Normal	6	55

Identified Aquatic Vegetation (N=11)		
Vegetation	**Frequency**	**Percent**
Filamentous algae*	2	18
Filamentous algae - minor**	1	9
Not applicable	8	73

* Algaecide application performed on these date

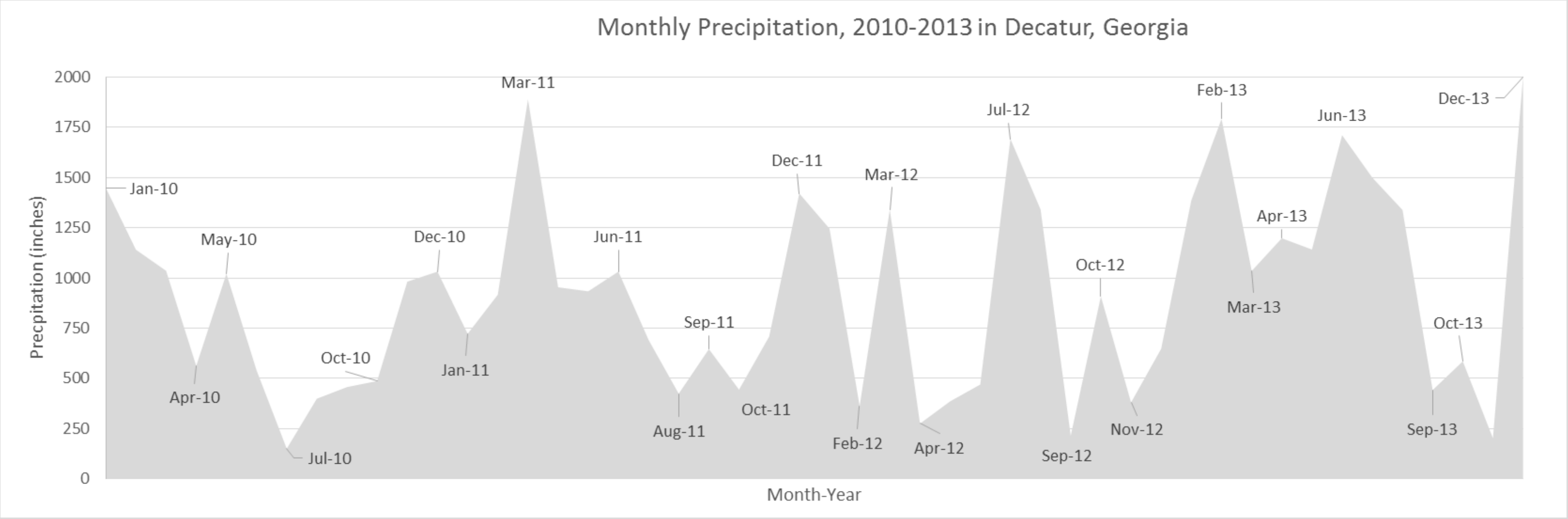
** Algaecide not applied due to windy condition

Data from lake service reports show that technicians visited the lake nine times in 2013 and twice in 2014. The fewest service visits occurred during the winter (December, January, February). At the time of the service visits, the average temperature of the lake ranged from 50 to 83 degrees Fahrenheit but was most frequently between 72 and 77 degrees Fahrenheit. On 7 of 11 occasions (64%) water clarity was 36 inches, but ranged from 18 to 36 inches. Water clarity was measured as depth to which a Secchi disk was visible. On approximately half of the visits (5/11) geese were noted as present. Filamentous algae was identified on three of the eleven dates, all in the spring (March, April, May). The two spillways from the lake were usually clear, clearing blockage or removing debris was occurred during two of the eleven service visits, one in June 2013 and one in May 2014.

Lake service reports also included site observations and maintenance work. Fish observations included signs of stress such as diseased fish or fish swimming near the water surface and gasping. Wildlife observations included signs of beaver, muskrat, and geese; the former two damage large trees and build dams while the latter can increase lake fecal coliform counts and destroy landscapes. Aesthetic appearance was judged in the context of lake color, algae levels and weed coverage. Maintenance included control of observed algae and exotic, invasive weeds, using EPA-approved algaecides and herbicides. (J. Brown, personal

Appendix K: Monthly Precipitation Data

Figure 1: Sum of precipitation by month over four years at the site used as a proxy for precipitation at Lake Avondale.



Appendix L: Photos from Lake Avondale

Figure A

Figure B

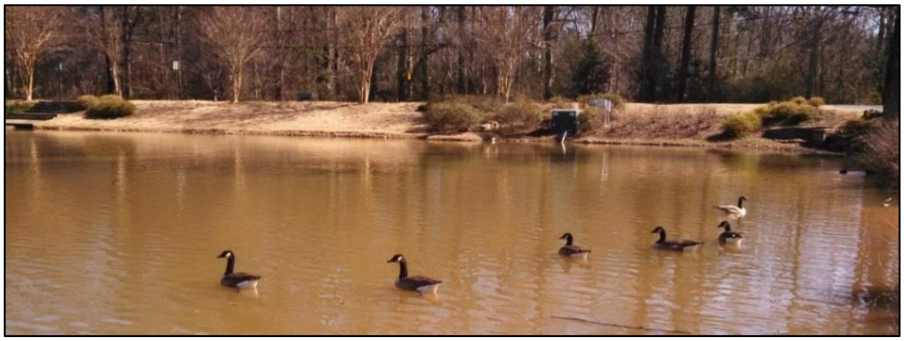


Figure C

Figure A: Geese moving from the south bank of Lake Avondale into the water

Figure B: Six Canada geese swimming in Lake Avondale

Figures C: One of several signs politely telling people not to feed geese.

Photos credit of Lauren Cunningham
February 28, 2016

Appendix M: Relative Location of Lake Avondale

The relative location to Lake Avondale in Dekalb County, Georgia and to waterbodies downstream in the Upper Ocmulgee Watershed.